DISSOLVED ORGANIC MATTER IN WETLAND SOILS AND STREAMS OF SOUTHEAST ALASKA: SOURCES, CONCENTRATION AND CHEMICAL QUALITY

A

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ABSTRACT

Dissolved organic matter (DOM) transported from terrestrial to aquatic ecosystems is an important source of C, N and energy for the metabolism of aquatic heterotrophic bacteria. I examined the concentration and chemical quality of DOM exported from coastal temperate watersheds in southeast Alaska to determine if wetland soils are an important source of biodegradable dissolved organic carbon (BDOC) to aquatic ecosystems. I addressed this question through a combination of high resolution temporal and spatial field measurements in three watersheds near Juneau, Alaska by using a replicated experimental design that characterized DOM export from three different soil types (bog, forested wetland and upland forest) within each of the watersheds. PARAFAC modeling of fluorescence excitation-emission spectroscopy and BDOC incubations were used to evaluate the chemical quality and lability of DOM. Overall, my findings show that wetland soils contribute substantial biodegradable DOM to streams and the response in BDOC delivery to streams changes seasonally, with soil type, and during episodic events such as stormflows. In particular, the chemical quality of DOM in streamwater and soil solution was similar during the spring runoff and fall wet season, as demonstrated by the similar contribution of protein-like fluorescence in soil solution and in streams. These findings indicate a tight coupling between wetland DOM source pools and streams is responsible for the export of BDOC from terrestrial ecosystems. Thus, seasonal changes in soil-stream linkages can have a major influence on watershed biogeochemistry with important implications for stream metabolism and the delivery of labile DOM to coastal ecosystems. Soil DOM additions in small streams draining the three soil types showed that DOM leached from watershed soils is readily used as a substrate by stream heterotrophs and at the same time modified in composition by the selective degradation of the proteinaceous fraction of DOM. These findings indicate terrestrial DOM inputs to streams are an important source of C to support stream heterotrophic production. Thus, the production of protein-rich, labile DOM and subsequent loss in stream runoff has the potential to be an important loss of C and N from coastal temperate watersheds.

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CHAPTER ONE

Introduction

Dissolved organic matter (DOM), which includes organic forms of carbon (DOC), nitrogen (DON) and phosphorus (DOP), is a complex mixture of soluble organic compounds derived from both terrestrial and aquatic sources. Different types of DOM influence aquatic chemistry and biology differently because they vary in chemical properties and biological availability to heterotrophs (McKnight et al. 1985). The qualities of DOM differ in relation to the original source-material, which falls broadly into allochthonous (terrestrially-derived) and autochthonous (derived from within the aquatic system) source pools. Autochthonous sources include algal cell death and senescence, grazing or "sloppy feeding" and extracellular release (Bertilsson and Jones 2003). Allochthonous sources include throughfall, root exudates, plant, root and soil organic matter (SOM) degradation, extracellular release, and the primary and secondary metabolites of microorganisms (Kalbitz et al. 2000).

Streamwater DOM is primarily derived from terrestrial sources, and the processes controlling DOM concentrations are largely a function of watershed characteristics such as soil type, water flowpaths through the soil and wetland coverage (summarized by Mulholland 2003). Wetlands are a particularly important source of DOM to aquatic ecosystems, and runoff from wetland soils can have a profound impact on the chemistry and biology of surface waters (Gorham et al. 1998). At the watershed-scale, wetland influence on DOM concentrations in surface waters has been linked to percentage peat cover (Dillon and Molot 1997; Mattson et al. 2005), percentage wetland cover (Eckhardt and Moore 1990) and wetland type (Xenopoulos et al. 2003). Within wetlands, controls on DOM production and export at the plot scale include temperature (Freeman et al. 2001), soil water table level (Fraser et al. 2001; Blodau et al. 2004) and discharge (Schiff et al. 1997; Pastor et al. 2003). As a result, the delivery of DOM from wetland soils to streams is an intricate process controlled by the interaction between production

consumption, and the degree of hydrologic connectivity with streams. Despite the recognition that wetlands are a substantial source of DOM to surface waters, the chemical quality and biodegradability of DOM from different wetland types and how it varies seasonally is not well understood.

DOM export during storms

The transport of DOC during storms is a well studied area because of its overall importance in the annual watershed DOC export budget (Hinton et al. 1997; Jones et al. 1998). For example, Hinton et al. 1997 documented in a central Ontario headwater stream that a single storm during the fall accounted for 31% of the autumn DOC flux. DOC typically enters streams via two main flowpaths: 1) groundwater, which typically delivers recalcitrant forms of DOC and, 2) shallow soil flowpaths, which deliver organic matter recently leached from organic horizons and shallow, organic-rich mineral soil horizons (Schiff et al. 1997). The relative contribution of both flowpaths to DOC fluxes varies seasonally due to changes in temperature, precipitation, soil water table levels and DOC production/consumption relationships (Schiff et al. 1997). Thus, the increase in streamwater DOC associated with high flows is typically coupled with a change in the chemical quality (Hood et al. 2006; Vidon et al. 2008) and biodegradability of DOC (Buffam et al. 2001).

Biodegradable DOM

Streamwater DOM in temperate regions is a complex mixture of mostly terrestrially-derived, organic compounds that vary in their biological availability. Because DOM comprises most of the reduced available carbon for the metabolism of aquatic and soil heterotrophs, scientists have developed a variety of techniques to study the biodegradability of DOC (BDOC) in natural environments (see Marschner and Kalbitz 2003). Researchers typically use one of three approaches to evaluate BDOC (reviewed

by McDowell et al. 2006): 1) measurement of DOC removal during controlled laboratory incubations (Holmes et al. 2008); 2) quantifying the kinetics of CO_2 production during controlled laboratory incubations (Wickland et al. 2007) and 3) measuring the removal of DOC as water moves through a bioreactor with microbes growing on glass beads (Yano et al. 2000). Other common approaches to assess BDOC in natural environments involve using indirect measures, such as elemental ratios (Hunt et al. 2000), molecular weight as determined by ultrafiltration (Meyer et al. 1987) and spectroscopic analyses including $SUVA_{254}$ and fluorescence spectroscopy (Kalbitz et al. 2003).

Spectroscopic analyses of DOM

Conventional analysis of aquatic DOM has focused on bulk measurements, due primarily to the heterogeneous nature of DOM and the analytic and interpretive difficulties associated with characterizing DOM fractions. In spite of these constraints, recent advances in spectroscopic analyses enable the rapid and precise characterization of DOM. For example, the specific UV absorbance ($SUVA_{254}$) of DOC, which is the average absorptivity at 254 nm, is highly correlated with aromatic C content (determined by ^{13}C NMR; Weishaar et al. 2003). Thus, SUVA measurements provide information about the chemical quality and biodegradability of DOM (Kalbitz et al. 2003).

As an alternative, fluorescence spectroscopy has been used successfully to trace changes in the chemical quality of aquatic DOM in watershed-scale studies. Aquatic humic substances, which comprise the largest fraction of DOM, account for a significant portion of the fluorescence occurring in natural waters (Green and Blough 1994). In particular, the quinone moieties contribute significantly to the fluorescence of humic substances (Klapper et al. 2002; Cory and McKnight 2005). More than 50% of the fluorescent component in natural waters is due to these quinone-like fluorophores (Cory and McKnight 2005). Quinones, a class of biomolecules found in all living organic material in aquatic and terrestrial ecosystems, act as electron transporters and pigments in cells throughout the electron transport system (ETS). Since quinones can cycle between

different oxidation states, the occurrence of different quinone fluorophores is a product of the redox conditions found in the environment (Cory and McKnight 2005).

Fluorescence spectroscopy can be used to generate the fluorescence index (FI = ratio of emission intensity at wavelengths 450/500 nm, obtained at excitation 370 nm) and three dimensional scans (excitation-emission matrices; EEM) of DOM (McKnight et al. 2001). The FI in conjunction with EEMs have been used to distinguish the source (autochthonous vs. allochthonous) of aquatic DOM as well as monitor seasonal changes in the chemical quality of aquatic DOM (McKnight et al. 2001; Hood et al. 2003). Excitation-emission fluorescence spectroscopy can also be analyzed using the multivariate modeling technique parallel factor analysis (PARAFAC), a three-way decomposition method similar to principal component analysis (Stedmon et al. 2005; Cory and McKnight 2005). PARAFAC allows the fluorescent signal of DOM to be decomposed into unique fluorescent groups whose abundance is related to DOM precursor material. This multivariate technique decomposes the fluorescent signature of aquatic DOM into individual components, thereby providing information about the composition and origin of DOM. Consequently, PARAFAC is well suited to the heterogeneous nature of DOM.

Research overview and chapter descriptions

DOM influences an array of biological, physical and chemical processes. Yet, there have been few integrated studies at the watershed-scale, particularly in high-latitude watersheds or in mesic to wet environments, aimed at developing an understanding of how major DOM source pools, such as wetlands, influence the quantity and quality of DOM delivered to streams and facilitate the loss of labile DOM from terrestrial ecosystems. My dissertation research addresses this information gap and contributes to our understanding of how ecological and hydrological processes interact to control biogeochemical processes at the watershed-scale. My dissertation research was conducted in coastal temperate watersheds in southeastern Alaska. In the coastal

temperate biome, which extends from northern Vancouver Island to Prince William Sound in Alaska, basic information is lacking about the variability of aquatic nutrient regimes, the interactions between abundant wetlands and stream chemistry and the chemical nature of DOM in forest watershed streams. Results from this dissertation provide new information on major influences on carbon dynamics in wetland dominated watersheds in a region where there are few anthropogenic stressors to complicate watershed interpretations. Moreover, insights from my research are transferable to other watersheds where wetlands play an important role in aquatic DOM dynamics.

The central aim of my dissertation is to improve our understanding of the role of terrestrial ecosystems, particular wetlands, in influencing the quantity and quality of stream DOM in coastal temperate watersheds of southeast Alaska. Within this area of research I focus specifically on whether wetlands are an important source of biodegradable DOM to aquatic ecosystems. I addressed this question through a combination of field and lab experiments in three watersheds and in three different soil types (bog, forested wetland and upland forest) near Juneau, Alaska. The bog and forested wetland were selected because these wetlands represent the most typical mapped wetland communities in southeast Alaska (USDA 1997), and the upland forest was selected to provide a mineral soil contrast to the two wetland types.

Within southeast Alaska, approximately 29% of the land area is classified as wetlands, with coverage ranging from <5% to 95% of total watershed areas. The most extreme result of the difference in wetland coverage is the simultaneous presence of brownwater (high DOC) and clearwater (low DOC) streams in adjacent watersheds. Because the proportion and type of wetland and mineral soils varies widely among southeast Alaskan watersheds, the concentration and chemical quality of DOM exported from individual watersheds may respond in very different ways through time. The well-defined, carbon rich watersheds of southeast Alaska therefore present an excellent opportunity to develop a process-level understanding of the coupling between different soil DOM source pools and stream biogeochemistry.

Watershed-scale biogeochemistry integrates seasonal changes in biotic and abiotic processes occurring in linked terrestrial and aquatic ecosystems. Thus, understanding how the chemical composition of soil DOM varies in different terrestrial source pools is important for elucidating the biogeochemical role of DOM within the soil profile and along the soil-stream continuum. In Chapter 2 of my dissertation, I evaluated how the chemical quality and biodegradability of soil solution DOM varies among four different wetland and forest soil types. This, in turn, provides an improved understanding of the potential for different soils to contribute labile DOM to aquatic ecosystems. I further evaluate the use of PARAFAC modeling of fluorescence excitation-emission spectroscopy as a tool to identify unique terrestrial sources of DOM from coastal temperate watersheds in southeast Alaska.

As allochthonous DOM moves through a watershed from its source in the soils to the watershed outlet, the composition of DOM reflects both source material and distance downstream along the soil-stream continuum. Thus, sampling along a soil-stream continuum is an ideal way to test hypotheses at the watershed-scale. In Chapter 3 of my dissertation, I evaluated the chemical quality and lability of DOM along a soil-stream continuum in three soil types in southeast Alaska. My primary hypothesis was that BDOC in soil solution and in streamwater for both wetland and upland forest sites is determined by the interaction between BDOC production/removal processes and seasonal changes in soil hydrology. I further proposed that the percentage of BDOC in streams would be higher during spring snowmelt and the fall wet season compared to the summer growing season, corresponding with high biotic demand during the summer growing season.

Dissolved organic matter concentration increases during floods in most streams and as a result, knowledge of how different DOM source pools (e.g. wetlands) influence the concentration and chemical quality of DOM exported during stormflows is essential for elucidating the cycling of C in watersheds. In Chapter 4 of my dissertation, I evaluated the importance of storms for facilitating the loss of labile DOM from wetland and upland forest watersheds. My hypothesis was that during storms, soil surface

horizons and streams will become tightly linked which will result in an increase in streamwater DOC and BDOC yields, and that shifts in the amount of BDOC will be dependent on the extent and type of wetland present within a watershed.

In Chapters 2-4 of my dissertation, I evaluated the production and export of biodegradable DOM from terrestrial to aquatic ecosystems. Because DOM is an important energy source for aquatic heterotrophs, DOC uptake studies are useful in elucidating the role of wetland-derived DOC in stream metabolism (e.g. Wiegner et al. 2005). In the fifth chapter of my dissertation, I conducted a series of slug additions using DOC derived from watershed soils and salmon carcasses to investigate the fate and metabolic importance of common allochthonous sources of DOC in temperate forest streams of southeast Alaska. In addition, I used fluorescence excitation-emission spectroscopy to evaluate longitudinal changes in the fluorescent properties of DOM during additions. My hypothesis was that stream uptake of DOC would be greatest during the spring runoff and fall wet season compared to the summer growing season, corresponding with terrestrial inputs of labile DOM to streams.

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CHAPTER TWO

FLUORESCENCE CHARACTERISTICS AND BIODEGRADABILITY OF DISSOLVED ORGANIC MATTER IN FOREST AND WETLAND SOILS FROM COASTAL TEMPERATE WATERSHEDS IN SOUTHEAST ALASKA[1]

Abstract

Understanding how the concentration and chemical quality of dissolved organic matter (DOM) varies in soils is critical because DOM influences an array of biological, chemical, and physical processes. We used PARAFAC modeling of excitation-emission fluorescence spectroscopy, specific UV absorbance ($SUVA_{254}$) and biodegradable dissolved organic carbon (BDOC) incubations to investigate the chemical quality of DOM in soil water collected from 25 cm piezometers in four different wetland and forest soils: bog, forested wetland, fen and upland forest. There were significant differences in soil solution concentrations of dissolved organic C, N, and P, DOC:DON ratio, $SUVA_{254}$ and BDOC among the four soil types. Throughout the sampling period, average DOC concentrations in the four soil types ranged from 9 – 32 mg C L^{-1} and between 23-42% of the DOC was biodegradable. Seasonal patterns in dissolved nutrient concentrations and BDOC were observed in the three wetland types suggesting strong biotic controls over DOM concentrations in wetland soils. PARAFAC modeling of excitation-emission fluorescence spectroscopy showed that protein-like fluorescence was positively correlated (r^2=0.82; $p<0.001$) with BDOC for all soil types taken together. This finding indicates that PARAFAC modeling may substantially improve the ability to predict BDOC in natural environments. Coincident measurements of DOM concentrations, BDOC and PARAFAC modeling confirmed that the four soil types contain DOM with

[1]Fellman JB, D'Amore DV, Hood E, Boone RD (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.

distinct chemical properties and have unique fluorescent fingerprints. DOM inputs to streams from the four soil types therefore have the potential to alter stream biogeochemical processes differently by influencing temporal patterns in stream heterotrophic productivity.

Key Words: Biodegradable dissolved organic carbon, dissolved organic matter, fluorescence, PARAFAC, peatland, soil biogeochemistry

Introduction

Dissolved organic matter (DOM) is a mixture of soluble organic compounds derived from both terrestrial and aquatic sources and in soils, plays an important role in the cycling of C, N and P. DOM controls the nutrient balance in terrestrial ecosystems by acting as a vector for dissolved losses of N, P and C (Qualls et al. 1991). DOM also provides a substrate for microbial metabolism, facilitates the transport of metals and plays an important role in soil formation (Kalbitz et al. 2000). These qualities of DOM differ in relation to the precursor organic material; thus, understanding how the chemical composition of soil DOM varies spatially and temporally is important for elucidating the biogeochemical role of DOM within the soil profile and along the soil-stream continuum.

Wetlands are an important source of dissolved organic carbon (DOC) to aquatic ecosystems (Mulholland 1997). As a result, wetland inputs of DOM to streams can have a profound impact on the chemistry (Billet et al. 2006) and biology (Sun et al. 1997) of aquatic ecosystems. In particular, aquatic DOC concentrations have been shown to be significantly correlated with peatland coverage (Aitkenhead et al. 1999), wetland area (Gorham et al. 1998) and wetland type (Xenopoulos et al. 2003). Despite the recognition that wetlands are a substantial source of DOC to surface waters, the chemical quality of DOC from different wetland types and how it varies seasonally is not well understood. Moreover, the importance of wetlands as a potential source of biodegradable DOC to support stream heterotrophic productivity has received little attention.

The biodegradation of DOC (BDOC) is an important process controlling DOC dynamics in soils, and controls on BDOC to a large extent are still poorly understood in soils (Kalbitz et al. 2000). In particular, DOM derived from wetland soils contains high concentrations of dissolved humic substances that have conventionally been considered recalcitrant and largely unavailable for bacterial degradation (Geller 1986). However, evidence suggests that this recalcitrant DOM may be more available than previously presumed and that terrestrially-derived humic substances might represent an important component of the streamwater BDOC pool (Moran and Hodson 1990; Volk et al. 1997). Since DOM is an important source of C and energy for microbial heterotrophs and given its heterogeneous nature, scientists have developed a variety of simple indicators for BDOC in natural ecosystems.

One common approach is to use elemental ratios, such as C:N, H:C or O:C, as an indicator of the biodegradability of DOM (Meyer et al. 1987; Hunt et al 2000). Another approach is the use of specific UV absorbance ($SUVA_{254}$), an indicator of aromatic C content, which has been shown to be negatively correlated with BDOC (Kalbitz et al. 2003a; Saadi et al. 2006). However, Marschner and Bredow (2002) found no relationship between $SUVA_{254}$ and BDOC and suggested BDOC of non-aromatic compounds varied greatly. A third approach uses molecular weight as determined by ultrafiltration (Meyer et al. 1987). The traditionally accepted model of biodegradation is that as the size of the molecule increases, the degree of recalcitrance to bacterial breakdown decreases (Saunders 1976). This view is no longer widely accepted since studies indicate high molecular weight compounds can be readily utilized by microbes (Amon et al. 1996). These findings suggest there are multiple factors controlling the biodegradability of DOM and that more advanced techniques for assessing BDOC in natural environments are necessary.

Recent advances in fluorescence spectroscopy enable the rapid and precise characterization of DOM and provide an alternative to the traditional approaches for predicting BDOC. Laboratory incubation studies have shown fluorescence spectroscopy can be used successfully to obtain information about the biodegradability of DOM

(Kalbitz et al. 2003a; Wu et al. 2003; Saadi et al. 2006). Excitation-emission fluorescence spectroscopy (EEMs) can also be analyzed using the multivariate modeling technique parallel factor analysis (PARAFAC), a three-way decomposition method similar to principal component analysis (Stedmon et al. 2003, 2005; Cory et al. 2005). PARAFAC decomposes the fluorescence spectra of DOM into independent components whose abundance can be related to differences in composition and source material. PARAFAC analyses have been used successfully in soil DOM studies to differentiate between different terrestrial sources (Ohno and Bro 2006) and to investigate the sorption of DOM onto mineral soils (Banaitis et al. 2006).

We used PARAFAC modeling of fluorescence EEMs, $SUVA_{254}$ measurements and BDOC incubations to investigate the chemical quality of DOM from four different forest and wetland soil types in coastal temperate watersheds of southeast Alaska. Our goal was to understand how the chemical quality and biodegradability of soil solution DOM varies between the different soil types. This, in turn, provides an improved understanding of the potential for different soils to contribute labile DOM to aquatic ecosystems. We further evaluate the use of PARAFAC modeling of fluorescence EEMs as a tool to identify unique terrestrial sources of DOM from coastal temperate watersheds in southeast Alaska.

Methods

Site descriptions and experimental design

DOM was examined in soil solution samples collected near Juneau, Alaska (58.2° N, 134.2° W). Juneau has a maritime climate with a mean annual precipitation of 1400 mm and a mean monthly temperature ranging from -2° C to 14° C at sea level. The heavily glaciated, mountainous terrain of southeastern Alaska, the cool climate and the abundant precipitation create a landscape mosaic of carbon-rich peatlands mixed with coniferous

forests dominated by *Picea sitchensis* and *Tsuga heterophylla*. Overall, wetlands account for approximately 30% of the land area in the Tongass National Forest (USDA 1997).

Three replicate field sites were established for four different forest and wetland soil types (bog, forested wetland, fen and upland forest) during the spring of 2006, yielding a total of 11 sites (only two replicate upland forest sites). The bog and forested wetland sites were selected because these wetlands represent the most typical mapped wetland communities in southeast Alaska (USDA 1997). The fen sites were included to represent the wetland diversity present in southeast Alaska, and the upland forest sites were selected to provide a mineral soil contrast to the three wetland types studied. All three wetland types are peatlands, characterized by the accumulation of organic matter due to frequent near-surface soil saturation.

The bog sites were mapped as a complex of deep, moderate to well decomposed peat (>1 m deep) that has accumulated over glacial till and were typical of the slope bog wetland type (NWWG 1988). Water and nutrient supply to the bog is dominated by atmospheric inputs although groundwater and surface runoff can be locally important. The forested wetland sites were typical of the raised peatland swamp (NWWG 1988) with 0.5-0.75 m deep peat overlaying glacial till. Forested wetland sites have formed on the same deposits as the bog, although forested wetlands maintain a different hydrologic regime where soil hydraulic conductivity is greater than in the bog but soil saturation is sufficient to create anoxic conditions.

Fen sites were typical of the rich fen (NWWG 1988) wetland type and have greater graminoid and forb diversity as well as more robust growth. Nutrient and water supply to the bog and fen sites are typified by extremes since fens receive large inputs through surface water and groundwater from the surrounding uplands as well as via precipitation. These hydrologic and geochemical inputs are responsible for the more neutral pH in fens. Upland forest sites are spodosols (Typic Humicryod) where soils are moderately deep and moderately well-drained, due to the steep slope present at the sites. The soils are colluvial material derived from bedrock dominated by igneous intrusive

material. The soils at all sites were characterized by soil profile descriptions to 1 m, although we present data from the top 25 cm (Table 2.1).

Field sampling

Soil solution samples were collected eight times for each site from May 9, 2006 until October 17, 2006. This period of time corresponds to the approximate length of the snow free season. Soil solution samples were collected from four, 25 cm deep piezometers and combined, yielding one sample from each of the sites per sample date. Piezometers were constructed from 3.1 cm PVC pipe and inserted in a small grid across the site. The 25 cm piezometer depth corresponds to the approximate acrotelm/catotelm boundary for all wetland sites. Piezometers were used to sample soil solution in the mineral soils because our interests were in collecting a bulk sample that is representative of the upland forest. Therefore, the soil solution in the upland forest represents a composite of DOM from the O (0-15 cm), E and upper B horizons (15-25 cm). All soil solution samples were field-filtered using pre-combusted, Gelman A/E glass fiber filters (nominal pore size 0.7 μm) and stored in the refrigerator until analysis, which occurred within 48 hours.

Dissolved C, N and P analyses

Concentrations of DOC (determined by non-purgeable organic carbon analysis) and total dissolved N (TDN) from soil solution samples were determined by high-temperature combustion using a Shimadzu TOC-V Organic Carbon and Total Nitrogen Analyzer with lower detection limits of 0.4 mg C L^{-1} for DOC and 0.1 mg N L^{-1} for TDN. Ammonium (NH_4-N) and nitrate (NO_3-N) were measured on a Dionex Ion Chromatograph (cation ICS-1500; anion DX-600), and dissolved organic N (DON) was calculated as the difference between TDN and inorganic N (NH_4-N and NO_3-N). The calculated error or lower quantification threshold for DON values during analytical runs was 0.2 mg N L^{-1} (square root of the sum of the squared analytical errors of TDN, NH_4-N and NO_3-N).

Soluble reactive phosphorus (SRP) was measured using the ascorbic acid method (Murphy and Riley 1962), total dissolved phosphorus (TDP) was measured using a persulfate digestion (Valderrama et al. 1981) in conjunction with the ascorbic acid method, and dissolved organic phosphorus (DOP) was calculated as the difference between TDP and SRP. A 10 cm quartz flow through cell was used for both SRP and TDP analyses to enable the detection of low P concentrations (1.0 μg P L^{-1}).

Spectroscopic analyses and PARAFAC modeling

Specific UV absorbance ($SUVA_{254}$) was measured using a 1.0 cm quartz cell on soil solution DOM following the procedures of Weishaar et al. (2003). Samples were allowed to warm to room temperature, analyzed on a Genesys 5 spectrophotometer and $SUVA_{254}$ was calculated as the UV absorbance at 254 nm per L mg-C^{-1} m^{-1}. Fluorescence excitation-emission matrices (EEM) of DOM were measured on a Fluoromax-3 (Jobin Yvon Horiba) fluorometer with a xenon lamp following the procedures of Hood et al. (2007). EEMs were created by measuring fluorescence intensity across excitation wavelengths ranging from 240 - 450 nm and emission wavelengths ranging from 300 - 600 nm. Samples were diluted to avoid inner filter effects by adding Milli-Q water to soil solution samples to provide an optical density of 0.02 at 300 nm (Green and Blough 1994). EEMs were corrected for instrument bias and Raman normalized using the area under the water Raman peak at excitation wavelength 350 nm.

PARAFAC modeling of fluorescence EEMs was conducted with MATLAB using the PLS_toolbox version 3.7 (Eigenvector Research Inc. 2006) following the procedures described in Stedmon et al. (2003; 2005). PARAFAC can take overlapping fluorescence spectra and decompose the data into score and loading vectors that are quantitative estimates of the relative concentrations of the components. If the correct number of fluorescent components is selected using the PARAFAC model, the components can be compared for each sample by determining the relative contribution of

each component to the total DOM fluorescence. Since DOM is a complex mixture of organic compounds, it is doubtful that each component represents a pure or specific fluorophore; rather, each component more likely represents a group of fluorophores with very similar fluorescence characteristics (Stedmon et al. 2005). We therefore refer to fluorescence components in this study as "humic-like, fulvic-like or protein-like" since these components are likely a mixture of similar fluorophores rather than pure fluorophores.

Using PARAFAC modeling, we identified a total of nine unique components within the fluorescence EEMs (Table 2.2). We validated our PARAFAC model using core consistency diagnostics (Ohno and Bro 2006) followed by a split plot analysis (Stedmon et al. 2005). The core consistency provides a quantitative measure of how well the spectral loadings represent variation in data. If the core consistency is not close to 100%, a different number of components should be selected. The core consistency score in our nine component model was 98.1% and the model explained 99.7% of the variability in the data set. To perform a split plot analysis, we randomly divided our data array into two separate halves of 165 EEMs each (total data set of 330 EEMs), applied the PARAFAC model to each half separately and repeated the analysis stepwise from 7-10 components. We selected nine components as the best model fit since we found good agreement in the spectral loadings for each dataset.

The percent contribution of each of the components was determined by quantifying the relative abundance of each component in comparison to the other components identified by the PARAFAC model. All nine components identified by our model have been previously identified as either part of a PARAFAC model (Stedmon et al. 2003, 2005; Ohno and Bro 2006) or through visual analysis of EEMs (Coble et al. 1996; Baker 2001). Of the nine components identified by the PARAFAC model, we focused our analyses on the following four components: component 1 (humic-like fluorescence), component 4 (fulvic-like fluorescence), component 8 (tryptophan-like fluorescence) and component 9 (tyrosine-like fluorescence). These four components were selected because they are commonly observed fluorophores in other studies and on

average, the relative contribution of the four components taken together accounted for approximately 51% (average of four soil types) of the total DOM fluorescence.

Biodegradable DOC incubations

In this study, we refer to BDOC as the DOC utilized by heterotrophic microbes through two different processes: 1) complete mineralization of C to obtain energy, and 2) incorporation of C into microbial biomass. BDOC was measured following a slightly modified protocol described in Qualls and Haines (1992). Soil solution samples were initially analyzed for DOC concentrations and then filtered through a 0.2 μm filter to remove the majority of microbial biomass. After filtration, 23 mL of the filtrate was transferred to ashed amber glass bottles and 2 mL of a bacterial inoculum was added. Caps were placed loosely on the bottles to allow air movement, and samples were incubated at 25° C for 30 days in the dark. After 30 days, the solution was re-filtered through a 0.2 μm filter, DOC was measured, and BDOC was calculated as the difference in DOC before and after the 30 day incubation. DOC analysis was also preformed on the bacterial inoculum and additional DOC provided to the soil water sample (ranged from 0.1 to 0.3 mg C L^{-1}) was added to the initial sample DOC concentration.

The bacterial inoculum was prepared by first collecting soil from the riparian zone at one of the study sites. Approximately 10 g of sieved, moist soil was combined with 50 mL of deionized water, gently shaken for 10 minutes, and allowed to settle over night. The bacterial inoculum was next filtered through a pre-combusted, Whatman GF/D filter, transferred to a pre-combusted glass bottle, diluted 1:1 with deionized water and incubated at 25° C for 24-48 hours before addition to the sample solution.

Statistical analyses

We used a mixed-model (Proc Mixed; SAS Institute, Inc. 2003), repeated measures analysis of variance (ANOVA) with a compound symmetry (CS) covariance structure in

conjunction with a Tukey's pairwise differences test to evaluate the effects of soil type on nutrient concentrations, BDOC and the relative contribution of PARAFAC components. All values for different sample dates were considered as repeated measurements. Because we were only interested in statistically comparing the four soil types, we did not statistically evaluate the temporal patterns within each soil type. Linear regression models were used to evaluate relationships between BDOC and the chemical characteristics of DOM using Proc GLM, SAS (SAS Institute, Inc. 2003).

Results

Dissolved C, N and P concentrations

For all sample dates taken together, average soil solution concentrations of C, N and P varied by more than 100% across the four different soil types (Table 2.3). Average DOC concentrations ranged from 9 mg C L^{-1} in the upland forest to 32 mg C L^{-1} in the forested wetland and were not significantly different between the forested wetland and bog ($p>0.05$). Concentrations of DOC in the fen and upland forest were significantly less than in the bog and forested wetland ($p<0.05$). There was no significant difference in DON concentrations between the three wetland types, whereas DON concentrations for the upland forest were significantly less than the three wetland types ($p<0.05$). The DOC:DON ratio in the forested wetland was significantly greater than those for the other three soil types ($p<0.05$), while the fen had the lowest DOC:DON ratio and was significantly less than in the forested wetland and bog ($p<0.05$). Despite having significantly lower DON concentrations than the other soil types, the DOC:DON ratio in the upland forest was lower than the bog and significantly lower than in the forested wetland. DOP concentrations in the fen were significantly greater than in the bog and the upland forest but did not differ from those in the forested wetland ($p<0.05$). DON and DOP were the dominant fractions of total dissolved N and P for all soil types and NH_4-N dominated the pool of DIN. Concentrations of NH_4-N and SRP were significantly greater

in the fen than those in the other soil types ($p<0.05$), while both NH_4-N and SRP were significantly less in the upland forest than the other soil types ($p<0.05$). Similar to DOC, the upland forest had the lowest average N and P concentrations observed.

Concentrations of soil solution DOC in the bog and forested wetland exhibited minima in the spring and fall and peaked at greater than 35 mg C L^{-1} during the mid-summer growing season (Fig. 2.1a). The upland forest showed a contrasting seasonal pattern where the greatest DOC concentrations (15-17 mg C L^{-1}) were observed during the spring/early summer and fall months. DON concentrations were high for all soil types during the spring sampling, decreased during the summer growing season to a low of 0.1 mg N L^{-1} in the upland forest and gradually increased during the autumn wet season (Fig. 2.1b). DOP concentrations for all soil types were greatest during the spring sampling followed by a gradual decrease throughout the remainder of the growing season (Fig. 2.1c). DOC:DON ratios in the bog and forested wetland were lowest during the spring and fall months compared to the summer growing season, while seasonal variation in DOC:DON ratios was small in the fen and upland forest (Fig. 2.1d). Seasonal variation in DIN and SRP concentrations was small in the bog, forested wetland and upland forest, whereas NH_4-N and SRP concentrations in the fen were greater during the summer compared to the summer and fall.

Spectroscopic properties of DOM and PARAFAC modeling

$SUVA_{254}$ of DOC proved to be a good indicator of differences in the chemical quality of soil DOM between soil types (Fig. 2.2a). Average $SUVA_{254}$ values ranged from 3.5 L mg-C^{-1} m^{-1} in the fen to 4.4 L mg-C^{-1} m^{-1} in the forested wetland and were significantly lower for the fen than those for the other soil types ($p<0.05$). This range in $SUVA_{254}$ values corresponds to an aromatic C content of approximately 25-34% when inferred from the linear model developed by (Weishaar et al. 2003). In evaluating the temporal patterns in $SUVA_{254}$, there was very little variation in $SUVA_{254}$ in the upland forest and fen (Fig 2.2b). However, $SUVA_{254}$ in the bog and forested wetland was lowest during the

spring, increased during the summer months and decreased slightly during the fall as $SUVA_{254}$ values returned to 4.1 L mg-C^{-1} m^{-1} in the bog and 4.2 L mg-C^{-1} m^{-1} in the forested wetland.

Visual analysis of the fluorescence EEMs for soil solution samples collected on June 17 revealed both similar and unique fluorophores among the different soil types (Fig. 2.3). In particular, all four soil types had a primary fluorescence peak at approximately 240 nm excitation and 450-460 nm emission. This fluorophore, which has been attributed to humic-like material of terrestrial origin (Stedmon et al. 2003) was very prominent at the bog while it was less well developed in the other soil types. Moreover, the fen had a fluorescence peak at approximately 280 nm excitation and 334 nm emission. This fluorophore, which has been linked to the amino acid tryptophan (Coble et al. 1996), was very prominent at the fen but it was less well developed at the bog and upland forest and non-detectable in the forested wetland EEM.

The humic-like component 1 (determined by PARAFAC modeling) was the dominant fluorescent component in soil solution DOM for all of soil types and was significantly greater in the bog than in the other three soil types ($p<0.05$; Fig. 2.4). In contrast, the fulvic-like component 4 was significantly greater in the forested wetland than in the other soil types ($p<0.05$). The ratio of the humic-like component 1 and the fulvic-like component 4 varied across the four soil types and was 1.7 for the forested wetland, 2.6 for the upland forest, 10.2 for the fen and 22 for the bog. Component 8, tryptophan-like fluorescence, was significantly greater than the tyrosine-like component 9 for the fen, upland forest, and forested wetland sites ($p<0.05$); whereas, there was no significant difference between the two components in the bog ($p>0.05$). The contribution of the protein-like fluorescence (the sum of tyrosine and tryptophan-like components) was significantly greater for the fen (23.4%; $p<0.05$) than for all other soil types, and the bog (10.4%) was significantly greater than the forested wetland (4.6%; $p<0.05$), but did not differ from the upland forest (10.1%; $p>0.05$).

Biodegradability of DOC

Consistent with the low DOC:DON ratios and low $SUVA_{254}$ values, soil solution BDOC was significantly greater for the fen than in the other three soil types ($p<0.05$; Fig. 2.5a), while BDOC was significantly greater in the bog than in the forested wetland but did not differ from the upland forest ($p<0.05$). During the incubations, an average of 6.2, 7.3 and 2.7 mg C L^{-1} was consumed for the fen, bog and upland forest sites, respectively. Average BDOC concentrations in the forested wetland (7.2 mg C L^{-1}) were greater than in the fen, although the fraction of BDOC was nearly half (23%) that reported for the fen (42%). Similar to $SUVA_{254}$, there was very little temporal variation in BDOC for the upland forest; however, BDOC was greatest in the spring and fall compared to the summer months in the three wetland types (Fig. 2.5b). DOC:DON ratios, $SUVA_{254}$ values and the contribution of the humic-like component 1 were all negatively correlated with soil solution BDOC for all sites taken together (Fig. 2.6a-c). Therefore, as the C:N ratio and the aromatic C content of the DOM increased, the biodegradability of DOM decreased. In addition, protein-like fluorescence was a strong predictor of DOC biodegradability for all soil types taken together (Fig. 2.6d).

Discussion

Dissolved C, N and P concentrations

The organic C, N and P concentrations reported in this study fall within the range reported in other studies of forested (Qualls and Haines 1991; Michalzik et al. 2001) and wetland soils (Fraser et al. 2001; Blodau et al. 2004), which supports the idea that DOM concentrations in wetland soils are significantly greater than upland forest soils. The organic forms of N and P dominated soil solution for all soil types and suggests that DON and DOP are an important component of nutrient cycling in coastal temperate soils. The significantly lower concentrations of DOM in the upland forest are not surprising given the shallow depth of the O horizon as well as the potential for sorption of DOM by

underlying mineral horizons (McDowell and Likens 1988; Qualls and Haines 1991). The different organic C, N and P concentrations observed between the wetland types are likely a function of distinct ecosystem nutrient dynamics caused by differences in site characteristics (i.e. soil properties), hydrologic inputs, and dominant vegetation. For example, the greater DOC concentrations in the bog and forested wetland could result from seasonal water table drawdown in combination with greater rates of organic matter decomposition and subsequent DOC production in the aerobic surface horizons (McKnight et al. 1985; Fraser et al. 2001). The fen in contrast had significantly lower DOC concentrations than the bog and forested wetland and suggests that continuous soil flushing in fens results in low pore water DOC concentrations (Urban et al. 1989).

Seasonal changes in DOC and DON concentrations have been previously documented in both wetland (Devito et al. 1989; Fraser et al. 2001) and forested landscapes (Qualls et al. 1991; Yano et al. 2004). Concentrations of DOC and DON exhibited contrasting seasonal patterns in the bog and forested wetland suggesting controls on DOC production and/or removal may be different than those for DON. During the spring snowmelt period, low wetland DOC concentrations can be attributed to prolonged soil saturation and subsequent dilution of soil pools of DOC (Fraser et al. 2001; Worrall et al. 2002). However, DOP and DON concentrations exhibited maxima during the spring and suggests decreased biotic demand (Devito et al. 1989) in combination with soil freeze-thaw events (Fitzhugh et al. 2001) can result in a pool of DON and DOP in soil solution that is potentially available to flush to streams. As a result, DOC:DON ratios exhibited minima during the spring. With the onset of the summer growing season, biotic demand for DON and DOP increases, water table drawdown occurs followed by higher rates of DOC production, which results in a DOC:DON ratio typically greater than 40 in the bog and 50 in the forested wetland. DOC concentrations once again decrease during the late summer/fall wet season as the supply of DOC becomes exhausted and DOC:DON ratios approach near spring values. Our findings suggest strong biotic control over DOM concentrations in wetland soils, which is similar to previous research in five Canadian peatlands that found N and P

retention during the summer growing season and net N and P export during the spring (Devito et al. 1989).

For upland forest sites, temporal patterns in DOM concentrations were similar indicating similar controls on the production and/or retention of DOM in forest soils (Neff et al. 2000). The greater DOM concentrations during the spring and fall months can be attributed to rising water tables associated with snowmelt and large precipitation events. When these events occur, water infiltrates into the soil causing water tables to rise into the organic horizons. Soluble organic material that has built up in the organic layers can be solubilized and potentially leached from the soil. These findings suggest concentrations of DOM are not tightly controlled by microbial demand for N and P in the soil but rather both production/degradation and physical removal processes interact to control DOM concentrations in upland forest soils.

Biodegradable DOC

The biodegradable fraction of DOC in the upland forest reported in our study (30%) was similar to values reported previously for pine and hardwood forest soils in central Massachusetts (10-45%; Yano et al. 2000) as well as for mixed hardwood soils in the southern Appalachians (20-30%; Qualls and Haines, 1992). In evaluating BDOC in wetland soils, approximately 40% of the initial DOC was consumed during incubations in Japanese mountain bog pools (Satoh and Abe 1987) and an average of 45% was consumed from freshwater marshes (Mann and Wetzel 1995). Moreover, 22% of the initial DOC was consumed from two cedar bogs in the Pine Barrens region of New Jersey (Wiegner and Seitzinger 2004); thus, our estimates of BDOC in wetlands (23-42%) fall within the range of other incubation studies. In our study, the amount of DOC consumed from the forested wetland (7.2 mg C L^{-1}) was greater than in the fen (6.2 mg C L^{-1}), although the percentage of DOC consumed in the forested wetland was nearly half than reported for the fen. This suggests that both the percentage and the amount of DOC consumed could be equally as important when evaluating BDOC in soils.

Seasonal patterns in BDOC were observed in the three wetland types which is consistent with other studies of wetlands soils that have found BDOC to be greatest during the spring and fall compared to the summer (Wiegner et al. 2004). Similar to the seasonal patterns observed in $SUVA_{254}$ values and DOC:DON ratios, concentrations of BDOC strongly reflect biotic controls in wetland soils. In contrast, we observed no seasonal patterns in BDOC in the upland forest which is consistent with the results in other hardwood forests soils (Boyer and Groffman 1996). However, other studies in forest soils have found seasonal changes in BDOC (Qualls and Haines 1992; Yano et al. 2000). These findings suggest that in upland forest soils, multiple factors interact to control BDOC concentrations because labile DOC is actively removed by the heterotrophic community while at the same time, modified in its composition by the adsorption of recalcitrant fractions of DOM by mineral soils (Qualls and Haines 1992).

Effects of nutrients on biodegradable DOC

Many factors can affect the amount of BDOC in soil solution including temperature, nutrient availability, water stress, bacterial community composition and the chemical characteristics of DOM (Del Giorgio and Davis 1998). As a result, BDOC is determined by the dynamic balance between the production and consumption of DOM in the soil. Previous BDOC experiments have shown N and P can limit bacterial growth efficiencies. In our study, BDOC was correlated with both DOC:DON ratios and protein-like fluorescence, which is consistent with laboratory trials that have shown amino acids to be a readily available source of C, N and energy for heterotrophic microbes (Ellis et al. 2000). We also found BDOC to be mildly correlated with concentrations of both DOP (r^2=0.41; $p<0.05$) and DON (r^2=0.35; $p<0.05$) but more importantly, BDOC was poorly correlated with DIN (r^2=0.21) and SRP (r^2=0.25). These results indicate microbes were predominantly using organic sources of N and P to satisfy growth demands. However, given that the total N and P concentrations were relatively low in comparison to the amount of DOC consumed during incubations (for every 1 mg C L^{-1} consumed, microbes

require 40 μg N L^{-1} and 8 μg P L^{-1} to satisfy growth requirements using a bacterial growth efficiency of 0.4 and a bacterial molar ratio for C:N of 10 and C:P of 50), we suggest that much of the DOC consumed during incubations was not incorporated into biomass but rather was respired as CO_2 through waste respiration, as observed in Wiegner and Seitzinger (2004). Therefore, N and potentially P could have limited microbial uptake of DOC during incubations in the bog, forested wetland and upland forest soils, which is consistent with the idea that net primary production is frequently limited by N in freshwater wetlands (summarized by Aerts et al. 1999) and in temperate forests (Vitousek and Howarth 1991).

Indicators of biodegradable DOC

The ratio of DOC:DON in soil solution DOM proved to be a good predictor of biodegradable DOM supporting the idea that microbes grow more efficiently on DOM with low C:N ratios (Hunt et al. 2000; Wiegner and Seitzinger 2004). We also found a strong negative correlation between BDOC and $SUVA_{254}$, consistent with other studies showing a relationship between aromatic C content and BDOC (Kalbitz et al. 2003a; Marschner and Kalbitz 2003; Saadi et al. 2006). These results suggest that seasonal changes in the N and aromatic C content of DOM can influence the biodegradability of DOM in soils.

PARAFAC components were good predictors of BDOC for all soil types taken together. The humic-like component 1 was negatively correlated with BDOC, which is consistent with previous studies showing that fluorophores with long emission wavelengths are highly conjugated and more aromatic in nature (Coble et al. 1996; Stedmon et al. 2003). Therefore, the humic-like component 1 provides an independent indicator that aromatic C content can be used to predict BDOC in soil waters. The relative contribution of protein-like fluorescence was a very strong predictor of BDOC in soil solution. Previous studies have also used simple fluorescence indicators, such as a humification index (Kalbitz et al. 2003a) or tryptophan-like fluorescence intensities (Wu

et al. 2003; Saadi et al. 2006), to study DOM biodegradation. However, there are several reasons why protein-like fluorescence may be a more useful predictor of BDOC compared to other fluorescent indicators. First, PARAFAC modeling determines the relative contribution of tryptophan and tyrosine-like fluorescence to the total pool of DOM fluorescence. Even though fluorescence intensities may be used to predict total hydrolyzable amino acid concentrations (Yamashita and Tanoue 2003), protein-like fluorescence is a better indicator of more favorable C:N ratios for the microbial utilization of DOM.

Tyrosine and tryptophan-like fluorescence also appear to indicate differences in the form or degree of amino acid degradation. Tyrosine has been shown to fluoresce well in its monomer form or when tryptophan is present in low concentrations, suggesting that tyrosine-like fluorescence indicates more degraded peptide material (Mayer et al. 1999; Yamashita and Tanoue 2003, 2004). These same studies have also suggested that samples dominated by tryptophan-like fluorescence may indicate the presence of intact proteins or less degraded peptide material. Our findings suggest that using the combined fluorescent signal for both amino acids more effectively predicts the biodegradability of DOM than using tryptophan-like fluorescence alone. Overall, the strong positive relationship between protein-like fluorescence and BDOC in our study suggests that PARAFAC analysis of DOM may represent a substantial advancement over other optical measurements in the ability to predict the biodegradability of DOM in soil solution.

Relationships between soil types, BDOC and the chemical quality of DOM

The relative contribution of PARAFAC components differed between the four soil types suggesting there are distinct differences in the chemical properties and lability of DOM between the soil types. The fen sites had the greatest fraction of BDOC among the soil types, which is consistent with the high protein-like fluorescence, low C:N ratios and low aromatic C content. Minerotrophic fens have been shown to possess greater rates of primary production (summarized by Aerts et al. 1999), plant litter decay and enhanced

rates of nutrient cycling than in bogs. As a result, the highly productive vascular plants, either through root exudates of carbohydrates and amino acids (Eviner and Chapin 1997) or litter decay (Yano et al. 2000), are likely the reason for the abundance of labile DOM present in the fen. This finding corroborates other studies (McDowell and Likens 1988; Yano et al. 2000) that suggest there is a significant contribution of recently fixed C to biodegradable DOM in the soil.

Significant differences in PARAFAC components also existed among the bog, forested wetland and upland forest. The humic-like component 1 is the dominant fluorescent component in the bog which is consistent with the idea that DOM in peat bogs is largely comprised of humic acids (Gondar et al. 2005). In the forested wetland and upland forest sites where organic horizons overlay mineral soils, the fulvic-like component 4 contributes greater to DOM fluorescence than in the bog and the humic-like to fulvic-like ratio is less than 3. This finding corroborates previous research in a northern hardwood forest showing that humic acids dominate the surface organic horizons and decrease with depth in the soil profile until the more mobile fulvic acids eventually became the dominant fraction in the lower horizons (Ussiri et al. 2003). Another possible reason for the greater fulvic acid content in the upland forest and forested wetland is the potential for lateral transport of DOM downslope through the soil, which has been suggested to occur in forested histosols of southeast Alaska (D'Amore and Lynn 2002). This type of water movement would most likely transport fulvic-rich DOM because humic acids usually precipitate out and accumulate in organic horizons and fulvic acids tend to remain soluble and move downward with percolating water (Ussiri et al. 2003).

The significantly greater contribution of tryptophan-like fluorescence in comparison to tyrosine-like fluorescence in the upland forest, forested wetland and fen indicates that the protein containing DOM is of relatively recent origin or is relatively unaltered (Mayer et al. 1999; Yamashita and Tanoue 2003, 2004). This would suggest that the lability of this DOM is closely related to the chemical quality of the DOM precursor material. In particular, plant litter extraction experiments have shown that

higher quality litter contributes more BDOC to soils than low quality litter (Boyer and Groffman 1996). Therefore, a potential reason for the low quality DOM at the forested wetland is the high lignin content and aromatic litter of *Tsuga heterophylla* (C:N ratio >80; Prescott and Preston 1994), which is the dominant conifer in forested wetlands.

Tryptophan and tyrosine-like fluorescence were not significantly different in the bog in contrast to the other three soil types. This proportionally higher tyrosine-like fluorescence suggests greater degradation of amino acid containing DOM in the bog. Water movement in bogs has been shown to be predominantly in the vertical direction, rather than in lateral directions (McKnight et al. 1985). This long residence time for DOM in the bog soils could lead to a high degree of microbial modification of the original source material. Moreover, research from Mer Blue bog, Canada has shown that the fluorescent properties of soil solution DOM changed from plant-derived to more microbial-like with depth in the soil profile, which was attributed to the microbial consumption of available DOM (Fraser et al. 2001). We therefore propose the pool of DOM in bog soil waters reflects both substantial microbial modification of the original source material and subsequent production of more microbial-like DOM. Since this DOM released into bog soil solution can occur through the biodegradation of microbial cell walls as well as the release of microbial metabolites (Guggenberger et al. 1994; Kalbitz et al. 2003b), such as carbohydrates and proteins, we suggest the high protein-like fluorescence and labile DOM present in the bog is the result of the production of this microbial-like DOM.

We compared DOC:DON ratios with the ratio between the humic-like component 1 and the fulvic-like component 4 and found that as the DOC:DON ratio increases, there was a decrease in the humic:fulvic ratio (r^2=0.46; $p<0.001$; data not shown). This finding indicates that the humic-like component 1 has a greater N content, which is consistent with the lower C:N ratios of extractable humic acids in comparison to fulvic acids (Ussiri and Johnson 2003; Gondar et al. 2005). Even though the contribution of DOM fluorescence to the total pool of DOM is still unknown, DOC:DON analysis reveals components 1 and 4 of our PARAFAC model resemble humic and fulvic acids extracted

from soils. Our results suggest DOM fluorescence combined with PARAFAC analysis could be used as a proxy for tracing the dynamics of the bulk pool of DOM in natural ecosystems.

Conclusions

We found an average of 23-42% of the DOM in soil solution from the four soil types is biodegradable. Even though the bulk of the DOM pool (58-76%) was found to be refractory, 2.7 to 7.3 mg C L^{-1} of DOC was consumed during incubations from the four soil types. This suggests that the DOM derived from wetland soils could be an important component of the streamwater pool of BDOC. The temporal changes observed in DOM concentrations indicate DOM inputs to streams from the different soil types have the potential to alter stream biogeochemical processes differently by influencing stream heterotrophic productivity. We further suggest that DOM dynamics within the three different wetlands may respond differently to climate change or different management practices and that these wetland types should be evaluated separately in future assessments of wetland ecosystem function. Therefore, attempts to lump these wetlands into a homogenous ecosystem for climate models should be conducted with caution.

Coincident measurements of $SUVA_{254}$, BDOC and PARAFAC modeling of fluorescence EEMs confirmed that different terrestrial source pools contain DOM with distinct chemical properties and that these terrestrial source pools have a unique fluorescent fingerprint. Since PARAFAC modeling of DOM fluorescence is a precise and rapid technique for tracing DOM dynamics in soils, its application for intensive temporal and spatial sampling protocols is possible. Taken together, our findings suggest that PARAFAC analysis of fluorescence EEMs has the potential to be used as an ecological tool to trace the movement of DOM from different terrestrial source pools along the soil-stream continuum.

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