AN ABSTRACT OF THE DISSERTATION OF

Yuriko Yano for the degree of Doctor of Philosophy in Forest Science presented on June 12, 2002.

Title: Characteristics of Dissolved Organic Matter (DOM) and its Stabilization in a Forest Soil.

Abstract approved ________________________________

Phillip Sollins

________________________________

Kate J. Lajtha

Dissolved organic matter (DOM) plays an important role in transport of C and essential nutrients such as N, P and S. DOM is also critical for the formation of soil organic matter (SOM), which is the largest terrestrial C pool. Nonetheless, we lack a basic understanding of what controls immobilization and mobilization of DOM. I conducted a parallel laboratory and field study to examine: 1) the effect of litter quality on DOM chemistry, and 2) the effect of DOM chemistry on immobilization in the mineral soil. For field study, an ongoing long-term manipulation of litter inputs (Double Litter=doubled annual leaf litter inputs; Double Wood=doubled woody litter inputs; and Control= normal litter inputs) in a Douglas-fir old-growth forest in the Pacific Northwest was chosen.

In the lab study, degree of litter decomposition strongly influenced the chemical composition of the water extracts. For both needle and wood extracts, the hydrophobic acid fraction increased and the hydrophilic neutral fraction decreased from newly-fallen to well-decomposed DOC sources.

Contrary to the laboratory results, no difference was found in composition of the O-horizon leachate among Double Litter, Double Wood and Control treatments after 4 years of litter manipulation, in spite of significant differences in total C, total N and C:N ratios of the O-horizon material. Possibly, microbial degradation decreased differences in DOM chemistry. Alternately, DOM production from native O-horizon material may be much greater than from newly added litter.

In field and lab studies, the removal of two acid fractions (hydrophobic and hydrophilic acids) accounted for most of the total DOC decrease. Because the hydrophobic neutral fraction was little sorbed, ligand exchange rather than hydrophobic interaction was suggested to be the major mechanism of DOM sorption.

Concentrations of DOM in incoming water and DOM removal were positively correlated with a slope of ~1.0 and negative intercept regardless of season and %hydrophilic neutral, the most biodegradable fraction, of incoming DOM, suggesting that the removal was mainly abiotic (sorption) and that there was constant net release of DOM from the soil layers independent of sorption.

# Characteristics of Dissolved Organic Matter (DOM) and its Stabilization in Forest Soil

By
Yuriko Yano

A DISSERTATION

Submitted to

Oregon State University

In partial fulfillment of
the requirements for the
degree of

Doctor of Philosophy

Presented June 12, 2002
Commencement June 2003

# Acknowledgement

I thank my two advisors; Dr. Kate Lajtha for being always patient and supportive and finding a way out when I came across a problem, and Dr. Phil Sollins for giving me opportunities to learn not only subjects that are related to my dissertation but also things such as philosophy in science and the history of human societies.  I also thank my committee members Drs. Mark Johnson, Dave Myrold, and Peter List for their suggestions, insightful comments, and support on my project.

It would have been impossible to complete my project without the support of many professors, who kindly offered me the use of their lab space and equipment.  These are: Drs. Bob Griffiths, Kermit Cromack, Jerry Qualls, John Baham, and Mark Harmon. Additional thanks goes to Dr. Lisa Ganio for her help on my statistical analysis.  I would also like to thank the following people who contributed to the completion of this dissertation in their special ways.  Thanks to Jay Sexton, John Moreau, and Suzanne Remillard for removing a number of down logs on the study site after a destructive snow storm in the winter of 1998; Bruce Caldwell, Scott Holub, Julie Spears, Shana Pennington, and Bonnie West for helping me in the field and lab; Shannon Claeson, Kari O'Connell, Michele Pruyn, and Elly Vandegrift for keeping me sane by dragging me out of the lab when it was needed; Marc Kramer, Steve Perakis, Chris Swanston, and Rota Wagai for being the only people around me who were happy to share my passion for DOM with me; my wonderful housemates – Cesar Llave, Anna Versluis, and Kris Whitbeck – for providing me of extra space and comfort; and my parents, Kunihiko and Kyoko Yano, and my brother, Kenji Yano, who were supportive no matter what the situation I was in and taught me how to see a positive side of the life during a hard time.

Thanks to all my friends for their patience and support.  These include: Dominique Bachelet, Rachel Beitner, Nancy Chaney, Tina Draisbach, Kei Fujimura, Charlie Lefevre, Bob Peck, Admir Giachini, Marya Madsen, Etsuko Nonaka, Eduardo Nouhra, Jodi Sorenson, and Steve Wondzell.

**Last but not least, special thanks goes to Erik and Chava Beever for getting me started writing this dissertation and welcoming me with big smiles (wag! wag!) and kisses (lick! lick!) when I returned from work.**

Doctor of Philosophy dissertation of Yuriko Yano presented on June 12, 2002.

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

Yuriko Yano, Author

# CONTRIBUTION OF AUTHOR

Bruce Caldwell was involved with designing the study and establishing the study site for Chapter 2.

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# CHAPTER 1
# INTRODUCTION

## GLOBAL C CYCLING – PROBLEMS

Atmospheric $CO_2$ has risen rapidly since the late 19th century due to the increased flux of $CO_2$ to the atmosphere through human's fossil fuel combustion and land-use change (Aber and Melillo 2001). Because $CO_2$ is one of the dominant greenhouse gases on Earth, most global climate models predict substantial increases in atmosphere temperatures (1.5-5.5 °C) if $CO_2$ continues to rise (Schlesinger 1997). Patterns of precipitation as well as distributions of plant and animal communities are also predicted to be altered by the global warming. To sustain a habitable environment on Earth, we must understand how to control $CO_2$ production and sequestration.

Forests are significant sinks for atmospheric $CO_2$ (Ciais et al. 1995; Fung 2000; Pacala et al. 2001; Clark 2002). Therefore, understanding what causes increases and decreases of C stored in forest ecosystems is critical for modeling the global C budget, and for establishing forest management plans for controlling atmospheric $CO_2$.

The major processes that control the size of vegetation C pools (e.g., photosynthesis, respiration) are well documented and modeled (e.g., Melillo et al. 1993; McGuire et al. 1997; Cao and Woodward 1998; Tian et al. 1998). However, modeling changes of SOM pools has not been as successful as modeling vegetation, in spite of the fact that soil organic matter (SOM) is the largest organic-C pool on land. Short-term (i.e., decadal, and perhaps even century-scale) changes in SOM are difficult to measure due to large background SOM stores, and we lack a basic understanding of the processes that form and stabilize SOM. Therefore, understanding the fundamental processes that control stabilization and destabilization of SOM is essential for modeling changes in terrestrial C stores.

## THE ROLE OF DOM IN SOM FORMATION

Stabilization of dissolved organic matter (DOM) is a major process of SOM formation. DOM contains C (DOC), as well as organically bound N (DON), and can distribute those elements from plant detritus in the forest floor to mineral soil. In forest ecosystems, DOM concentration is generally highest immediately below the O horizon and decreases as the solution percolates through mineral soil horizons (e.g., McDowell and Likens 1988; Qualls and Haines 1991; Michalzik et al. 2001). Abiotic sorption onto soil minerals and biotic immobilization are two known DOM stabilization processes. Abiotic sorption has generally been considered to be more important than biotic immobilization, because only a small portion of total soil DOC (3-19 %) has been determined to be labile (McDowell and Likens 1988; Qualls and Haines 1992) and this percentage is too small to explain a 100-fold reduction in DOC between O and B horizons. However, Yano et al. (2000) measured a much higher proportion (10-40%) of bulk DOC as biodegradable in a hardwood and a conifer forest in Massachusetts, suggesting that abiotic sorption may not always be the predominant process that stabilizes DOM into SOM.

## DOM CHARACTERISTICS AND DYNAMICS – BACKGROUND

### Origin and Chemistry of DOM

Dissolved organic matter (DOM) is operationally defined as those organic molecules that pass a filter of a certain pore size, most commonly 0.7 µm. Sources of DOM can include plant tissue, litter, plant metabolites (e.g., root exudates, mucilage, sloughed cells), soil organic matter (SOM), and decaying microbial cells or microbial metabolites (e.g., extracellular enzymes). Therefore, DOM in soil consists of a wide variety of chemical compounds.

The small proportion of DOM that has been chemically identified is mostly low molecular weight compounds such as amino acids, proteins, and sugars that are free from humic substances (reviewed in Kalbitz et al. 2000). Because most DOM is complex, an operational definition (e.g., by functional groups, molecular size, surface charge, UV absorption) is often used to characterize DOM. A commonly used method separates DOM based on differences in affinity between DOM and three types of resins (hydrophobic, cation-exchange, and anion-exchange; Leenheer 1981; and modified by Qualls and Haines 1991) (Fig 1-1).

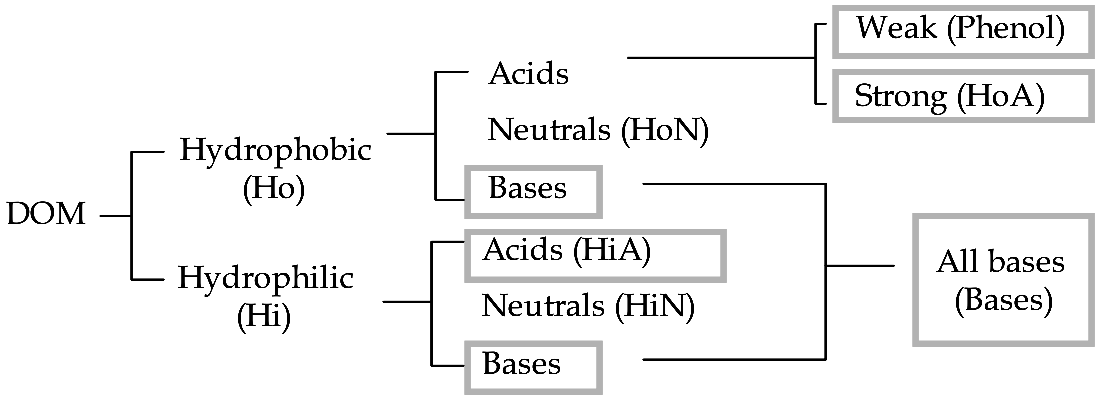


**Fig. 1-1.** Functionally different DOM fractions separated by the chemical fractionation method (Leenheer 1980; and modified by Quallis and Haines 1991). Six fractions shown in boxes are reported in this study.

Using these resins in combination, DOM can be separated into 6 functional groups. The weak hydrophobic acid fraction (Phenol) includes small phenolic compounds (e.g., tannin and flavenoids). Strong hydrophobic acids (HoA) are mainly microbially altered plant-derived material (lignin and ligno-cellulose), have high content of aromatic C, and include larger molecular size humic substances and humic-bound amino acids and carbohydrates. The hydrophobic neutral fraction (HoN) is plant-derived material that is less microbially altered, containing waxes and fatty acids. This fraction also contains microbial lipids. The hydrophilic acid fraction (HiA) contains partly microbially synthesized and partly plant-derived material that is lower molecular size humic-like substances with high carboxyl-to-C ratios. The hydrophilic neutral fraction (HiN) contains non-humic-bound carbohydrates, mainly of microbial origin, and may contain simple sugars (e.g., hexose, deoxysugars). The hydrophobic and hydrophilic base fractions contain free amino acids, peptides and proteins. Several studies have shown that these operationally defined chemical fractions are well associated with certain chemical structures or properties. Using the combination of $^{13}$C-NMR, pyrolysis-mass spectrometry techniques, and wet-chemical analyses, Guggenberger et al. (1994) found evidence for higher degree of oxidative biodegradation (i.e., more side-chains are oxidized) for HiA than for HoA.

## DOM production in soil -- conceptual models

Recently proposed conceptual models suggest that DOM is produced in soil water in a two-step process (Christ and David 1996; Kalbitz et al. 2000; Wagai and Sollins 2002): 1) replenishment of a leachable organic matter pool, followed by 2) leaching of the organic compounds in the leachable pool (Fig. 1-2). The first step involves both biotic and abiotic processes (all arrows from “solid material” to “leachable OM pool” in Fig. 1-2) and occurs as: release of soluble organic compounds from live plant and microbial cells such as root exudation and extracellular-enzyme secretion (biotic, arrows marked 1), dissolution of soluble compounds from dead plant and microbial cells (abiotic, arrow marked 2), or breakdown of insoluble organic compounds by extracellular enzymes (biotic, arrows marked 3). Organic compounds in this leachable pool can undergo abiotic or biotic alteration (e.g., polymerization, arrows marked 4), or can be removed from the pools by biotic uptake (arrows 5) or abiotic sorption (arrows marked 6) to a solid material. When soil dries, DOM in the leachable pool precipitates out on plant debris or SOM (abiotic, arrows marked 6).

The second step is abiotic. Pore size, preferential flow, and concentration gradient between the leachable OM pool and moving water control DOM leaching into groundwater (arrows marked 7). Thus, both abiotic and biotic processes in the soil control the production and chemistry of DOM.

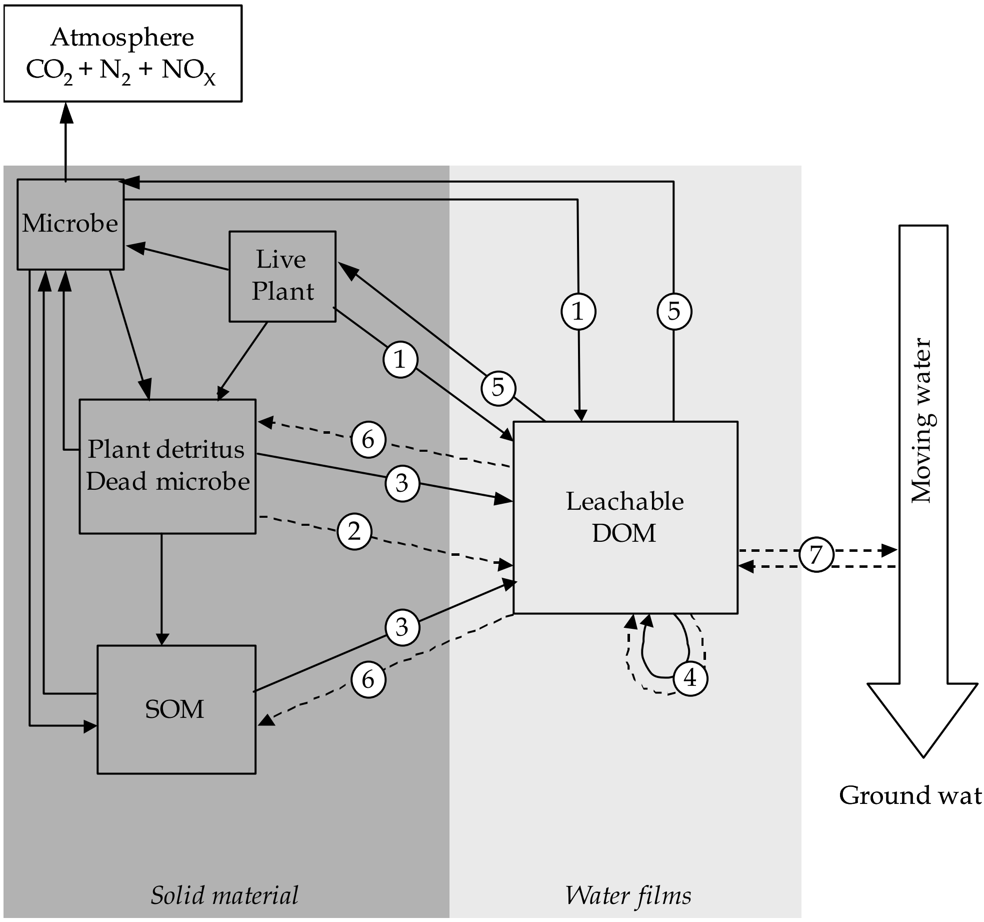


**Fig. 1-2**. Conceptual model of the processes involved in formation of DOM. Dark-shaded box indicates solid OM pools in soil, and light-shaded box indicates water films exist on soil particles. Solid lines indicate biotic processes and broken lines indicate abiotic processes. See text for explanations of numbered arrows.

## Dynamics and chemistry of DOM

The degree of abiotic sorption varies according to the chemistry of DOM. Using the fractionation scheme of Leenheer (1981), Dai et al. (1996) studied sorption of forest floor DOM by a B horizon soil. They observed that 50% of the HoA and 10% of the HiA fractions were sorbed on the mineral soil, with significantly less sorption observed for the other fractions. Recent studies have also found that Ho-DOC has a stronger affinity for mineral soil than does HiA (Kaiser and Zech 1997, 1998).

Qualls and Haines (1992) found that the biodegradability of bulk DOM was associated with the proportion of HiN fraction in the initial solution. Similarly, Jandl and Sollins (1997) found that most biodegradable DOM was in the HiN fraction. Because the C and N content differs for the chemical fractions (i.e., HiA and Bases are N-rich, HoA and HoN are C-rich; Qualls and Haines 1991) and sorption and degradability are different for each fraction, changes in composition are expected with removal of DOM with increasing soil depth.

## Effect of litter type on DOM chemistry

The chemical composition of plant OM is known to differ from tissue to tissue. In general, leaves are rich in N due to high enzyme levels needed for photosynthesis. Woody tissues, on the other hand, are extremely rich in C and poor in N due to the high content of structural compounds such as lignin, cellulose and ligno-cellulose. Recent $^{13}$C-NMR analyses have shown that leaves contain more alkyl C than do wood or fine roots (Zech et al. 1997). Correspondingly, the contribution of aromatic C to total OM is greater in leaves (Kögel-Knabner 1997). In addition to litter type, the degree of decomposition also alters the chemical composition of plant litter. Generally, as litter decays alkyl C levels increase and O-alkyl C level (e.g., carbohydrate C and C-O esters) decrease (Kögel-Knabner 1997).

Differences in plant litter quality may have profound effects on soil solution chemistry and ecosystem processes. Current DOM process models include biological decomposition as a key factor that control DOM chemistry and dynamics. Currie and Aber (1997) related DOC in forest-floor leachate to decomposition rates of litter, which varied by the chemical composition of litter (%lignin, %cellulose, %extractives, and %N). Neff and Asner (2001) linked biotic and abiotic processes in their DOC model. The authors also used lignin:N ratio to predict DOM chemistry. In both approaches, the underlining assumption was that the chemical composition of DOM is determined by litter quality, but, to my knowledge, this assumption has not been tested in the field. To model DOM dynamics and SOM formation, we need a basic understanding of the effects of litter quality on DOM composition, and of DOM composition on SOM formation (sorption).

## Research questions

In the following two chapters I examine three questions in field and laboratory study:

1. How does plant litter quality affect DOM chemistry?
2. How do differences in plant litter inputs (quality and amount) affect the amount and C:N of SOM?
3. How does DOM chemistry affect potential abiotic sorption?

Chapter 2 covers the first two questions. I examine the effect of litter quality (type= needle vs. wood; decay stage= freshly-fallen vs. well-decomposed) on DOM chemistry. I report the chemistry of O-horizon leachate and C:N of SOM under forest floors where litter input had been manipulated (field study) and chemistry of extracts derived from various sources (laboratory study), and discuss the effects of litter quality on DOM and SOM.

Chapter 3 covers the third question. I examine the changes in DOM composition with soil depth in the field to evaluate differences in the affinity of specific DOM to the mineral soil (field study). I also examine DOM sorption in a laboratory incubation experiment, and discuss chemical controls on DOM sorption.

# CHAPTER 2

# THE EFFECTS OF LITTER QUALITY ON DISSOLVED ORGANIC MATTER (DOM) AND DOM DYNAMICS IN A CONIFEROUS OLD-GROWTH FOREST SOIL IN PACIFIC NORTHWEST

Yuriko Yano, Kate Lajtha, Phil Sollins, and Bruce A. Caldwell

## ABSTRACT

Dissolved organic matter (DOM) plays an important role in SOM formation in temperate rain forests, yet we lack the basic understanding of what controls immobilization and mobilization of DOM in soil. Abiotic sorption and biotic immobilization are the two major processes that remove DOM from soil solution and stabilize it into SOM, and the chemical properties of DOM are believed to affect these biotic and abiotic processes. I examined chemical controls on DOM removal in the mineral soil of a Douglas-fir old-growth forest at the long-term DIRT (Detritus Input and Removal Treatments) study site at the H. J. Andrews Experimental Forest, Oregon, USA. An ongoing manipulation of litter inputs (Double Litter=doubled annual leaf litter inputs; Double Wood=doubled woody litter inputs; and Control= normal litter inputs) at this site provided an opportunity to study the effects of litter type on DOM chemistry, DOM removal in the soil, and soil C and N. A parallel laboratory study was conducted to examine the effect of litter type on DOM chemistry.

Newly harvested root litter tended to produce more DON per weight than other types, suggesting that fine root litter could be the largest DON source in this forest ecosystem, if reallocation of nutrients in senescing fine roots does not retrieve N sufficiently.

The degree of litter decomposition, from newly fallen leaf (Oi) or wood (Class 1) to highly decomposed leaf (Oa) or wood (Class 5), strongly influenced the chemical composition of the extracts. The hydrophobic acid fraction (HoA) increased (from 27 to 47% and from 16 to 62% of total DOC for leaf and wood litter, respectively) and the hydrophilic neutral fraction (HiN) decreased (from 30 to 5% and from 36 to 13% of total DOC for leaf and wood litter, respectively) from the extracts of newly fallen to well-decomposed DOC sources. Contrary to the laboratory results, the composition of the O-horizon leachate was similar among Double Litter, Double Wood and Control treatments after 4 years of litter manipulation, in spite of significant differences in total C, total N and C:N ratios of the O-horizon material. Possibly, microbial degradation of O-horizon material prior to leaching decreased differences in DOM chemistry under

different DIRT treatments. An alternative explanation is a greater contribution from native O-horizon material to DOM rather than from newly added litter.

The concentrations of DOC and DON in the outgoing water were constant regardless of the treatment, season, and the concentrations of incoming water for both shallow (0-30 cm) and deep (30-100 cm) soil layers. In other words, the concentrations of DOM in incoming water and outgoing water were positively correlated with a slope of ~1.0 and negative intercept, suggesting constant net release of both DOC and DON from the soil layers independent of net removal (sorption). The intercepts for the shallow soil layer were more negative than those of the deeper layer for both DOC and DON, suggesting larger net release of DOM from the shallow than from the deeper soil layer. Little influence of season and soil depth on regression slope suggests that most DOM removal was due to abiotic sorption.

## INTRODUCTION

Soil organic matter (SOM) is the largest organic C pool in the biosphere and accounts for virtually the entire ecosystem N reserve (Schlesinger 1997). Thus, the balance between gains and losses of SOM is an important control on atmospheric $CO_2$ and N available to plants via mineralization. As significant amount of C and N enters mineral soils as DOM, the retention of this DOM in soil is an important process for SOM formation, and thus C and N dynamics. Two major mechanisms have been hypothesized for DOM retention in mineral soils: abiotic sorption and biotic immobilization (McDowell and Likens 1988; Qualls and Haines 1992). The importance of sorption in DOM removal is, however, still under debate due to the difficulty in separating abiotic from biotic processes (Kalbitz et al. 2000).

In forest ecosystems DOM concentration is generally highest immediately below the O horizon, and most of the DOM is removed as the solution percolates through mineral soil (Solinger et al. 2001; McDowell and Likens 1988). Abiotic sorption is often considered to be more important than biotic immobilization in removing DOM from soil water, and is greatly affected by soil mineralogy (i.e., clay type, surface area; Davis 1982; Nambu and Yonebayashi 2000). McDowell and Likens (1988) reported that the concentration of carbohydrate, which was assumed to be a labile C, was very small in the soil solution (~4 -6% of total DOC). Similarly, using a laboratory batch incubation of soil solution, Qualls and Haines (1992) found that a fraction of rapidly degradable DOC in Oa horizon solution was too small (~15%) to explain a 100-fold reduction in DOC between the O and B horizons. However, in a different forest ecosystem, rapidly degradable DOC, measured by a flow-through bioreactor, was 10 - 40% of total DOC (Yano et al. 2000), which suggests that abiotic sorption may not be the overwhelmingly dominant mechanism for the removal of DOM in some forest ecosystems.

The chemical composition of DOM affects its sorption potential. For example, aromatic-C is more strongly sorbed to Al- and Fe-hydrous oxides than is alkyl-C (Kaiser et al. 1997). Several techniques have been proposed to separate DOM into fractions related to functionally important characteristics such as biodegradability and sorption. One commonly used technique separates DOM into hydrophobic and hydrophilic fractions by its affinity to different types of resins (Leenheer 1981; Qualls and Haines 1991). It is well documented that hydrophobic DOM has a stronger affinity to mineral soil particles than does hydrophilic DOM (Dai et al. 1996; Kaiser and Zech 1998a, b).

DOM chemistry may be determined by litter type. Generally, leaves and fine roots contain more nutrients (e.g., N and P) than wood (Aber and Melillo 1991). Recent $^{13}$C-NMR analyses have shown that leaves are richer in alkyl C than wood and fine roots (Zech et al. 1997). Correspondingly, the contribution of aromatic C to total OM is greater in leaves (Kögel-Knabner 1997). Decomposition generally increases proportions of alkyl C and decreases O-alkyl C (Kögel-Knabner 1997). Because the decomposition rate of plant litter is closely related to its chemical composition (Melillo et al. 1982), and microbial decomposition controls DOM production in O horizon soil (Gödde et al. 1996), differences in litter type as well as the degree of decomposition may produce DOM with different chemical characteristics. At present, however, it is still not clear what controls DOM chemistry, and we lack the understanding of links between the quality of DOM sources (i.e., different tissue types in different decomposition stages) and DOM chemistry, DOM sorption, and consequently, the dynamics of DOM.

The primary objectives of this field and lab study were to understand the chemistry and behavior of DOM originating from litter of different types (i.e., leaves vs. wood vs. fine roots) and stages of decomposition. The ongoing long-term DIRT (Detritus Input and Removal Treatments) study site established in an old-growth stand at the H. J. Andrews Experimental Forest in the Cascade mountains in western Oregon provided an opportunity to investigate links between litter type and DOM chemistry in the field. In the lab study, I examined the influence of tissue type and degree of decomposition on the chemistry of water-extractable DOM.

## METHODS

### Study sites

Plant litter inputs have been manipulated at the DIRT plots (44°15′N, 122°10′W, 726 m elevation) since 1997. The mean annual temperature at the headquarters site at the Andrews Experimental Forest is 7.9 °C and the mean annual precipitation is 237cm yr$^{-1}$, mostly rain. Over 70% of the precipitation occurs during a "wet season", between November and March on average (Sollins et al. 1980). The transitions between a "wet season" and "dry season" are generally clear, although the timing can vary from year to year by ~2 months. During my study period, I defined the wet seasons to be Oct.-May of 1999-2000 and Nov.-May 2000-2001, periods when soil water collection was possible. N deposition to this area is ~2 kg N ha$^{-1}$yr$^{-1}$ (Sollins et al. 1980), close to levels in other pristine environments where anthropogenic N deposition is minimal (Hedin et al. 1995). The DIRT site was established in an undisturbed old-growth Douglas-fir (*Pseudotsuga menziesii*) stand. Other plant species found at the site include western hemlock (*Tsuga heterophylla*), vine maple (*Acer circinatum*), western red cedar (*Thuja plicata*), and Oregon grape (*Berberis nervosa*) and mosses covering the forest floor. Soils are derived from volcanic parent materials and have been classified as Humic Dystrudepts. The soils have strong Andic tendencies: high amorphous Al hydroxide content (oxalate-extractable Al= 1.1 %) and a pH in NaF near 11 (data not shown). Prior to the establishment of the DIRT plots, the soils had a thin O horizon (2-5 cm) that was difficult to separate from the moss layer, lying on a 10-20 cm thick A horizon with abundant fine roots, over a 20-30 cm thick B horizon with less fine root biomass.

### Field litter input manipulation

In 1997 six litter input treatments, replicated 3 times, were randomly assigned to the plots. Plots measure 10 m x 15 m, although a few are 10 m x 10 m where there was no space for a larger plot. One plot was approximately 125% larger than the standard size because physical obstacles (e.g., a large recent mound from uprooted tree) blocked access to undisturbed soil. In this study, only three of the six treatments were used (Table 2-1).

Because of great heterogeneity in the amount of fallen logs and mosses accompanying them, most logs lying on the ground and moss layer were removed from plots prior to the start of the treatment to standardize initial conditions. The litter on No Litter plots was excluded with 1 mm-mesh screens, transferred to Double Litter plots, and spread evenly 3-4 times per year: once at the end of the dry season, twice or more during the wet season in normal years (November – March), and once at the beginning of the dry season. Any large branches and stems that fell on screens were discarded. Between 1997 and 2001, the total mass of aboveground fine litter added to Double Litter plots was 5760 kg ha$^{-1}$ yr$^{-1}$. To double the mass of woody debris in the forest floor of Double Wood plots, extremely decomposed woody debris (Class 5) from an adjacent area was added in the three summers between 1997 and 1999, and the chips of large pieces of newly fallen Douglas-fir (Class 1) in the summers between 1998 and 2000. Total mass of Class 5 debris added to the plots was 21.6 Mg ha$^{-1}$ and Class 1 debris 86.4 Mg ha$^{-1}$.

**Table 2-1.** Litter input treatments at the DIRT study site. Treatments used in this study are shown in bold letters.

| Treatment | Method |
|---|---|
| **Control** | **Normal litter inputs** |
| **Double Litter** | **Doubled annual leaf (needle) and fine litter inputs** |
| **Double Wood** | **Doubled existing woody debris in the forest floor** |
| No Litter | Aboveground litter (leaf and fine litter) inputs excluded by screening |
| No Roots | Belowground litter (fine root) inputs excluded by trenching |
| No Inputs | Above- and belowground litter inputs excluded by screening and trenching |

### Soil water collection and treatment

Five tension lysimeters (Prenart) were installed in each plot according to the method described by Lajtha et al. (1999), three at 30 cm and two at 100 cm depth in the mineral soil. Water percolating through the soil column was collected approximately monthly during 2 wet seasons, Oct. 1999 - May 2000 and Nov. 2000 - May 2001, except for a 2-3 month period when access to the plots was blocked by deep snow. One zero-tension lysimeter was installed at the bottom of the O horizon (0 cm) in each treatment plot to collect O-horizon leachate, and soil water was collected 3 times at 2-3 month intervals during the 2000-2001 wet season. To minimize biological and chemical alteration of solution, all samples were retrieved within 72 hours after the application of tension (for tension lysimeters) or the beginning of water collection (for zero-tension lysimeters). All samples were transferred on ice to Oregon State University where volume was measured and samples were filtered thorough combusted Whatman GF/F glass fiber filers, and stored frozen until analysis for dissolved organic carbon and nitrogen (DOC and DON) and chemical fractionation.

## Soil sampling

Plots were divided into 5 m x 5 m subplots prior to soil sampling. O-horizon material was collected from random points within each subplot in fall 2001and bulked into a single composite sample per plot. A-horizon soil (0-10 cm) was collected in summer 2001 with an Oakfield corer and bulked into a single composite sample per subplot to yield a total of 4-6 samples per plot. Samples were immediately transported to Oregon State University and all O-horizon material and a portion of the A-horizon soil were air-dried. Dried soils were then ground to pass a 100-mesh screen for analysis of total C and N.

## Collection of plant litter

### Needle

To obtain Douglas-fir needle litter along a gradient of decay, I collected O-horizon material from three young Douglas-fir stands where the O horizon was thick enough (~4 cm) to be separated by decay stage, had no moss cover, and contained litter derived mostly from Douglas-fir (85.5% on average by weight). The material was then gently separated into Oi, Oe, and Oa layers with a knife. Oi horizon is composed of freshly fallen needles with minimal decay, Oe horizon contains partially decomposed needles, and Oa horizon is composed of well-decomposed material whose origin is unrecognizable.

### Wood

I collected wood from three newly-fallen (Class 1) and three well-decomposed (Class 5) Douglas-fir logs from the McDonald-Dunn Research Forest, located about 90 km northwest of the Andrews Experimental Forest. I used the classification method for logs, described in Sollins (1982), which is widely used in the Pacific Northwest of U.S.A: Class 1 logs are freshly-fallen, bark and all wood sound, and current-year twigs still attached; Class 5 logs are extremely decomposed such that sapwood and bark are absent and wood is mainly fragmented and cannot be lifted intact. I assumed that the class 5 logs that I used were Douglas-fir based on the nature of the logs and the history of the stand, because identification of Class 5 logs to species is difficult. Class 1 logs were further separated into bark, sapwood, and heartwood.

### Fine Roots

Douglas-fir seedlings were grown on a mixture of the Andrews Experimental Forest mineral soil and commercial silica sand (soil : silica sand = 1 : 1) for 1 - 1.5 years. The mixture instead of native soil was used for the ease of fine-root harvest. The seedlings were then harvested by gentle washing in DI water. Roots were submerged in water no longer than 30 minutes. All roots had morphological features of mycorrhizal symbioses. The distribution of root-diameter size was; 72.7%, 20.1%, and 6.76% by weight for <1.0 mm, 1.0-2.0mm, and >2.0 mm, respectively. All roots harvested were pooled to a single composite sample because of low total mass.

### Lab extraction of plant litter

Subsamples of all litter (Oi, Oe, Oa, Class 1 and 5, and root) were air-dried and ground to pass a 30-mesh screen. The ground litter was then extracted in DI water with solid-to-water ratios of 1: 40, 1: 25, and 1: 25 for leaf, wood, and root litter, respectively. The extraction was conducted in a shaker at 100 rpm at 22°C for 48-68 hours followed by centrifugation at 7000 rpm for 15 minutes. The supernatant of each extract was then filtered through Whatman GF/F glass fiber filers, and stored frozen. The frozen samples were used for the determination of initial concentrations of DOC and DON as well as for further chemical analysis. A portion of the ground litter was further ground to pass a 100-mesh screen for the analysis of total C and N.

## Chemical analysis

### DOC and DON

All soil water collected in lysimeters and plant litter was analyzed for DOC with high temperature platinum-catalyzed combustion (Shimadzu TOC-5000A HTCO carbon analyzer).

Nitrate-N was measured using the hydrazine sulfate reduction method and $NH_4$-N was determined using the Berthelot reaction method using a Scientific Instruments Autoanalyzer. Total dissolved N (TDN) was measured using Koroleff's (1983) persulfate digestion procedure, followed by $NO_3^-$ analysis. Because $NO_2$-N in soil water or solutions in an aerobic environment is negligible (Qualls et al. 1991, Currie et al. 1996), DON was calculated as:

$$[DON] = [TDN] - [NO_3^- \text{-N}] - [NH_4\text{-N}]$$

Because DON was calculated by difference, values were sometimes slightly negative, in which case a value of 0 mg $L^{-1}$ was assigned.

### Chemical fractionation

Soil water collected in lysimeters and extracts of plant litter were fractionated into 6 functional fractions by a method modified from Qualls and Haines (1991) and Leenheer (1981). In brief, the method fractionates DOM by its affinity to 3 different types of resins (hydrophobic, cation-exchange, and anion-exchange). The weak hydrophobic acid fraction (Phenol) includes small phenolic compounds (e.g., tannin and flavonoids). The strong hydrophobic acids (HoA) are mainly microbially altered plant-derived material and have high content of aromatic C and include larger molecular size humic substances and humic-bound amino acids and carbohydrates. The hydrophobic neutral fraction (HoN) is less microbially altered plant-derived material and contains waxes, fatty acids, and microbial lipids. The hydrophilic acid fraction (HiA) contains partly microbially synthesized and partly plant-derived material that is lower molecular size humic-like substances with high carboxyl-to-C ratios. The hydrophilic neutral fraction (HiN) contains non-humic-bound carbohydrates, mainly of microbial origin, and may contain simple sugars (e.g., hexose, deoxysugars). The hydrophobic and hydrophilic base fractions contain free amino acids, peptides and proteins. Because the proportions of hydrophobic base fraction were very small for all soil solutions and litter extracts tested in this study (≤2 % of total DOC), both hydrophobic and hydrophilic bases were combined and reported as a base fraction (Bases).

### Total C and N

Total C and N in the O-horizon material, A-horizon soil, and plant litter were determined by Micro-Dumas combustion analysis at the Stable Isotope/Soil Biology Laboratory of the University of Georgia, Georgia, USA.

### Data Analysis

All measurements of DOC and DON for soil water were grouped by plot (*n*=3) within each year. Repeated measures analysis was used to determine any effects of treatment, time, and depth on DOC, DON, and DOC:DON ratio. Chemical fraction composition was determined for the O-horizon leachate and litter extracts. A paired t-test was used to detect the effect of litter type or litter-input treatment on the composition of chemical fractions. A one-way analysis of variance (ANOVA) was used to look for treatment effects on the chemistry of bulk soils, litter, and DOM. Values were natural-log or square-root transformed to meet appropriate normality prior to the analysis, followed by back-transformation to obtain least square means and 95% confidence limits. The SAS System (SAS Institute Inc. 1999) was used for all statistical analyses.

Extraction and all chemical analyses for Class 1 wood were done on bark, sap wood, and heart wood, separately. Values for whole wood were then back calculated based on %tissue volume of whole wood for Douglas-Fir with mean log diameter of 52 cm (bark=12%, sap wood= 30%, and heart wood=58%) (Harmon 1992).

Net removal (sorption) of DOM in the soil column was determined for each sampling event as the difference in DOM concentration between the two soil depths (0-30 cm and 30-100 cm), after correction for the effect of evapotranspiration (ET). Water loss via ET in the mineral soil between 0 and 100 cm was assumed to be 21% of O-horizon leachate based on a previous study at the near by site (Sollins et al. 1980). I assumed that 61% of the total loss occurred within the first 0-30 cm soil layer, based on the distribution of fine (diameter < 1 mm) and small roots (1 mm < diameter < 5 mm) within the mineral soil of a stand nearby (Santantonio and Hermann 1985).

## RESULTS

### Chemical properties of litter and litter extracts

#### C and N of litter and extracts

The litter of various types (needle, wood, and root) and stages of decomposition (from newly fallen to highly decomposed) showed large differences in total C, N, and C:N ratio (Table 2-2). The concentration of C in the needle litter decreased with the degree of decomposition (i.e., C content of Oa is ~56% that of Oi litter). The pattern of N content was different. Regardless of the degree of decomposition, wood litter had ~10-

40% lower total N than did needle or root litter and thus, the C:N of wood litter was higher (189-527) than that of the needle or root litter (30.6-66.9). For both needle and wood litter, C:N ratios of well-decomposed litter were less than half of the values for freshly-fallen litter.

The ratio of extractable-organic C to bulk C decreased with the degree of litter decomposition. On average, percentage of extractable C for freshly-fallen litter (Oi, Class 1, and fine root) was over 10 times greater than for well-decomposed liter (Oa and Class 5) (Table 2-3). Fine root litter showed the largest proportion of extractable-organic C among all litter sources (7.6 %). The ratio of extractable-organic N to bulk N was, on the other hand, relatively small and did not vary greatly across the litter type (Table 2-3). Fine roots produced more DOC per gram litter material than did needle or wood litter. In addition, the DOC:DON ratio of the root extract (249) was 1/3 to 1/7 that of needle (840) or wood litter (1719). Although these differences were not significant, the lack of significance is perhaps due to no replication for the root extract (n=1). Along the decay gradient within each litter type, DOC:DON ratio generally followed the pattern of C:N of the litter from which it was extracted (Table 2-2). When the DOM source was freshly-fallen litter, the C:N ratio increased from the source to the extract (needle: from 66.9 to 840, wood: from 527 to 1719, and root: from 59.4 to 249). On the other hand, for well-decomposed litter, C:N ratio decreased from the source to extract (needle: from 30.6 to 27.7, wood: from 189 to 65.5).

## Composition of chemical fractions

Differences in litter type produced differences in the chemistry of lab-extracted DOM. HiN was the largest fraction for new wood and root extracts (36-37%, Fig. 2-1-A) while HoA, HiN, and HiA fractions were equally abundant for needle extracts (27, 30, and 25% of total DOC, respectively). The degree of decay appeared to have a stronger influence on the composition of DOC than did differences in litter type.

**Table 2-2.** Total C, N, and C:N ratio of litter and DOM water-extracted from the litter. Numbers in parentheses indicate lower and upper 95% confidence limits. Bold letters refer to significant differences within each variable across all litter types. No significant differences were found for %N or DON either among litter types or across decay stages. All measurements for litter and extracts were n=3, except for the root extract (n=1). Statistics are only shown for columns where significant difference were found.

| | | Bulk litter | | | Water-extractable DOM | | |
|---|---|---|---|---|---|---|---|
| | | C | N | C:N | DOC | DON | DOC:DON |
| Litter type | Decay stage | % | | | mg/L (water:litter=40:1) | | |
| Needle | Oi | 47.3 **b** | 0.71 | 66.9 **b** | 384.7 **a** | 0.16 | 840 **ab** |
| | | (42.5, 52.0) | (0.50, 0.92) | (56.8, 78.8) | (156.9, 943.3) | (0.03, 0.98) | (148, 4755) |
| | Oe | 39.0 **ab** | 0.99 | 39.6 **ab** | 146.9 **ab** | 1.09 | 173 **ab** |
| | | (34.3, 43.7) | (0.78, 1.20) | (33.6, 46.6) | (59.9, 360) | (0.21, 2.65) | (41.9, 711) |
| | Oa | 26.8 **a** | 0.90 | 30.6 **ab** | 37.6 **b** | 0.43 | 27.7 **b** |
| | | (22.1, 31.5) | (0.69, 1.11) | (26.0, 36.0) | (15.3, 92.2) | (0.01, 1.54) | (4.9, 157) |
| Wood | Class 1 | 48.6 **a** | 0.09 | 527 **a** | 384.6 **a** | 0.30 | 1719 **a** |
| | | (43.8, 53.3) | (-0.12, 0.30) | (448, 621) | (156.8, 943.0) | (0.00, 1.29) | (418, 7078) |
| | Class 5 | 53.1 **b** | 0.28 | 189 **b** | 42.6 **b** | 0.73 | 65.5 **b** |
| | | (48.4, 57.8) | (0.07, 0.49) | (160, 222) | (17.4, 104.4) | (0.07, 2.07) | (15.9, 270) |
| Fine root | Newly harvested | 44.2 **a** | 0.74 | 59.4 **ab** | 843.1 **a** | 3.39 | 249 **ab** |

**Table 2-3.** The percentage of total C and N that is water-extractable for different litter types and decay stages. Numbers in parentheses indicate lower and upper 95% confidence limits. No significance was found across litter types and decay stages. All measurements for litter and extracts were n=3, except for the root extract (n=1).

| Litter type | Decay stages | Water-extractable C (%) | Water-extractable N (%) |
|---|---|---|---|
| Needle | Oi | 3.7 | 0.10 |
| | | (2.0, 6.0) | (0.07, 0.77) |
| | Oe | 1.6 | 0.47 |
| | | (0.6, 3.2) | (0.01, 1.56) |
| | Oa | 0.6 | 0.25 |
| | | (0.1, 1.7) | (0.01, 1.13) |
| Wood | Class 1 | 3.2 | 0.88 |
| | | (1.6, 5.3) | (0.14, 2.26) |
| | Class 5 | 0.4 | 0.63 |
| | | (0.0, 1.3) | (0.05, 1.84) |
| Fine root | Newly harvested | 7.6 | 1.14 |

For example, percent HiN differed more between Oi and Oa (*P*=0.001) than between Oi and Class 1 extracts (*P*=0.311). For both well-decomposed needle and wood litter, percent HoA increased and it became the largest fraction of all with increasing degree of decomposition (47 and 61% for Oa and Class 5, respectively). At the same time, the percent HiN decreased.

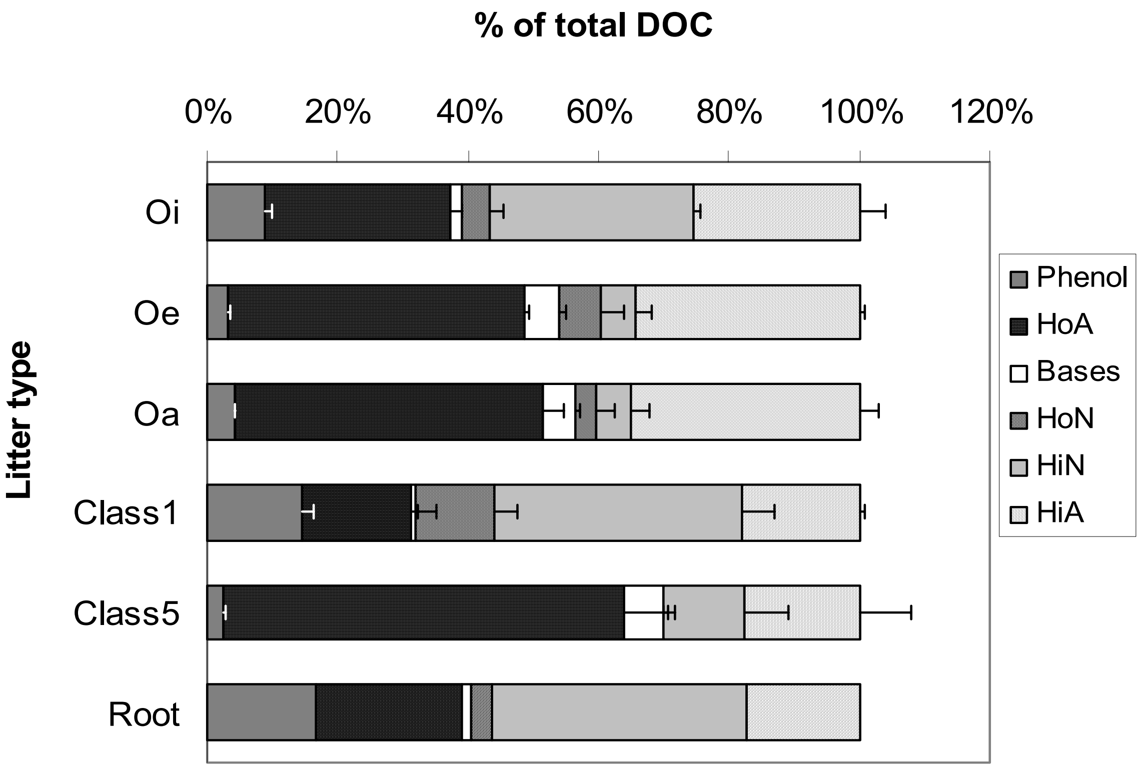


**Fig. 2-1.** Chemical composition of extracts for litter of different type/ decomposition gradient. Composition was expressed as the percent fraction of total water-extractable DOC.

## Chemical properties of O-horizon and soil

Four years of litter-input manipulation altered the chemistry of O-horizon material. The Double Wood treatment had the highest %C and the lowest %N. Consequently, the C:N of O-horizon under the Double Wood treatment was about twice that under other treatments (Table 2-4). Treatments had no significant effect on total C and N of the A-horizon soil.

## Chemistry of DOM – field collection

### DOC

DOC concentration under Double Wood tended to be more than under Control or Double Litter for 30-cm soil water for both years. Although this trend was not significant, mean DOC for Double Wood was 1.8 - 2.6 and 2.0 - 4.1 times that of Control or Double Litter, respectively (Table 2-5). Any suggestion of a trend disappeared by 100 cm depth. The concentration of DOC decreased significantly with increasing soil depth under all treatments for both years, indicating net DOC removal in the soil column (Table 2-5). The ranges of mean DOC concentration for 0-cm (O-horizon leachate), 30-cm and 100-cm soil water were 40 – 52, 2.9 - 12.3, and 2.1 - 3.9 mg-C/L, respectively. Approximately 88% of total DOC in the O-horizon leachate was removed in the upper 30 cm (in the second year). There was no significant difference in DOC between the two years regardless of treatment or depth.

**Table 2-4.** Least square means of total C, N, and C:N ratio of O- and A-horizon soils after four years of litter-input manipulation on the DIRT study site at the Andrews Experimental Forest. Values enclosed in the parenthesis are lower and higher 95% confidence limits. Bold letters refer to significant differences within each column.

| | O-horizon | | | A-horizon | | |
|---|---|---|---|---|---|---|
| | Total C | Total N | C:N | Total C | Total N | C:N |
| Treatment | % | | | % | | |
| Double Litter | 39.8 **ab** | 0.92 **a** | 43.9 **a** | 6.8 **a** | 0.23 **a** | 28.4 **a** |
| | (36.5, 43.1) | (0.74, 1.09) | (36.6, 52.5) | (3.4, 10.2) | (0.17, 0.29) | (20.2, 39.8) |
| Double Wood | 44.8 **b** | 0.60 **b** | 75.9 **b** | 7.2 **a** | 0.22 **a** | 32.1 **a** |
| | (41.5, 48.1) | (0.42, 0.77) | (63.4, 90.9) | (3.9, 10.6) | (0.16, 0.27) | (22.8, 45.0) |
| Control | 38.4 **a** | 0.97 **a** | 39.4 **a** | 6.2 **a** | 0.20 **a** | 29.6 **a** |
| | (35.1, 41.8) | (0.80, 1.15) | (32.9, 47.2) | (2.8, 9.6) | (0.15, 0.26) | (21.1, 41.6) |

### DON

Contrary to DOC, DON concentration differed among litter input treatments during the first year, although not during the second year. During the first year, the mean DON concentration under Double Wood was higher than under the other 2 treatments, Double Litter and Control, although the difference was significant only between Double Litter and Double Wood treatments ($P$=0.032, Table 2-5). DON concentration in the soil water decreased from the first to second year for both 30 ($P$=0.006) and 100 cm ($P$=0.010) depths. An effect of depth on DON was found only in the second year. Similar to the pattern observed for DOC, most (~96 % of) DON was removed in the top 30 cm soil layer.

### DOC:DON

DOC:DON ratio of the soil water was not affected by the litter input treatments during either year. Mean DOC:DON of the soil water at 30 cm increased from the first to second year ($P$=0.049) due to lower mean DON values the second year. Generally, DOC:DON ratio decreased with increasing soil depth (Table 2-5-A, B), but this was significant only in the second year.

### Chemical fractionations

The chemical composition of the O-horizon leachate collected in the second year was similar among treatments. Major components of the DOC were HoA (60-69% of total), HiA (25-46% of total), and Bases (2-4% of total); other fractions accounted for < 2% of total DOC (Fig. 2-2). No treatment effect was found for the three major fractions (i.e., HoA, HiA, and Bases) (paired t-test, $P$>0.05).

**Table 2-5.** Least square means of DOC, DON, and DOC:DON ratio of the soil water collected from DIRT site at the Andrews Experimental Forest during (A) the first and (B) second year. Note that soil water from 0 cm was not collected during the first year. Numbers in parentheses are lower and upper 95% confident limits. Bold letters indicate a significant treatment effect within each column. Statistics are only shown for columns where significant difference were found.

**A.** Year 1999-2000

| | 30 cm | | | 100 cm | | |
|---|---|---|---|---|---|---|
| | DOC | DON | DOC:DON | DOC | DON | DOC:DON |
| Treatment | (mg/L) | | | (mg/L) | | |
| Double Litter | 2.9 | 0.09 **a** | 31.1 | 1.1 | 0.12 | 9.5 |
| | (1.2, 7.1) | (0.04, 0.18) | (8.5, 114) | (0.7, 1.6) | (0.04, 0.38) | (1.9, 46.5) |
| Double Wood | 7.5 | 0.26 **b** | 23.6 | 1.4 | 0.25 | 7.8 |
| | (3.1, 18.4) | (0.12, 0.55) | (6.4, 86.2) | (0.9, 2.0) | (0.08, 0.77) | (1.3, 48.9) |
| Control | 4.1 | 0.14 **ab** | 58.2 | 0.9 | 0.08 | 28.5 |
| | (1.7, 10.1) | (0.07, 0.30) | (15.9, 213) | (0.6, 1.3) | (0.03, 0.25) | (5.8, 140) |

**Table 2-5.** (Continued)

**B.** Year 2000-2001

| | 0 cm | | | 30 cm | | | 100 cm | | |
|---|---|---|---|---|---|---|---|---|---|
| | DOC | DON | DOC:DON | DOC | DON | DOC:DON | DOC | DON | DOC:DON |
| Treatment | (mg/L) | | | (mg/L) | | | (mg/L) | | |
| Double | 51.1 | 0.75 | 68.6 | 3.0 | 0.05 | 69. 7 | 0.9 | 0.02 | 23.8 |
| Litter | (19.4, 134.3) | (0.30, 1.93) | (10.9, 431) | (1.1, 8.2) | (0.02, 0.13) | (27.3, 178) | (0.5, 1.6) | (0.01, 0.04) | (10.3, 54.8) |
| Double | 46.1 | 1.29 | 37.0 | 12.3 | 0.07 | 116 | 1.2 | 0.05 | 24.3 |
| Wood | (17.6, 121.3) | (0.51, 3.31) | (5.9, 233) | (4.5, 33.8) | (0.03, 0.16) | (45.6, 297) | (0.7, 2.1) | (0.02, 0.10) | (10.5, 55.9) |
| Control | 39.8 | 0.44 | 102 | 6.1 | 0.06 | 64.6 | 0.7 | 0.04 | 15.8 |
| | (15.1, 104.5) | (0.17, 1.12) | (16.2, 641) | (2.2, 16.9) | (0.03, 0.15) | (25.3, 165) | (0.4, 1.3) | (0.02, 0.08) | (6.9, 36.5) |

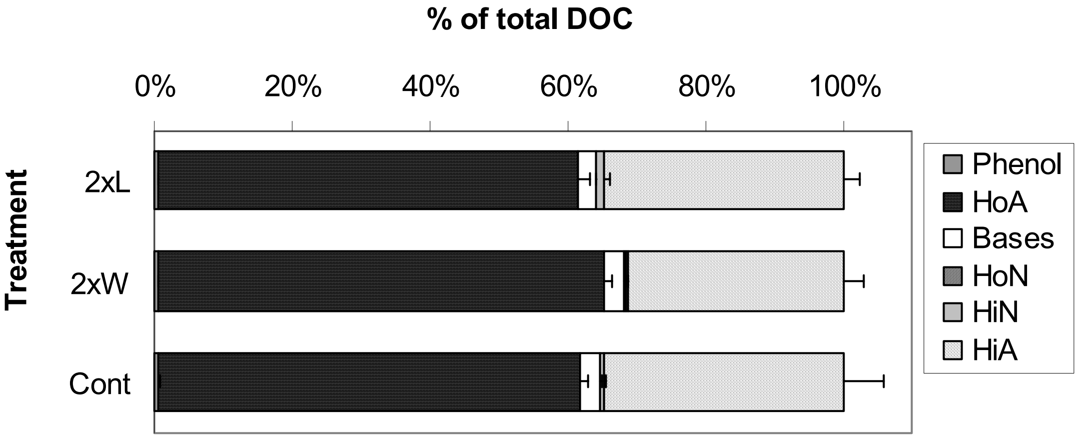


**Fig. 2-2.** Chemical composition of O-horizon leachate collected on DIRT site, HJA during the wet season of 2000-2001.

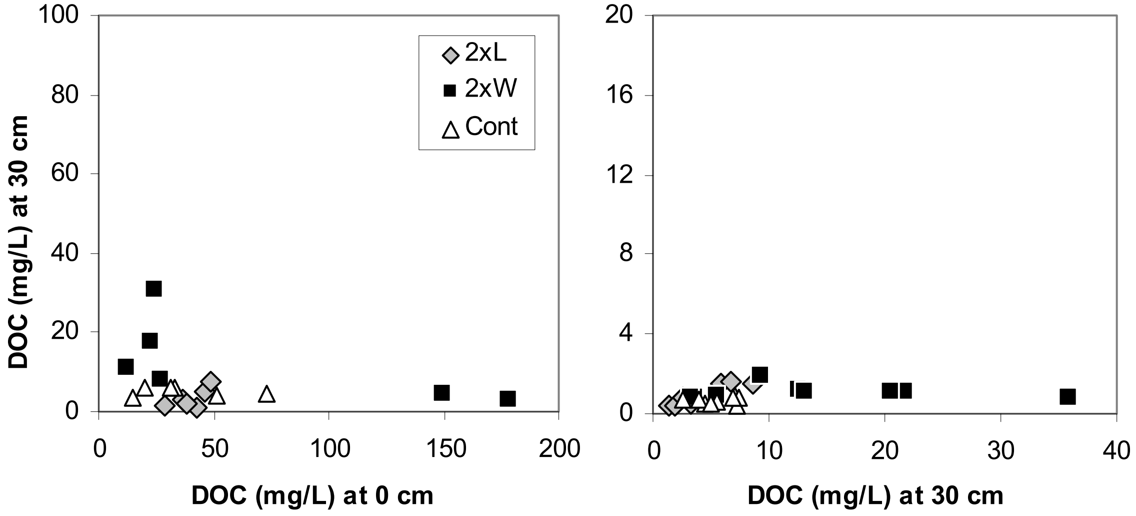
Net removal and release of DOM

The concentrations of DOC and DON in the outgoing water were constant regardless of the treatment, season, and the concentrations of incoming water for both shallow (0-30 cm) and deep (30-100 cm) soil layers (Fig. 2-3). The correlations between DOC in incoming water and net DOC removal on a sampling event basis were very tight (Pearson's R ranged from 0.862 to 0.999) (Table 6, Fig 2-4-A, C). The slopes of regression lines for the correlations were close to 1.0, regardless of the treatment, season, or the depth of the soil layer (Table 2-6, Fig. 2-5). A similar relationship was found for net removal of DON in the same soil layer (Pearson's R ranged from 0.746 to 0.997) with the slope of the regression lines being close to 1.0 (Table 2-6, Fig 2-4-B, D). All intercepts of the regression lines were negative, which indicates a constant net release of DOC or DON from the soil layer, at least at low concentrations in the input water.

**Table 2-6.** Correlations between net DOM removal and DOM concentration in incoming water.

| Season | Soil layer (cm) | Regression line | Pearson's R | *P*-value |
|---|---|---|---|---|
| **DOC** | | | | |
| 1999 warm/dry | 30-100 | $[NR_{DOC}] = 1.01 \times [DOC_{in}] - 1.44$ | 0.862 | <0.001 |
| 1999-2000 cold/wet | 30-100 | $[NR_{DOC}] = 0.97 \times [DOC_{in}] - 0.90$ | 0.993 | <0.001 |
| 2000-2001 cold/wet | 0-30 | $[NR_{DOC}] = 1.04 \times [DOC_{in}] - 9.11$ | 0.989 | <0.001 |
| | 30-100 | $[NR_{DOC}] = 0.98 \times [DOC_{in}] - 0.76$ | 0.999 | <0.001 |
| **DON** | | | | |
| 1999 warm/dry | 30-100 | $[NR_{DON}] = 0.94 \times [DON_{in}] - 0.13$ | 0.894 | 0.001 |
| 1999-2000 cold/wet | 30-100 | $[NR_{DON}] = 0.94 \times [DON_{in}] - 0.14$ | 0.746 | <0.001 |
| 2000-2001 cold/wet | 0-30 | $[NR_{DON}] = 0.98 \times [DON_{in}] - 0.06$ | 0.997 | <0.001 |
| | 30-100 | $[NR_{DON}] = 0.96 \times [DON_{in}] - 0.03$ | 0.901 | <0.001 |

$[NR_{DOC}]$: net removal of DOC, $NR_{DON}$: net removal of DON. Both as (mg/L)
$[DOC_{in}]$: DOC concentration of incoming water to a soil layer as (mg/L).
$[DON_{in}]$: DON concentration of incoming water to a soil layer as (mg/L).


**A.** Soil layer 0-30 cm **B.** Soil layer 30-100 cm

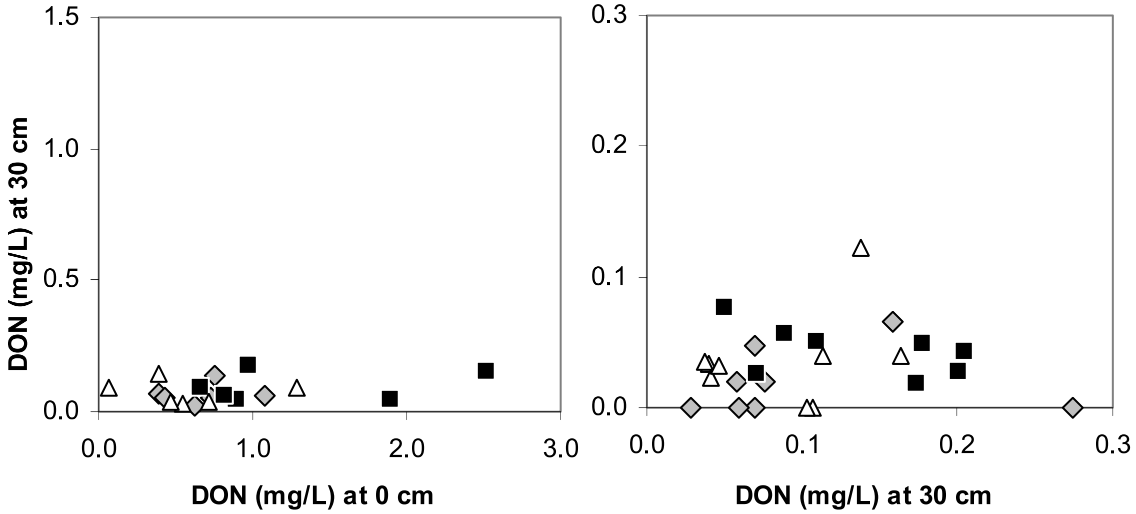

**C.** Soil layer 0-30 cm **D.** Soil layer 30-100 cm

**Fig. 2-3.** Constant release of DOM from two different soil layers. Relationship between DOC concentration in the incoming water and outgoing water for (A) 0-30 cm and (B) 30-100 cm soil layers, and for DON for (C) 0-30 cm and (D) 30-100 cm soil layers. Note the scale changes between graphs. The values of deep-soil DOM were corrected for concentration via evapotranspiration, and the net removal of DOM was calculated as: [shallow soil DOM] - [deep soil DOM]. Concentration factors used were 0.865 for the 0-30 cm soil layer and 0.900 for the 30-100 cm layer.

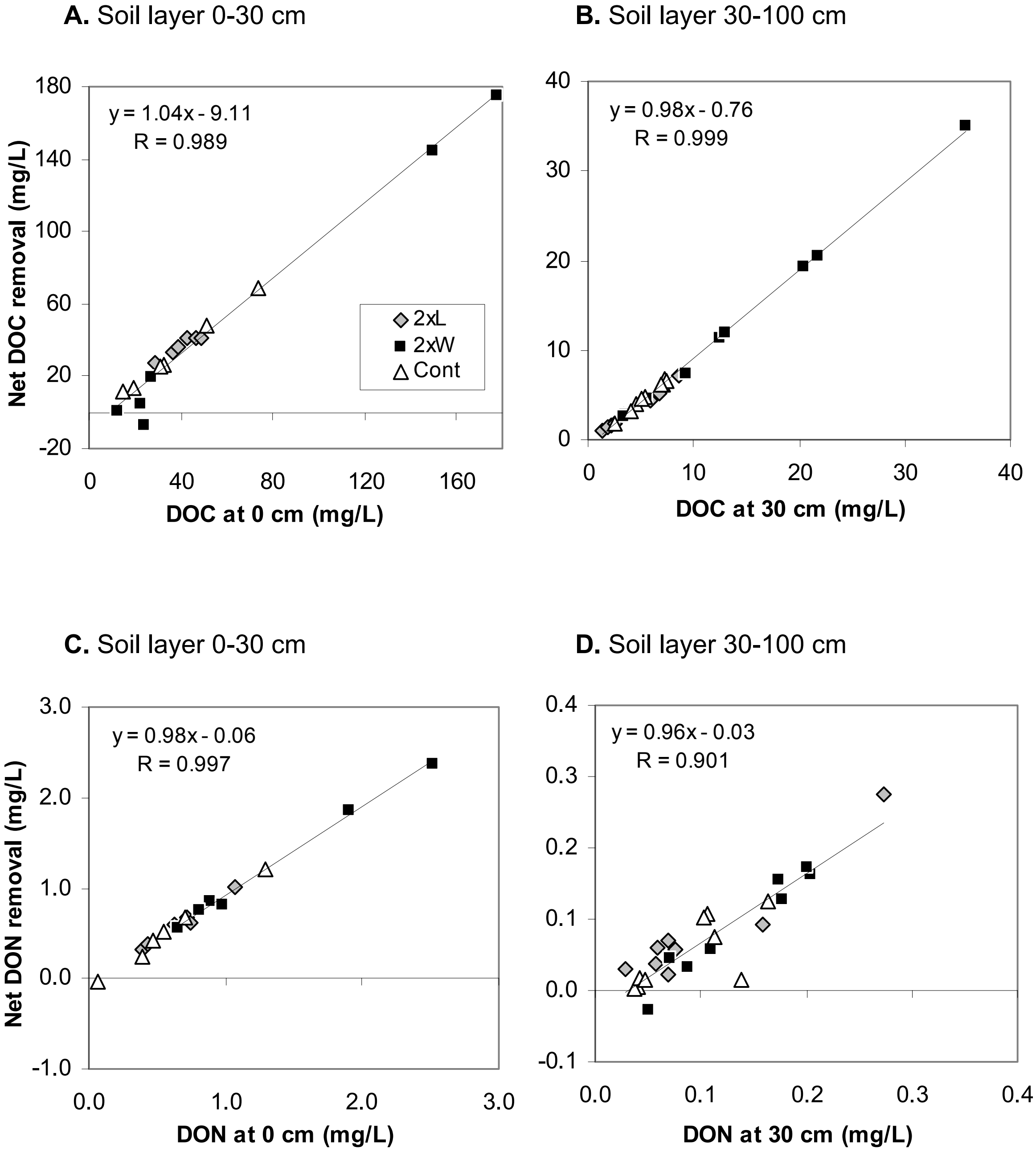


**Fig. 2-4.** Relationships between DOM concentration of the shallow soil water and the net removal of DOM. Only samples collected during the cold season of 2000-2001 water year were shown as examples. (A) and (B) show correlations for DOC and DON in the 0-30 cm soil layer, and (C) and (D) show those for the 30-100 cm soil layer. Note that scale changes between graphs. Values were corrected for water loss via evapotranspiration (see text and Fig. 2-3 caption).

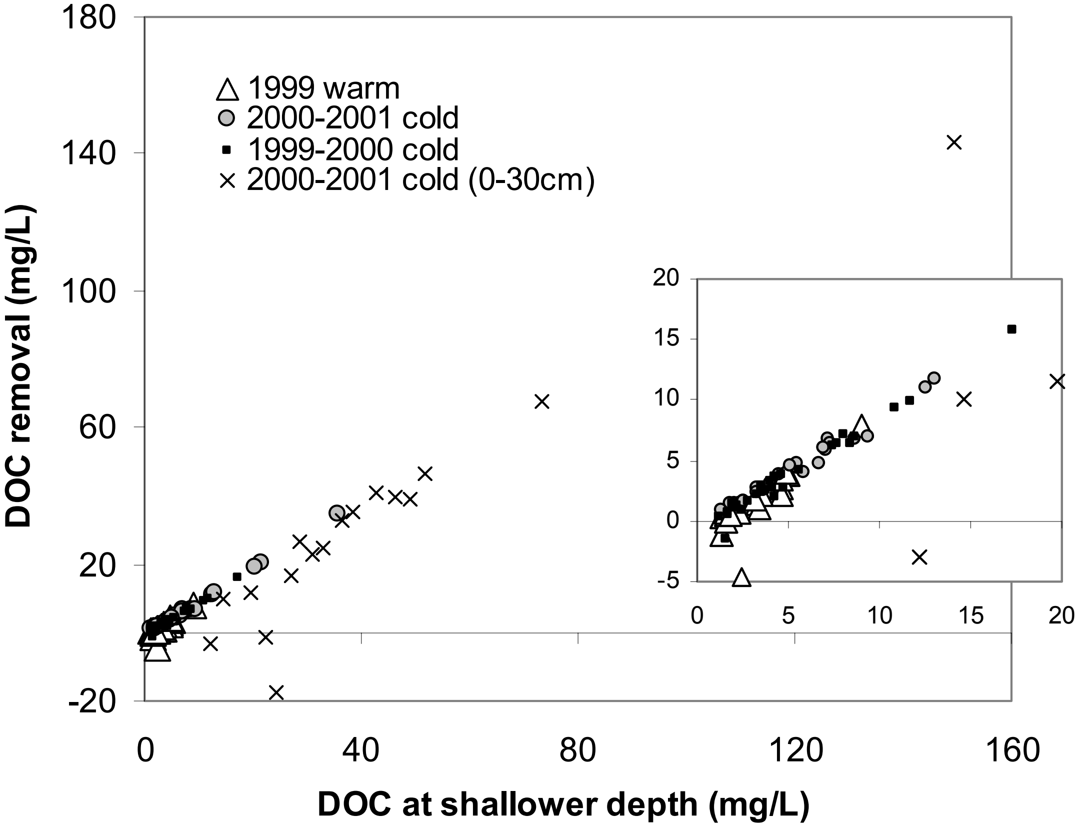


**Fig. 2-5.** Relationships between DOC removal and DOC concentration in the shallow water across season, year, and the depth of soil layer. DOC removal shown is for the soil layer of 30-100 cm, unless noted in the legend. Open triangles represent samples collected in the warm season of 1999 (June - Sept.); others were collected during the cold season (Oct. - May). A region of low DOC concentration is magnified and shown in a smaller graph inside the larger graph.

## DISCUSSION

### Chemical properties of litter and DOM chemistry

The C:N ratio differed by litter type. Sollins et al. (1980) reported organic matter inputs via leaf, wood, and fine root litter in a nearby Douglas-fir ecosystem of 2.3, 7.0, and 3.4 Mg $ha^{-1} \cdot yr^{-1}$. Combined with my finding that root litter produced the most DON per weight (>10 times that of leaf or wood litter, Table 2-2), root litter may be the largest DON source among various litter types in this forest ecosystem. Because I did not determined the amount of DON released from dead roots, it is difficult to conclude that fine roots are the largest DON source in the field conditions. Because plants can reallocate nutrients in roots before they senesce, it is possible that DON that can be released from dead roots in the field may be much smaller than what I observed for newly-harvested fine roots.

C:N ratio decreased with degree of decay as expected from loss of C via respiration during decomposition and/or the loss of C via leaching of DOM. The increases in C:N ratio from bulk litter to extracts suggest that the potentially leachable OM in newly-fallen needle and wood litter has a high C:N ratio. Chlorophyll, phospholipids, simple neutral sugars, and polysaccharides may be in this category. The lower C:N ratio of extracts compared to well-decomposed litter suggests that the C:N of the leachable OM is low. As litter decays, microbial breakdown of originally recalcitrant compounds (e.g., lignin, ligno-cellulose) could produce labile compounds that are prone to physical leaching. Because the C:N ratio of microbes is around 4-10 (Myrold 1998), leachable OM produced via microbial turnover would have a lower C:N ratio than its source. This idea is supported by my results: high percent HoN and HiN, which contain lipids and simple sugars, in the extracts of newly fallen litter, and high percent HoA and HiA in the extracts of well decomposed litter. HoA and HiA are produced via microbial degradation or polymerization of lignin and/or condensation of lignin-derived compounds (Guggenberger et al. 1994). Kaiser and Zech (2000) found a greater percent

Ho and smaller percent Hi in Oa-horizon extract than Oi extract. Their findings were consistent with my results where percent Ho increased (from 40 to 54% for needle, from 40 to 64% for wood extracts) and percent Hi decreased with increasing decomposition of DOM sources (Fig. 2-1).

## Chemical properties of soil

Several factors might explain why four years of litter manipulation altered the chemistry of the O-horizon material, but not the A-horizon soil. First, the lack of differences in DOM chemistry of O-horizon leachate would result in the retention of similar DOM compounds in the mineral soil.

Second, it is unlikely that incoming DOM and plant tissues that were physically mixed into the mineral soil over four years would be significant relative to native SOM. Assuming that 1) the DOC concentration of O-horizon leachate did not change during the period of litter-input treatment (45.7 mg-C/L, averaged for all treatments in Table 2-5), 2) water flux from O-horizon to the mineral soil at DIRT site was 19800 Mg ha$^{-1}$ yr$^{-1}$, as estimated by Sollins et al. (1980) for a nearby watershed, 3) the accumulation of SOM by physical mixing of O-horizon soil was < 0.1 Mg ha$^{-1}$ yr$^{-1}$ (also estimated by Sollins et al. (1980) for the same watershed), thus a possible high end of 'SOM accumulation rate via fine litter' at the DIRT site is 0.1 Mg ha$^{-1}$yr$^{-1}$, 4) total C of A horizon did not change during the study period in Control plots, thus %C of A horizon prior to the litter-input manipulation was 6.2 prior to the treatments (Table 2-5), and 5) bulk density of the A-horizon soil was 1.3 g/cm$^3$, then new organic-C that entered the mineral soil during the 4-yr treatment period was less than 3 % of C in SOM existed within 0-15 cm of the A horizon prior to the treatments Double Litter Double Wood, and Control.

### Effect of litter-input manipulation on DOM chemistry in the field

The reason why the DOM chemistry of the O-horizon leachate was similar among the DIRT treatments, even though litter type affected the chemistry of DOM in lab extractions, could be explained by microbial activity. Microbial immobilization and decomposition in the O-horizon could equalize differences in the chemistry of DOM derived from the different DOM sources. If DOC production were predominantly controlled by abiotic dissolution and sorption (by hydrogen bonding) between DOM and O horizon material, and if microbial modification played a relatively minor role in DOC production, as hypothesized in some recent studies (Qualls and Haines 1991, Qualls 2000), I would expect to see similar DOC composition between O-horizon leachate under Double Litter treatment and lab extract of Oi material, and between O-horizon leachate under Double Wood treatment and lab extract of Class 1 wood material (i.e., high HiN and HoN content). Instead, I found that the composition of O-horizon leachate was similar to that of lab extracts of Oa and Class 5 wood materials (i.e., high HoA and HiA content). Because both HoA and HiA are plant-derived compounds that have been highly modified by microbes (Guggenberger et al. 1994), high proportion of these fractions in the O-horizon leachate of all DIRT treatments would suggest that DOM production in the O horizon depends largely on microbial modification of OM. A paradox that extractable pool size was larger for the newly-fallen litter (Oi and Class 1 wood materials) than for well-decomposed litter (Oa and Class 5 wood materials) but that the chemical composition of O-horizon leachate showed the signals of well-decomposed litter may be explained by either or both of: 1) physical leaching of labile OM followed by immediate retention by organic-organic bond (i.e., hydrogen bond) within the O horizon (Qualls 2000), and 2) much smaller surface areas for newly-fallen litter than for well-decomposed litter that may eventually reduce the efficiency of physical leaching of OM.

Alternately, DOM added from the native O horizon existing prior to DIRT manipulations might have obscured the signal from the treatments.

## Net removal and release of DOM in the field

Two most plausible explanations to the strong 1:1 relationship found between net DOM removal and the concentration of DOM in incoming water, regardless of season are; 1) microbes in the soil were strongly C-limited rather than temperature or moisture and were able to take up most DOM in the incoming water, or 2) the contribution of abiotic sorption was more important than biotic immobilization to the removal of DOM in mineral soil. Because my result shows that %HiN of total DOC in the O-horizon leachate of the DIRT study site was <2% (Fig. 2-2), and because HiN is the most biodegradable fraction of all fractions (Michaelson et al. 1998, Jandl and Sollins 1997, Qualls and Haines 1992) and %HiN is strongly and positively correlated with biodegradability of total DOM, the former explanation would not be valid in this study. On the other hand, in a laboratory incubation, over 70% of total DOC can be removed by abiotic sorption (summarized in Neff and Asner 2001), supporting the idea that abiotic sorption rather than microbial immobilization plays a major role in DOM removal.

The similar slope between the two different soil layers (0-30 and 30-100 cm) suggests that DOM sorption did not differ between the two soil layers. This contradicts some recent findings that the magnitude of DOM sorption increased with decreasing soil organic matter content. For example, Kaiser and Zech (1998a) found weaker DOM sorption to A-horizon material (i.e., gentler slope and more negative intercept for the regression line of sorption isotherm) than to B-horizon in European forest soils. They also found that coating of synthetic clays with organic matter reduced DOC sorption (Kaiser and Zech 1998b). Similarly, Zysset and Berggren (2001) found stronger sorption of DOM to the Bs1 and Bs2 horizon of a Podzol than to the Bh horizon. These authors hypothesized that reduced sorption was caused by the saturation of sorption sites on minerals with organic matter. One potential explanation for little change in DOM sorption with increasing SOM content in the current study might be that the soil at my site has greater capacity to retain DOM than soils used in the studies above. The soils of the Andrews Experimental Forest are rich in amorphous sesquioxides, especially amorphous Al-hydroxides (i.e., oxalate-soluble Al = 1.1%, see Table 3-1; pH in NaF=

10.8, data not shown), which strongly sorb DOM (Kaiser and Zech 1998b). Alternately, no difference in the sorption patterns between the shallower and deeper soil layers could be because DOM was exposed to more mineral soil with available sorption sites as water percolates the soil columns than in the lab incubation studies mentioned above.

The slopes and intercepts of the regression lines relating net DOM sorption to DOM concentration in incoming water suggest nearly complete net sorption and constant release of DOM that are independent of each other, regardless of soil depth. Possibly, DOM in the O-horizon leachate is immediately and completely sorbed on the mineral soil via abiotic sorption. Sorbed DOM would be stable until microbial degradation slowly converts the sorbed DOM to leachable OM. This idea is consistent with a study by Kaiser et al. (2001) who hypothesized that an increase in $\delta^{13}C$ values of bulk DOC with increasing depth is due to two separate processes: preferential sorption of $^{13}C$-depleted Ho-DOC and the release of $^{13}C$-enriched Hi-DOC from SOM. Their hypotheses were also consistent with the study of Schiff et al. (1997) and Dai et al. (1996). The former study, using the $^{14}C$ dating technique, showed that DOC in groundwater was older than DOC derived from the forest floor. They attributed this to extensive recycling of DOC in the soil column before elution to groundwater. In the latter study, the authors found a shift in $^{13}C$-NMR composition from the forest floor leachate to Bhs horizon soil and concluded that DOC sorbed on the Bhs horizon underwent further decomposition. Slow but constant decomposition of SOM would constantly refill a water-extractable SOM pool, which is subject to leaching with the downward move of water (Christ and David 1996; Kalbitz et al. 2000; Wagai and Sollins 2002), and consequently cause $^{13}C$-enrichment and $^{14}C$-depletion of DOM in deep mineral soils.

When compared with the overall mean of incoming DOC (averaged over all treatments by year, calculated from Table 2-5), DOC constantly released from the mineral soil (intercepts of the regression lines in Table 2-6) equaled 20 % of average incoming DOC for the top soil layer (0-30 cm, for the first year), and to 11-19 % for the deeper soil layer (0-100 cm, for both years). Similarly, DON constantly released from soil equaled 10% of average incoming DON for the topsoil layer (for the first year). For the deeper soil layer, DON constantly released from soil was calculated to be 50-88 % of incoming DON (for both years).

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# CHAPTER 3

# CHEMICAL AND SEASONAL CONTROLS ON THE DYNAMICS OF DISSOLVED ORGANIC CARBON AND NITROGEN IN A CONIFEROUS OLD-GROWTH STAND IN PACIFIC NORTHWEST, USA.

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## ABSTRACT

Dissolved organic matter (DOM) plays an important role in transport of C and essential nutrients such as N, P and S. DOM is also critical for the formation of soil organic matter (SOM), which is a long-term reservoir for C and those nutrients. I studied chemical and seasonal controls on the production and removal of DOM in the soil of a Douglas-fir old-growth stand in the Pacific Northwest, USA. The O horizon was not usually the dominant source for dissolved organic C (DOC) as has been reported in many other studies. Rather, net DOC production from the shallow mineral soil (0-10 cm) was at times larger than from the O horizon. The shallow mineral soil was also the largest net source of dissolved organic N (DON), and the production of DON was not associated with the production of dissolved inorganic N (DIN). Both DOC and DON concentrations at the deep soil in this study site were within the ranges that had been reported for various forest and soil types.

The base fraction, which is sorbed on cation exchange resin and composed mostly of free amino compounds, accounted for a high proportion of the total DOC in the deep mineral soil water (up to 19 % of total DOC), in spite of low background N inputs.

In both field and lab studies, the hydrophobic acid fraction (HoA) was strongly removed from solution. The hydrophilic acid fraction (HiA) was also strongly removed from solution in the field study, but less so than HoA. In the lab study, strong removal of HiA was observed only for the extracts of well-decomposed litter. This suggests that the HiA fraction extracted from different sources may contain functionally distinct compounds that were not separated by the chemical fractionation method. Little net sorption of the hydrophobic neutral fraction (HoN) observed for most litter extracts in the lab study suggests that hydrophobic interaction may not be the predominant mechanism of DOM sorption.

## INTRODUCTION

DOM plays an important role in ecosystem C dynamics as well as the transport and storage of nutrients (N, P, S). DOM concentration is generally highest at the bottom of the O horizon and decreases with increasing soil depth, indicating that DOM is produced in the O horizon and removed in the soil. Processes that control DOM production and removal in soils are only partially understood. Recent studies suggest that DOM is produced by the combination of two processes: 1) a replenishment of potentially soluble SOM pool by decomposition of both new and old OM and by the release of microbial and plant metabolites (microbial polysaccharides, root exudates and sloughed cells), followed by 2) physical leaching of the OM in the pool (Christ and David 1996; Kalbitz et al. 2000; Qualls 2000; Wagai and Sollins 2002). On the other hand, microbial uptake and abiotic sorption to soil minerals are the two major processes that remove DOM from soil water and stabilize it into SOM (McDowell and Likens 1988; Qualls and Haines 1992). Studies have suggested that abiotic sorption is responsible for most DOM retention in soil. McDowell and Likens (1988) reported that the labile C fraction of soil solution (assumed to be mainly carbohydrates) is very small (approx. 3 - 6% of total DOC). Similarly, using a laboratory batch incubation of soil solution, (Qualls and Haines 1992) found that the labile (rapidly degradable) fraction of DOC in Oa horizon solution was too small (6 - 19%) to explain a 100-fold reduction in DOC between the O and B horizons. On the other hand, biodegradable DOC determined by a flow-through bioreactor ranged from 10 to 40% of total DOC of the O-horizon leachate hardwood and coniferous forest ecosystems in the Northeast (Yano et al. 2000). This would suggest that abiotic sorption might not always be the predominant process for DOM removal in the mineral soil. The relative importance of biotic and abiotic removal is still not clear due to the difficulty in evaluating these processes independently.

A number of studies have proposed various chemical and physical mechanisms that would cause DOM sorption, such as hydrophobic interactions (e.g., Jardine 1989), ligand exchange (e.g., Greenland 1971; Parfitt 1977; Kaiser and Zech 1998b), cation bridging, hydrogen bond, and Van der Waals forces (Sposito 1989; Qualls 2000). Therefore, differences in the chemistry of bulk DOM may affect the degree of sorption and, consequently, SOM formation. Due to the complex nature of both the sorbate and the sorbent in the natural environment, sorption studies have mostly been limited to lab incubation of known DOM compounds and simple minerals (i.e., synthesized clays). Sorption between natural DOM and mineral soils is less frequently studied. Studying changes in DOM chemistry with soil depth in the natural environment is thus essential to the understanding of the chemical controls on DOM dynamics in the field environment.

Chemical fractionation by resins can separate DOM into different functional groups based on the difference in the affinity of DOM to different types of resins (Leenheer 1981; Qualls and Haines 1991). The hydrophilic neutral fraction is rich in polysaccharides (Guggenberger et al. 1994; Dai et al. 1996) and is highly labile (Qualls and Haines 1992; Jandl and Sollins 1997), as opposed to the hydrophobic acid (anionic) fraction, which is highly aromatic and rich in carboxylic acids (Guggenberger et al. 1994; Dai et al. 1996). The base (cationic) fractions contain labile N compounds such as free amino acids, peptides and proteins.

The objectives of this study were to examine the chemical and seasonal controls on the dynamics of dissolved organic C and N, and to understand the relationship between DOM chemistry and its sorption potential. I examined changes in total DOM and its chemistry by soil depth and season for soil water collected in lysimeters in a temperate forest ecosystem in western Oregon, USA. I also examined in a lab incubation study the sorption potential of DOM extracted from different sources. The composition of DOM was determined using the chemical fractionation mentioned above.

## METHODS

### Study site

The study was conducted at the H. J. Andrews Experimental Forest located in the

Cascade Mountains in western-central Oregon, USA (44°15′N, 122°10′W, 726 m elevation). The area is characterized as a temperate rain forest with mean annual temperature of 7.9 °C and mean annual precipitation of 237cm $yr^{-1}$. Over 70% of the precipitation occurs during the wet season, mostly as rain, between about November and March (Sollins et al. 1980), although the months and length vary year to year. During the period of this study (1999-2001), I defined wet seasons to be Oct.-May of 1999-2000 and Nov.-May of 2000-2001. Background N deposition in this area is ~2 kg N $ha^{-1}yr^{-1}$ (Sollins et al. 1980). In 1998, I established experimental plots in an old-growth Douglas-fir stand adjacent to the DIRT experiment site (see Chapter 2). Soils are derived from volcanic parent materials and reveal strong Andic tendencies such as high amorphous Al hydroxide content (based on oxalate-extractable Al, Table 3-2) and high pH in 1M NaF (10.8; Yano, unpublished data). The soils had a thin O horizon (~ 2 cm) that was difficult to separate from the moss layer, lying on a 10-20 cm thick A horizon with abundant fine roots, over a 20-30 cm thick B horizon with less fine root biomass.

### Collection of water samples

In the fall of 1998, I set up five 5 m x 5 m plots in a ~0.2-ha area of the forest. Within each plot, one throughfall (TF) collector, one zero-tension lysimeter (at the bottom of the O horizon, or 0 cm), and 4 ceramic-cup tension lysimeters were installed at 10, 20, 30, and 70 cm in the mineral soil. The lysimeters within each plot were located at least 2 m apart.

To minimize biological degradation and chemical alteration of DOM in the water collected in the TF collectors and zero-tension lysimeters, water samples were retrieved within 24 hrs after the end of rain events between April and November. The rest of the year, when rain events are intermittent, water was collected approximately every 3 weeks except for a 2-3 month period when access to the plots was blocked by deep snow. Samples collected before the first wet season (the first 10 months after lysimeter installation) were not included in the analysis to avoid effects of lysimeter installation on

DOM chemistry. All samples were transferred on ice to Oregon State University, where volume was determined and samples were filtered through combusted Whatman GF/F glass fiber filers. The samples were then subsampled for the analysis of DOC and DON, and the remainder was combined by season for each wet season using a volume-weighted method. The combined samples were then stored frozen until chemical fractionation analysis.

### Plant litter extracts for lab sorption

To test the effect of DOM chemistry on sorption potential, I used DOM extracted from several different sources: Douglas-fir needles and wood in various stages of decomposition, and fine roots. Needle litter was collected from three young Douglas-fir stands at a site in the Andrews Experimental Forest where the O horizons were much thicker (~4 cm) and composed mostly of Douglas-fir needles (85.5% on average by weight) than in most old-growth stands, including my study site (~2 cm thick, composed of a moss layer with minimal accumulation of needles). From the O horizon, two horizons were separated out with a knife: Oi horizon (freshly fallen needles with minimal decay) and Oa (well-decomposed unidentifiable material).

Wood litter from two decomposition classes, Class 1 and Class 5 logs, was collected from Douglas-fir stands. Classification of logs was based on the method of described in Sollins (1982) that is widely used for ecosystems in the Pacific Northwest. Class 1 logs are newly-fallen, with current-year twigs still attached and with minimal decomposition of bark and wood. Class 5 logs are extremely decomposed such that sapwood and bark are absent and wood is mainly fragmented and cannot be lifted intact. Class 1 logs were further separated into bark, sap wood, and heart wood.

Roots were harvested from Douglas-fir seedlings grown in a mixture of the mineral soil of the study site and commercial silica sand (soil : silica sand = 1:1) in pots for 1.5 yrs. Upon harvest, the soil-silica sand mixture was removed by gentle washing in DI water (root-water contact time < 30 min). The distribution of root-diameter size was:

72.7%, 20.1%, and 6.76% for < 1.0 mm, 1.0-2.0 mm, and >2.0 mm, respectively. All litter was air-dried, ground to pass 30-mesh screen, then extracted in DI water in a shaker at 100 rpm at 20°C for 48-68 hours. Solid-to-water ratios for the extraction were 1:40 for needle, and 1:25 for wood and root litter. Solid was separated by centrifugation at 7000 rpm for 15 min, then the supernatant was filtered through Whatman GF/F glass fiber filers, analyzed to determine initial concentration of DOC and chemical fraction composition (see below). The composition of chemical fraction for these extracts is shown in Table 3-1.

**Table 3-1.** Chemical fraction composition of litter extracts. See the text below for details about the fractionation method.

| Litter type | Decay stages | Hydrophobic |  |  | Bases | Hydrophilic |  |
|---|---|---|---|---|---|---|---|
|  |  | Phonol | HoA | HoN |  | HiA | HiN |
|  |  | % |  |  |  |  |  |
| Needle | Oi | 9 | 27 | 4 | 2 | 25 | 30 |
|  | Oa | 4 | 47 | 3 | 5 | 35 | 5 |
| Wood | Class 1 | 14 | 16 | 11 | 1 | 17 | 36 |
|  | Class 5 | 2 | 62 | 0 | 6 | 18 | 13 |
| Fine root | New | 16 | 21 | 3 | 1 | 17 | 37 |

Lab sorption

B-horizon soil (Table 3-2) collected from an area near the study plots was used for a sorption test. The soil was sieved to < 2 mm without drying to minimize changes in soil mineralogy and stored at 3 °C prior to use. The litter extracts were thawed, filtered through pre-rinsed Durapore membrane filters (nominal pore size 0.45 μm), and diluted to 40 mg-C/L. Seventy-five mL each of these solutions, as well as DI water as a control, were added to 7.5 g of the field-moist soil (solid : solution ratio = 1 : 10, n=3). Prior to incubation, the pH of the suspension was adjusted to 4.1 ±0.2, the pH of most litter extracts. The suspension was shaken gently at 3 °C in the dark for 75 hours. A preliminary test showed that this incubation period was necessary for the DOC concentration of the suspension to reach equilibrium, and that C loss via respiration was not significant (<4%) of total DOC loss.

**Table 3-2.** Characteristics of B-horizon soils used for the sorption test. Values in parentheses show 1 SE (n=3).

| % of dry soil | | | g/kg dry soil | | | | | |
|---|---|---|---|---|---|---|---|---|
| Total C | Total N | C:N | $Fe_d$ | $Fe_o$ | $Fe_p$ | $Al_d$ | $Al_o$ | $Al_p$ |
| 1.31 | 0.2 | 15.7 | 38.2 | 11.0 | 4.8 | 13.8 | 11.0 | 5.2 |
| (0.0) | (0.1) | (2.1) | (0.3) | (0.3) | (0.0) | (0.1) | (0.1) | (0.2) |

$Fe_d$, $Al_d$: dithionite-citrate extractable Fe and Al.
$Fe_o$, $Al_o$: ammonium oxalate extractable Fe and Al.
$Fe_p$, $Al_p$: sodium pyrophosphate extractable Fe and Al.

The suspension was centrifuged for 40 min at 7000 rpm and filtered through pre-rinsed 0.45 µm-Durapore membrane filters to remove particulate organic matter. DOC concentration before and after the incubation was analyzed to determine sorption to the mineral soil. For the chemical fractionation analysis, the three replicates were combined after DOC measurements of post-incubation samples to ensure enough volume for analysis.

## Chemical analysis

### <u>DOC and DON</u>

All lysimeter and TF samples were analyzed for DOC with high temperature platinum-catalyzed combustion (Shimadzu TOC-5000A HTCO carbon analyzer). Dissolved organic N was determined as the difference between total dissolved N (TDN) and dissolved inorganic N. TDN was measured by persulfate digestion procedure (Koroleff 1983), followed by $NO_3^-$ analysis. Nitrate-N was measured using the hydrazine sulfate reduction method and $NH_4$-N was determined using the Berthelot reaction method using a Scientific Instruments Autoanalyzer. Because $NO_2$-N in soil water or solutions in an aerobic environment is negligible (Qualls and Haines 1991, Currie et al. 1996), DON was calculated as:

$[DON] = [TDN] – [NO_3^-\text{-N}] – [NH_4\text{-N}]$

Because DON was calculated by difference, values sometimes fell slightly below 0 mg $L^{-1}$. Negative DON values were considered to be 0 mg $L^{-1}$.

## Chemical fractionation

Lysimeter water (all depths except for 30 cm), TF, and litter extracts were fractionated into 5 functional fractions using the method of Qualls and Haines (1991). In brief, the method fractionates DOM by its hydrophobicity, then into acid (anionic), base (cationic), and neutral fractions by its affinity to 3 different types of resins (hydrophobic, cation-exchange, and anion-exchange resins). The weak hydrophobic acid fraction (Phenol) includes small phenolic compounds (e.g., tannin and flavonoids). The strong hydrophobic acid fraction (HoA) includes microbially altered plant-derived material of larger molecular size humic substances and humic-bound amino acids and carbohydrates. The hydrophobic neutral fraction (HoN) is less microbially altered plant-derived material and contains waxes, fatty acids, and microbial lipids. The hydrophilic acids (HiA) are partly microbially synthesized and partly plant-derived material that are lower molecular size humic-like substances with high carboxyl-to-C ratios. The hydrophilic neutral fraction (HiN) contains non-humic-bound carbohydrates, mainly of microbial origin, and may contain simple sugars (e.g., hexose, deoxysugars). The hydrophobic and hydrophilic base fractions contain free amino acids, peptides and proteins. Because the proportion of hydrophobic base fraction was very small for all soil solutions and litter extracts tested in this study (≤2 % of total DOC), both hydrophobic and hydrophilic bases were combined and reported as a base fraction (Bases).

### Data analysis

All measurements of DOC and DON for the water samples were grouped by 3 seasons within the same wet season: Oct. to Nov. (fall), Dec. to Mar. (winter), and April to May (spring). To examine the effects of soil depth and season on total DOC, DON, and DOC:DON ratio, I used repeated measures analysis with time as a repeated factor. ANOVA was used to detect differences in DOC sorption among different litter extracts. Values were natural-log transformed to obtain appropriate normality prior to the analysis, followed by back-transformation to obtain least square means and 95% confidence limits. To evaluate the relationship between initial DOC chemistry and sorption, correlation analysis was used.

Extraction and all chemical analyses for Class 1 wood were done on bark, sap wood, and heart wood, separately. Values for whole wood were then back calculated based on %tissue volume of whole wood for Douglas-Fir with mean log diameter of 52 cm (bark=12%, sap wood= 30%, and heart wood=58%) (Harmon 1992).

Net DOM removal in B-horizon soil in the field was determined as the difference in DOM concentration between two soil depths, 30 and 70 cm, after correction for evapotranspiration (ET). Water losses via ET from 0-30 and 30-70 cm soil layers were estimated from water fluxes reported by Sollins et al. (1980) and from root distribution data for a nearby stand (Santantonio and Hermann 1985). I assumed that water flux within a 70 cm soil column (this study) was similar to that at 100 cm depth reported by Sollins et al. (1980), thus water lost to ET from the 0-30 and 30-70 cm soil layers was calculated to be 13 % of the volume of O-horizon leachate and 10 % of soil water at 30 cm, respectively. The SAS System (SAS Institue Inc. 1999) was used for all statistical analyses.

Although the composition of chemical fraction was not determined for 30 cm lysimeter water, I assumed that the composition at 30 cm soil water was similar to that at 20 cm.

## RESULTS

### DOC

DOC concentrations generally increased from TF to 0 or 10 cm in the soil, then decreased to 70 cm (Fig. 3-1), indicating net release of DOC from the O horizon and/or 0-10 cm mineral soil layer and net removal of DOC in the 10-70 cm soil layer. None of the increases in DOC were significant with the exception of the first winter, where the increase between TF and 10 cm soil water was significant. For all seasons, the lowest DOC concentrations were found at 70 cm, and the differences between the highest (found at either 0 or 10 cm depth) and the lowest (70 cm) DOC were significant, indicating net removal of DOC in the mineral soil. The magnitude of change in DOC concentration with soil depth was largest for the fall samples followed by spring samples, although the magnitude of change was not significant for the fall and winter due probably to large variability of DOC values.

The influence of season on DOC concentration differed with soil depth. For O-horizon leachate, DOC concentrations were always greater in the fall than in winter ($P < 0.001$ for the first year and $P = 0.021$ for the second year). At 10, 20, and 30 cm in the mineral soil, DOC concentrations in the first fall were consistently greater than in the winters and spring. Generally, the variability of DOC was greater in the shallow soil (0-10 cm) than in deep soil, and greater in fall than in winter or spring (Fig. 3-1).

### DON

The patterns of DON were generally similar to those of DOC. For all seasons, mean DON concentrations increased significantly from TF to the 10 cm soil water, although, contrary to DOC, the increase between TF and the 0 cm soil water was small, indicating that net release of DON was mostly from the 0-10 cm mineral soil layer. After reaching a maximum concentration at 10 cm (or 20 cm the first winter), DON concentration decreased to the levels of TF by 70 cm, indicating net removal of DON between10 and 70 cm (Fig. 3-1). The increase was greatest for the fall, and the lowest concentration in the mineral soil was found at 70 cm in all seasons, as found for DOC. Generally, the variability of DON was greatest at 10 cm where the highest DON was observed.

### DOC:DON

DOC:DON ratios were greatest and most variable in TF and the O-horizon leachate (48-113). Ratios decreased to a narrow range of about 20-34 in the 10 cm soil water and stayed relatively constant below 10 cm (20-28), regardless of season or depth (Fig. 3-1).

### Chemical fraction composition

Generally, HoA and HiA were the two dominant fractions in all the samples collected in the first year (Fig. 3-2). HoA dominated DOC in TF and the O-horizon leachate (53.4-66.5% of total DOC, Fig. 3-2). Percent HoA decreased with increasing soil depth and equaled % HiA at 70 cm. Percent Bases generally increased with increasing soil depth and became significant in the deep soil water (up to 19.5 % of total DOC). Percent Phenol, HiN and HoN were consistently small in TF and the soil profile in all seasons.

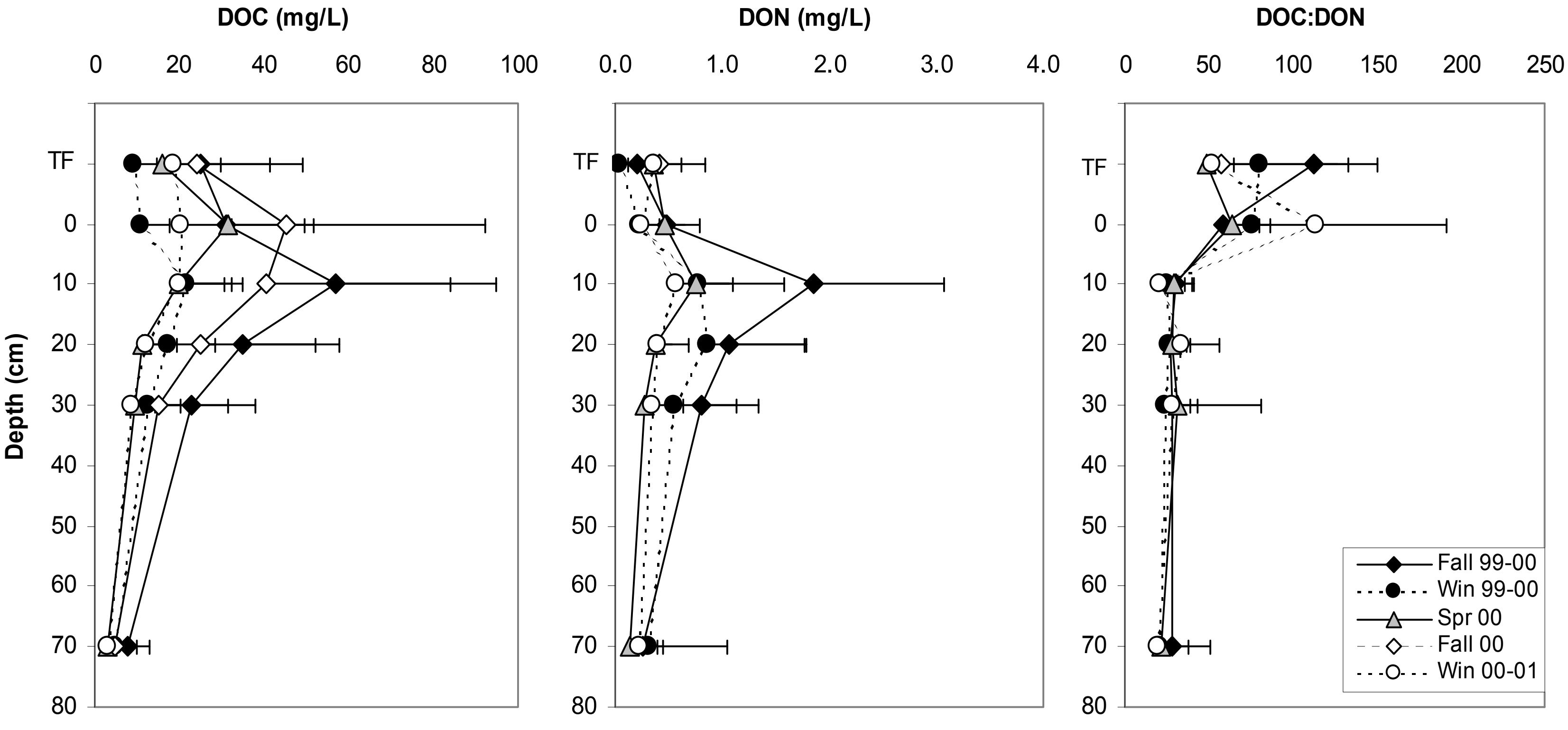

**Fig. 3-1.** Depth profiles of DOC, DON, and DOC:DON ratio for the samples of 1999-2000 and 2000-2001.

The increases and decreases in total DOC were generally associated with changes in the two dominant fractions, HoA and HiA (Fig. 3-3). For all seasons, the increase in total DOC from TF to the O-horizon leachate was associated with an increase in both percent HoA and HiA. Increases in the concentrations of HoA and HiA together accounted for 86-95 % of the total DOC increase, indicating that DOC produced within the O horizon and the 0-10 cm mineral soil layer was mostly in the form of acid. Of the total increase in these acid fractions, 45-68 % was due to the increase in HoA fraction. The decrease of total DOC concentration from 0 or 10 cm to 70 cm paralleled the decreases in both HoA and HiA fractions. The HoA and HiA fractions together accounted for 86-92 % of the total DOC decrease, of which 58-70 % was due to the decrease in HoA fraction, indicating preferential removal of HoA-DOC over HiA-DOC in the mineral soil. The concentrations of other fractions (Phenol, Bases, HoN, and HiN) changed somewhat with total DOC, but none of the changes were as influential to total DOC concentration as were changes in HoA or HiA.

### Net removal and release of DOM in the field

The correlation between DOM in incoming water and net DOM removal on a sampling event basis for 30-70 cm soil layer showed a tight relationship with negative intercept (Pearson's R = 0.935 for DOC, 0.969 for DON) (Table 3-3). The slopes of regression lines for the correlation were 0.9 for DOC and 0.88 for DON, indicating strong removal of both DOC and DON.

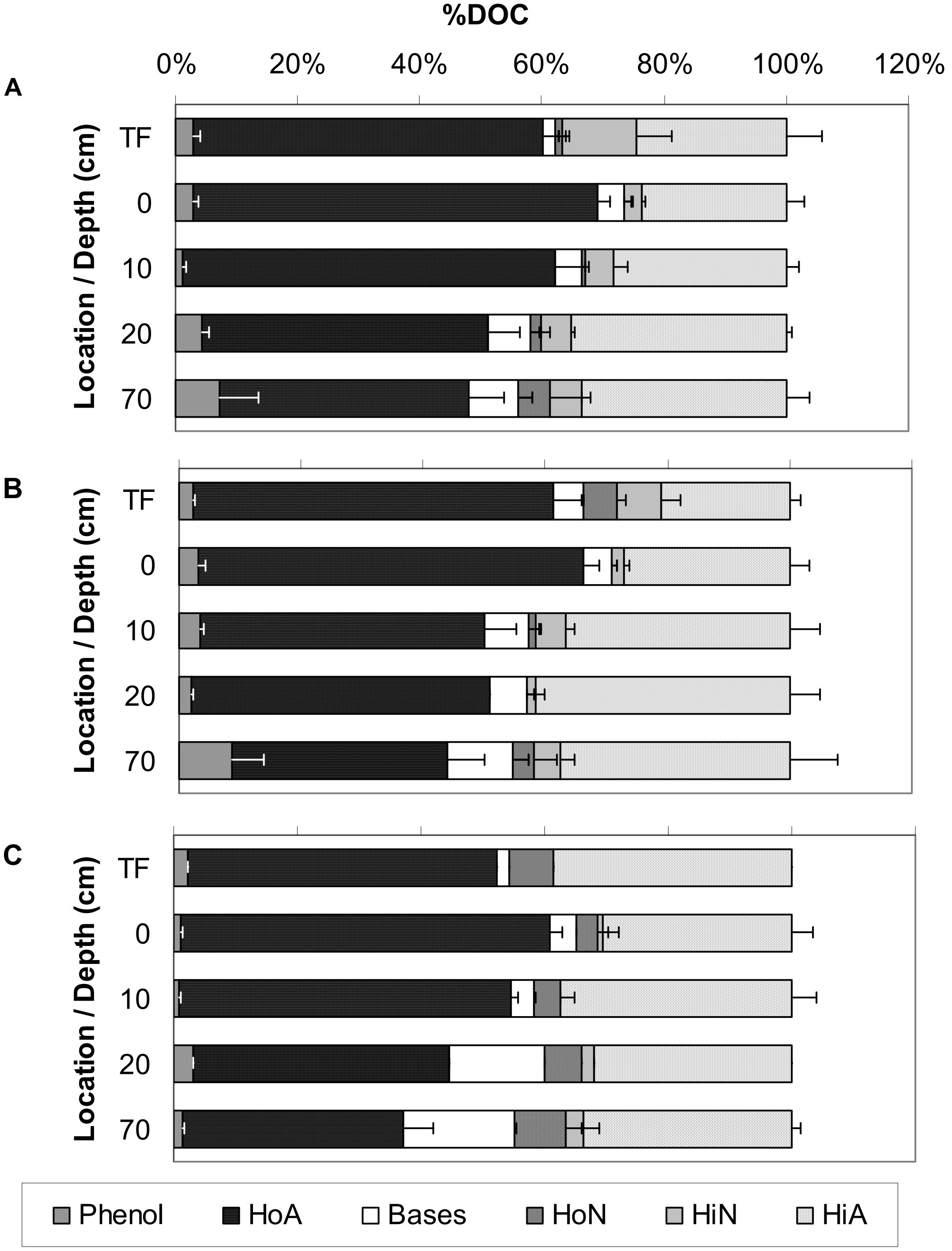

**Fig. 3-2.** Depth profiles of chemical fraction composition as percent of total DOC. Samples were collected in (A) fall 1999, (B) winter 1999-2000, and (C) spring 2000. TF=throughfall. Error bars indicate 1SE for n=3 to indicate analytical precision.

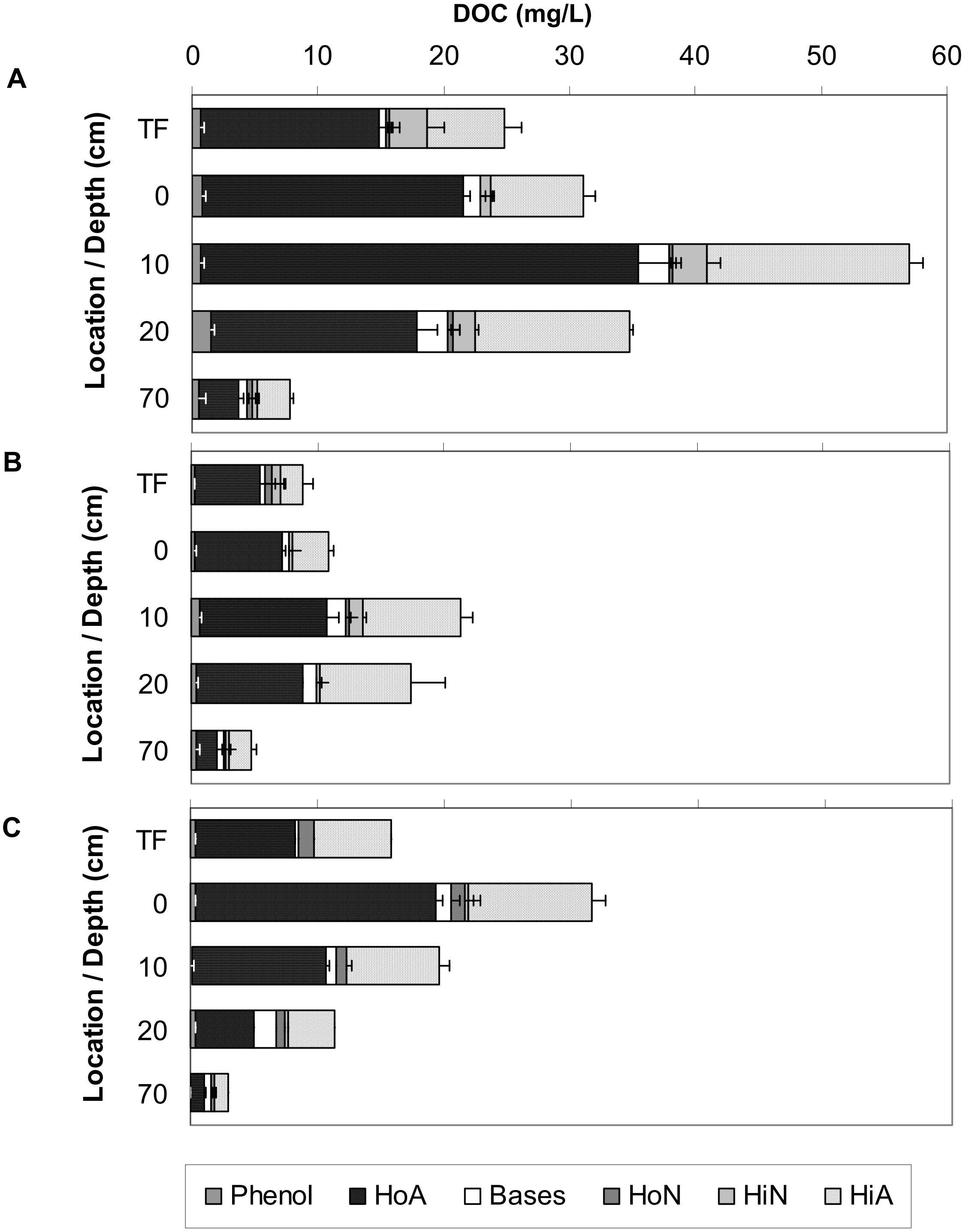

**Fig. 3-3.** Depth profiles of total DOC concentration and the distribution of chemical fraction within each sample location. The samples were collected in (A) fall 1999, (B) winter 1999-2000, and (C) spring 2000. TF=throughfall. Error bars indicate 1SE for n=3 to indicate analytical precision.

**Table 3-3.** Correlations between net DOM removal and DOM concentration in incoming water to a 30-70 cm soil layer.

| | Soil layer (cm) | Regression line | Pearson's R | *P*-value |
|---|---|---|---|---|
| DOC | 30-70 | $[NR_{DOC}] = 0.90 \times [DOC_{in}] - 2.54$ | 0.935 | <0.001 |
| DON | 30-70 | $[NR_{DON}] = 0.88 \times [DON_{in}] - 0.11$ | 0.969 | <0.001 |

$[NR_{DOC}]$: net removal of DOC, $NR_{DON}$: net removal of DON. Both as (mg/L)
$[DOC_{in}]$: DOC concentration of incoming water to a soil layer as (mg/L).
$[DON_{in}]$: DON concentration of incoming water to a soil layer as (mg/L).

### DOM removal (sorption) and chemical fraction composition

The lab sorption experiment clearly showed an influence of DOC source on DOC sorption. When litter types were compared, DOC derived from roots showed greater net sorption than DOC derived from needle (Oi, *P*=0.032) or wood (Class 1, *P*=0.033) (Fig. 3-5 A). In addition to litter type, DOC sorption was affected by the degree of litter decay. When DOC sources shifted from freshly-fallen to well-decomposed litter (Oi → Oa, Class 1 → Class 5), net DOC sorption increased by 34% for needle (*P*=0.029) and 36% for wood litter, although the increase for the wood litter was not significant (*P*=0.198).

The chemical composition of DOC sorbed to mineral soil in the lab stsudy differed among DOC sources. Greater total DOC sorption observed for old litter extracts was associated with greater removal of HoA and HiA (Oa and Class 5) (Fig. 3-5 B). Sorption of Phenol was important to total DOC removal for new litter extracts (Oi, Class 1, Root). HiN sorption was found only for new needle (Oi) and root extracts. The sorption of HoN was important only to Class 1 extract. The removal of HiN was found only for Oi and root extracts. Unlike other litter extracts, HiN in root extract was strong and composed of 31% of DOC that was sorbed (Fig. 3-5 B).

A positive trend was found between net DOC sorption and the % HoA of the in initial solution (R=0.335). This relationship was improved when the other two

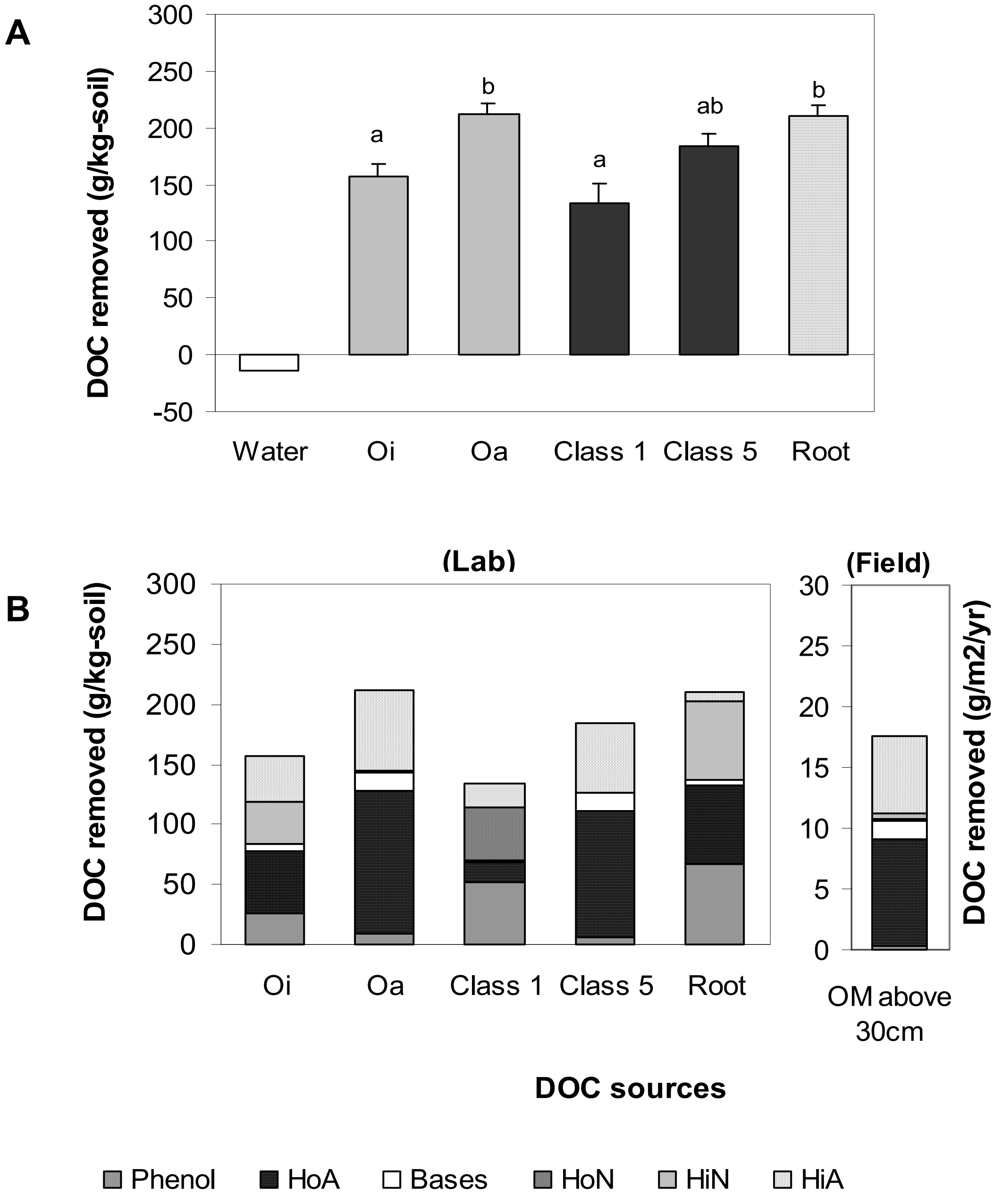

**Fig. 3-4.** DOC sorption in the laboratory incubation. DOC extracted from different sources was incubated with B-horizon soil. (A) Total DOC sorption. Water indicates net release of DOC from the B-horizon soil during the incubation. Error bars indicate 1SE for n=3 to indicate analytical precision. (B) Chemical fraction composition of DOC sorbed to the mineral soil determined as differences in chemical fraction composition between 30 (estimated) and 70 cm. See text for details about calculation. For both (A) and (B), sorption for Class 1 was calculated from DOC sorption of bark, sapwood, and heartwood. OM above 30cm: any organic matter that is a source of DOC that exists above 30 cm in soil.

hydrophobic fractions (Phenol and HoN) were included in the analysis (R=0.611, *P*=0.107) (Fig. 3-6), although neither Phenol nor HoN itself showed any trend with sorption.

In the field study, the chemical fraction composition of DOC that was removed between 30 and 70 cm mineral soil was similar to those of Oa and Class 5, i.e., most DOC removed in the soil was HoA or HiA with a small amount of Bases (Fig. 3-5 B).

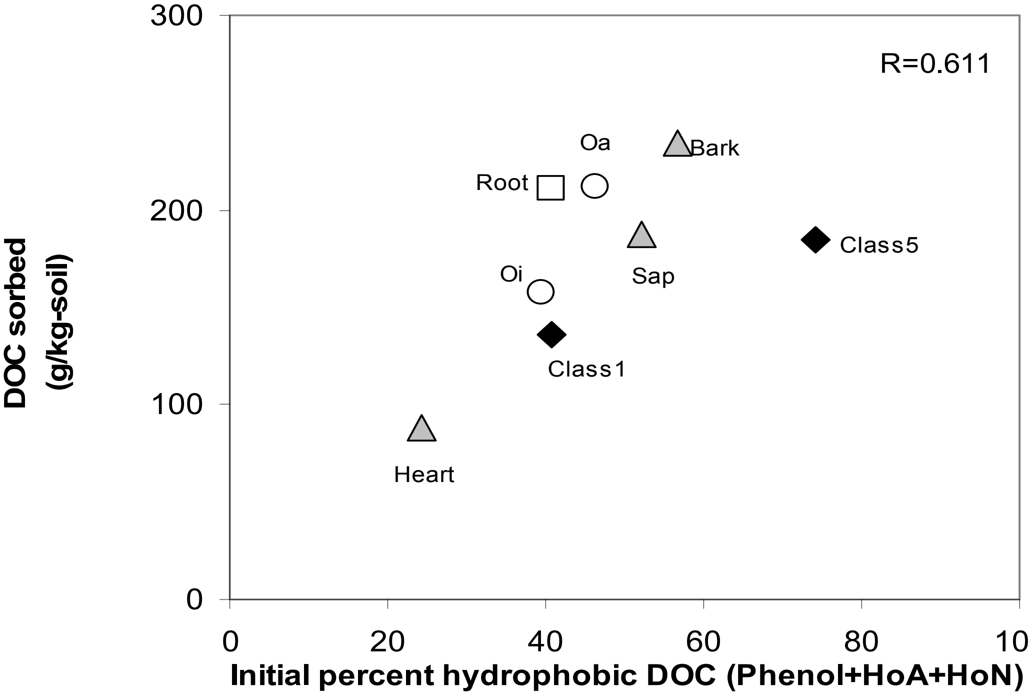

**Fig. 3-5.** Effect of initial DOC chemistry on laboraotry sorption. DOC sorbed was plotted for initial percent Ho-DOC (Phenol + HoA + HoN) fractions of total DOC.

To examine which chemical fraction is more subject to removal, % removal of initial fraction was compared. In the lab sorption study, in general, Phenol and Bases had high affinity to mineral soil (>70%), and neutral fractions (HoN and HiN) showed low affinity, except for HoN in Class 1 extract (Table 3-4). Affinity of strong acids (HoA and HiA) increased considerably with the decay of DOC sources; from 47 to 82% (HoA) and 40 to 63% (HiA) for needle litter, and from 24 to 52 % (HoA) and 25 to 100% (HiA) for wood litter.

For the field study, the greatest %removal was observed for the strong acid fractions (HoA and HiA). Percent removal of Phenol in the field study was not as high when compared to those of lab extract.

**Table 3-4.** Percent removal (sorption) of initial DOC for each chemical fraction. Removal was determined as a difference between before-and-after incubation for the lab study, and between 30 (estimated) and 70 cm DOC concentration for the field study.

| DOC sources | Decay stages | Hydrophobic | | | Bases | Hydrophilic | |
|---|---|---|---|---|---|---|---|
| | | Phonol | HoA | HoN | | HiA | HiN |
| | | % | | | | | |
| **Lab** | | | | | | | |
| Needle | Oi | 79 | 47 | ND | 100 | 40 | 29 |
| | Oa | 70 | 82 | 2 | 100 | 63 | ND |
| Wood | Class 1 | 83 | 24 | 85 | 74 | 25 | ND |
| | Class 5 | 82 | 52 | ND | 78 | 100 | ND |
| Fine root | New | 94 | 70 | 1 | 82 | 10 | 40 |
| **Field** | | | | | | | |
| OM above 30cm | | 44 | 75 | 29 | 60 | 70 | 55 |

ND: Not determined due to no HoN-DOC in the initial solution, or to no net removal during the lab incubation.
OM above 30 cm: Any organic matter above 30 cm that can be a source of DOC.

## DISCUSSION

### Sources of DOM

DOC concentration of TF at this study site (9-25 mg-C/L) was similar to those observed in other temperate coniferous forests (10-21 mg-C/L, Zech et al. 1994; Currie et al. 1996; Solinger et al., 2001). The DOM depth profiles suggest that the O-horizon of this study site was not always a large DOM source as has been observed in other temperate forests, where the highest DOM concentrations were found at the bottom of O horizon (e.g., Qualls and Haines 1991; Zech et al. 1994; Solinger et al. 2001). Rather, the mineral soil (0-10 cm) appeared to be a more significant source of DOM than the O-horizon, especially for DON. One explanation could be that the O horizon of this study site was quite thin (~ 2 cm), composed of a moss layer with minimal litter accumulation, and thus does not have the potential to add a significant amount of DOM to soil water. In other temperate coniferous forests the highest DOM concentrations were found just below the O horizon layer, the O horizon was thick enough to be further separated into sub layers (Oi, Oe, and Oa).

DOM in the shallow mineral soil (0-10 cm) can derive from root turnover, root exudates, microbial turnover, microbial breakdown of plant OM and SOM via extracellular enzymes, and physical dissolution of SOM. It is likely that the fine roots and/or microbial activity in rhizosphere contribute to the DOM production, because ~70% of fine and small roots were concentrated within the top 25 cm in a nearby Douglas-fir forest (Santantonio and Hermann 1985), and because the rhizosphere sustains the highest microbial activity in the soil (Kennedy 1998).

Large net DOM production in the shallow mineral soil (0-10 cm) in the fall may be due to an increase in root and microbial activities caused by an increase in water availability at the start of the wet season. Alternately, higher concentration in fall than winter in the shallow mineral soil may represent flushing of soluble OM produced by one or more processes mentioned above, but not flushed from the soil during the summer dry season. Simple dilution of soluble SOM by increased water flux could also result in the lower DOM concentration in the winters.

$NH_4^+$ and $NO_3^-$ showed no increase at 10-cm depth (Yano, unpublished data), thus, the net production of N in the shallow mineral soil was only as DON.

The DOC:DON ratio decreased from O-horizon leachate to 10 cm soil water while both DOC and DON concentrations increased. Thus, DOM compounds produced from the 0-10 cm soil layer were N-rich. A decrease in DOC:DON could also result from strong uptake of high-C:N DOM (e.g., carbohydrates) as DOM is produced.

Alternately, the decrease in DOC:DON ratio could be due to the dissolution of SOM of low C:N. C:N of the bulk soil next to this study plots was 26.6 (n=2), close to that of DOM in the soil water. The increase of total DOC between 0 and 10 cm in the mineral soil was also associated with large increases in HiA and small increases in Bases. Because both HiA and Bases are N rich relative to other fractions (DOC:DON was 10-52 for HiA and 3-10 for Bases, reported by Qualls and Haines 1991), net release of those fractions from the mineral soil layer would have reduced the DOC:DON ratio of bulk soil water.

A major portion of the net DOC increase from TF to the O horizon was due to increase in HoA-DOC (> 70 % of the total DOC increase), consistent with the findings of Guggenberger et al. (1994) who hypothesized that most DOC produced in the O horizon was a result of microbial decomposition rather than physical dissolution of plant materials. Increases in the concentration of HiA from 0 and 10 cm soil water (only in the fall and winter) were much larger than those between TF and 0 cm soil water. This would indicate that the process of DOC production in the mineral soil is different from that in the O horizon, i. e., the mineral soil produces significant amounts of HiA in addition to HoA. Guggenberger et al. (1994) found that HiA-DOC appeared to show higher degree of oxidative biodegradation (i.e., more side-chains are oxidized) than HoA-DOC, whose chemical structures are more similar to OM of plant underwent further decomposition.origin than are those of microbial origin. Possibly, HoA, once released from the O horizon or decomposing roots, then sorbed in the mineral soil, was further degraded to become potential HiA, although still sorbed on the solid phase. This idea is supported by the study by Dai et al. (1996) who found a shift in $^{13}$C-NMR composition from forest floor leachate to Bhs horizon soil and suggested that DOC sorbed on the Bhs horizon underwent further decomposition.

### Net removal of DOM in the field

A decrease of DOM with increasing soil depth indicates net DOM removal in the mineral soil. Because HiN is the most labile fraction of all (~15% of HiN was fast-degrading DOC; Jandl and Sollins 1997), and because the content of HiN at 30 cm was too small (3% of total DOC, estimated) to account for total net DOM removal in the B-horizon soil (90% of total DOC at 30 cm depth), the removal may be due mostly to abiotic sorption. The regression lines between incoming DOM to the B-horizon soil (30-70 cm layer) and net removal would suggest that most DOM entered that soil layer was removed (90% for DOC and 88% for DON). This pattern was similar to those observed for 30-100 cm soil layer at an adjacent location within the same stand, where 97-104% of incoming DOC and 94-98% of incoming DON were removed (Chapter 2).

The decrease of DOM concentration with increasing soil depth below 10 cm with minimal change in DOC:DON suggests that the DOM removal processes (either microbial uptake or abiotic sorption) did not favor either C- or N-rich compounds. This is consistent with the findings by Qualls and Haines (1992) that biodegradation of DOM in the mineral soil water did not favor either C or N (i.e., no change in DOC:DON ratio after biodegradation). In a separate study, they found consistent decrease in DOC:DON with increasing soil depth (Qualls and Haines 1991), which they concluded was primarily due to a shift in the distribution of chemical fractions from high to low DOC:DON, and that the shift was caused by differences in the sorption affinity of each fraction to soil minerals. In this study, however, the decrease in DOM concentration with increasing soil depth was not associated with N enrichment. Little N enrichment with soil depth in spite of the enrichment of HiA and Bases fractions (N-rich fractions) would suggest that the distribution of C and N in the chemical fractions for soil solutions in this study might be different from those that have been reported elsewhere.

## Chemical fraction composition and DOM removal (sorption)

The magnitude of removal (sorption) varied among the chemical fractions. Dai et al. (1996) reported that the HoA fraction was most strongly removed in the field, consistent with the results of my field study where the largest portion of DOC removed in the mineral soil was HoA (49% of total net removal). This pattern was consistent with the results of lab sorption study: HoA was generally a predominant fraction of DOC that was sorbed to the mineral soil (31-57% of DOC sorbed), except for Class 1 extract where sorption of HoA accounted only for 13% of DOC sorbed.

HiA was the second most strongly removed fraction in the field (37% of total net removal). This was partially supported by the lab incubation study, in which HiA was the second largest fraction of DOC (25-32%) that was removed, except for Class 1 and root extracts. This would suggest that HiA extracted from different sources might contain functionally different DOC compounds.

Although initial %HiN was similar among new litter extracts (Oi =30%, Class 1 =36%, and root =37%), HiN in Class 1 extract was not sorbed to mineral soil while HiN in Oi and root extracts were well sorbed. Thus, HiN in Oi and root extracts might contain functionally more similar DOC compounds compared to those in Class 1 extract. The reason of high net DOC removal for the new root extract, in spite of its low HoA content (i.e., root DOC had less than half the % HoA of Oa or Class 5 extracts), would be because of this exceptionally strong removal of HiN in the root extract.

Most DOC in Phenol and Bases (>70% of initial) was sorbed in the lab sorption study, suggesting that these fractions would contain DOC compounds with high affinity to mineral soil. In spite of its high %sorption, the importance of Bases to total DOC sorption was low due to its low initial content (1-6% of total initial DOC).

Although Ho-DOC is known to be more strongly sorbed than Hi-DOC (Kaiser and Zech 1998a; Kaiser and Zech 1998b), I did not find any trend of preferential sorption for HoN itself, with the exception for Class 1 wood extracts. Combined with the fact that HiA is the second most strongly sorbed fraction in the field, my results suggest that DOC sorption may be caused by ligand exchange between the hydroxyls of clays and acidic functional groups of DOC (e.g., Kaiser and Zech 1998b), rather than hydrophobic interaction between DOC and clays (attraction between hydrophobic portions of clay and DOC) as proposed by Jardine et al. (1989). The fact that the preferentially sorbed DOC fractions (HoA and HiA) are rich in carboxyls, and that the soil has high content of reactive amorphous Fe- and Al-oxides (i.e., high $Fe_o$ and $Al_o$, shown in Table 3-2), indirectly supports the idea of ligand exchange as the predominant sorption mechanism, as amorphous Fe- and Al-oxides are known to be strong sorbents for DOC via ligand exchange (Kaiser and Zech 1998b).

My results may also suggest that DOC sorption is not simply a result of ligand exchange between carboxylic functional groups and hydroxides on clays. Because HiA-DOC has more carboxylic functional groups per molecular weight than HoA-DOC (summarized in Qualls and Haines 1991), HiA would be more strongly sorbed than HoA-DOC, if ligand exchange were the dominant sorption mechanism. In addition, Phenol and Bases showed high affinity to mineral soil in the lab study, in spite of their low carboxylic functional groups retalive to the two strong acid fractions (HoA and HiA). Perhaps, a combination of ligand exchange and hydrophobic interaction would affect the sorption of DOC compounds. The latter mechanism may be less effective to small molecular weight organic acids, such as HiA, which may not have significant hydrophobic functional groups relative to its carboxyl functional groups, but may apply to larger molecular-weight HoA, which is less degraded and would have ample hydrophobic functional groups.

In addition to the two sorption mechanisms mentioned above, coprecipitation of DOC molecules or DOC molecules and clay particles via cation bridging might also be significant. Because of the young soil developed from volcanic parent materials, fast weathering rates of the parent materials might provide enough base cations for cation bridging, such as $Ca^{2+}$ and other polyvalent cations such as $Al^{3+}$. High pH of the mineral soil (~7.0) of this study site would suggest high base cation concentrations in the soil solution.

Differences in %sorption of Phenol, Bases and HiA between the field and lab studies might be because DOC compounds that were physically extracted from litter and that were leached in the field by moving water might be different even though they are fractionated into identical chemical fractions. Soil water collected in lysimeters would contain DOC derived from belowground DOC sources, such as the metabolites of roots and the turnover of microbial biomass, that would not have been included in the litter extracts.

### Profiles of chemical fraction composition

HoA and HiA were the two dominant fractions in TF as well as the soil solutions in this study, with a shift in dominance by HoA in the O-horizon leachate to dominance by HiA in the deeper mineral soil. This is consistent with many other studies for a wide range of forest types with different climate and soil types; e.g., a hardwood forest on an Ultisol (Qualls and Haines 1991), coniferous forests on Inceptisols and Spodosols (Guggenberger et al. 1994), a hardwood forest on a Spodosol (Vance and David 1991, David et al. 1995), and a coniferous forest on a Spodosol (Dai et al. 1996). In the case of volcanic-ash derived soil, however, HiA was the most important constituent followed by HoA (Antweiler and Drever 1983).

A striking difference in chemical fraction composition between other studies and this study was the importance of Bases. Percent Bases is generally ≤ 7 % of total DOC (Qualls and Haines 1991; Guggenberger et al. 1994; Dai et al. 1996) with the exception of the study of volcanic-ash soil (6-13 %; Antweiler and Drever 1983). In this study, percent Bases was even greater (up to 19 % of total DOC) and was the third largest fraction in the deep soil water. Because DOC concentrations at the deep soil in this study site (3-8 mg/L) were within the range that has been reported for various forest and soil types (0.8-12 mg/L in the B-C horizons, reviews by Herbert and Bertsch 1995; 0.6-5 mg/L in C horizons reviewed by Michalzik et al. 2001), a larger percent Bases would indicate that more DOC that is in the form of free amino compound was lost to the deep soil in my study site than other non-volcanic soils. A higher % Bases in the soil solution in this study as well as one by Antweiler and Drever (1983) might be due to lower retention of Bases by volcanic-ash derived soil minerals. Volcanic-ash derived young soils are characterized by high amorphous clay content (e.g., high sodium-oxalate extractable Al, Table 3-1; pH in 1M NaF of 10.8 in my site; data not shown), and volcanic ash weathers faster than most non-volcanic parent materials. Because the major component of Bases is cationic amino compounds, there might be a competition for cation-exchange sites between cations produced by weathering and Bases, resulting in a lower retention of Bases in the soil. In addition, because the high weathering rates would consume $H^+$ (e.g., soil water pH of ~7.0, current study; 4.5-6.5, Antweiler and Drever, 1983), the cationinc amino compounds of Bases may be deprotonated and lose affinity to the mineral soils. These hypotheses are consistent with the insignificant net retention of Bases in spite of extremely high cation exchange capacity of soil examined in this study (~40 $\mathrm{cmol_c/kg}$ for the B horizon, data not shown).

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# CHAPTER 4
# CONCLUSION

## EFFECTS OF LITTER QUALITY ON DOM

In the laboratory study, litter type and decomposition stage had strong effects on the amount of extractable DOM and its chemistry. Newly harvested fine root litter tended to produce more DOC and DON per weight than other litter types, suggesting a potential importance of root inputs to DOM dynamics in this forest ecosystem, although the degree of reallocation of nutrients from senescing fine roots to plants is unknown. The degree of litter decomposition also had a strong influence on DOM composition: HoA increased (from 27 to 47% and from 16 to 62% of total DOC for leaf and wood litter, respectively) and HiN decreased (from 30 to 5% and from 36 to 13% of total DOC for leaf and wood litter, respectively) from the extracts of newly fallen to well-decomposed DOC sources.

In the field, however, no difference was found in the composition and concentration of the O-horizon DOM among Double Litter, Double Wood and Control treatments after 4 years of litter manipulation, in spite of significant differences in total C, total N, and C:N ratios of the O-horizon material. Because the composition of DOM was similar to that of lab-extracts of well-decomposed litter, microbial degradation in the O-horizon material possibly made the DOM composition in the O-horizon leachate similar under different DIRT treatments. Alternately, lack of difference in the composition of O-horizon leachate may be due to a greater contribution from native O-horizon material rather than from newly added litter.

## EFFECT OF DOM CHEMISTRY ON REMOVAL (SORPTION)

In both field and lab studies, HoA was strongly removed from solution. HiA was also strongly removed from solution in the field study, but less so than HoA. Strong sorption of acid fractions suggests that sorption process may be driven by ligand exchange between hydroxides of clay surface and carboxyl functional groups of DOM. The low net sorption of the HoN observed in the lab study suggests that hydrophobic interaction may not be a major mechanism of DOM sorption. In the lab study, HiA was sorbed only from extracts of well-decomposed litter. This suggests that the HiA fraction in different solutions may contain functionally distinct compounds that were not separated by my chemical fractionation method.

## PRODUCTION AND REMOVAL OF DOM

The O horizon was not always a significant source of dissolved organic C (DOC) as has been reported in many other studies. Rather, net DOC production from the shallow mineral soil (0-10 cm) was at times larger than from the O horizon. The shallow mineral soil was also the largest net source of dissolved organic N (DON), and the production of DON was not associated with the production of dissolved inorganic N.

Percent Bases (mostly cationic free amino compounds) was higher at my study site (up to 19 %) than at other sites (≤ 7 %, for soils with non-volcanic parent material; 6-13 % for soil with volcanic parent material). Because DOC concentrations in the deep soil at this study site were within ranges reported for various forest and soil types, the amount of N lost in Bases is relatively large at this study site.

The concentrations of DOC and DON in the outgoing water were constant regardless of the treatment, season, and the concentrations of incoming water for both shallow (0-30 cm) and deep (30-100 cm) soil layers. In other words, the concentrations of DOM in incoming water and outgoing water were positively correlated with a slope of ~1.0 and negative intercept, regardless of the conditions. Combined with the fact that

%HiN of O-horizon leachate was < 2%, near complete removal of DOM suggest that DOM removal was abiotic (sorption) rather than biotic (microbial respiration and/or immobilization), and that the net release of DOC and DON was constant and independent of net sorption.

## FUTURE DIRECTIONS

Current DOM process models include biological decomposition as a key factor that controls DOM chemistry and dynamics. Currie and Aber (1997) related DOC in forest-floor leachate to decomposition rates of litter, which varied by the chemical composition of litter (%lignin, %cellulose, %extractables, and %N). Neff and Asner (2001) linked biotic and abiotic processes in their DOC model. The authors also used lignin:N ratio to predict DOM chemistry. In both approaches, the underlining assumption was that the chemical composition of DOM is determined by the chemical composition of litter. However, findings in Chapter 2 contradicted their assumption: the composition of O-horizon leachate did not differ under DIRT treatments. As discussed in the chapter, microbial decomposition of the O-horizon material possibly decreased differences in DOM chemistry under different DIRT treatments. Information that links litter chemistry and DOM chemistry is necessary for modeling DOM dynamics.

Other DOM models use physical and/or chemical interactions between DOM and mineral soil surfaces to predict DOM stabilization in mineral soils (e.g., Graber and Borisover 1998; Neff et al. 2001). Generally, these DOM models use sorption and desorption parameters determined in a laboratory sorption isotherm (regressions between initial DOC concentration and sorption, equivalent to regression lines in my field study, Fig. 2-4, Table 2-6). Neff and Asner (2001) summarized these parameters (slope and intercept) reported for a wide range of soil types (ranged from 0.01 to 0.73 for 6 different soil orders). However, these parameters determined in a laboratory study might not represent sorption and desorption rates in the field. In fact, the slope of laboratory isotherm for the mineral soil adjacent to DIRT site at Andrews Experimental Forest was

0.45, nearly half of that found in my field study (~1.0, Table 2-6). Study is therefore necessary to understand sorption and desorption parameters in the field.

Soil mineralogy controls sorption and desorption processes in soils. Understanding interactions between DOM chemistry and mineralogy on sorption and desorption processes is also essential for modeling DOM dynamics at the global scale.

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