Carbon and nitrogen mineralization from light- and heavy-fraction additions to soil

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Abstract

Mineralization of C and N from soil organic matter (SOM) can be altered when natural ecosystems are transformed for food and fiber production. We examined C and N dynamics in adjacent long-term minimally disturbed and disturbed soils from agricultural and forest sites. Light and heavy fractions (LF and HF, respectively) of SOM were collected by physical density separation using sodium polytungstate. Aerobic C and N mineralization of soil (WS), soil plus HF (S+HF) and soil plus LF (S+LF) mixtures were determined. Between 0.8% and 1.7% of C and 0.3% and 1.2% of N from WS was mineralized after 28 days. The proportion of C mineralized from HF was negligible in all sites, suggesting that the HF component of soils could be a major sink for C storage in soils. Larger proportions of N from HF were mineralized in disturbed than minimally-disturbed soils, suggesting greater protection of N in the HF of disturbed soils. The proportion of C mineralized from LF ranged from \(-0.3\%\) to \(3.2\%\), and was not consistent with C mineralization dynamics from the HF component of soils. It appeared that, while the LF component of soil contained C that was chemically and, to a lesser extent, physically protected from decomposition, more C was potentially mineralizable from the LF than the HF component of the agricultural and forest soils examined. In most soils, LF additions resulted in N immobilization rather than N mineralization. Our results indicate that HF is the main source of potentially mineralizable N whereas LF is a potential sink for mineral N, regardless of land management practices, in the agricultural and forest soils we examined. © 2000 Elsevier Science Ltd. All rights reserved.

Keywords: Soil organic matter; Heavy fraction; Light fraction; N mineralization; C mineralization

1. Introduction

The quantity and rate of soil organic matter (SOM) turnover are often altered when natural ecosystems are transformed into more intensively managed production systems. It is generally accepted that SOM contains fractions with a rapid turnover rate (e.g., weeks and months) as well as fractions with a slower turnover rate (e.g., years and centuries) (Schimel et al., 1985). Differences in turnover rates of SOM fractions may be due to physical protection of organic matter within soil aggregates as well as chemical protection from humification (Cambradella and Elliott, 1992). The fractions of SOM that turn over rapidly are believed to make a greater contribution to nutrient cycling than fractions that turn over slowly because they provide a more readily accessible source of energy for the saprotrophic soil organisms responsible for nutrient cycling (Janzen et al., 1992). It is becoming increasingly important to determine not only how land management practices affect the retention or loss of these fractions of SOM, but also how they affect nutrient cycling from SOM fractions. Improved understanding of these processes will provide valu-
able information for maintaining or implementing environmentally-sustainable land management practices in agricultural and forest soils.

Although models of SOM dynamics, such as the Century model, provide a useful theoretical framework to evaluate SOM dynamics (Parton et al., 1987), analytical methods do not exist to separate SOM fractions based on their relative turnover time. Physical fractionation of soils using density separation may help distinguish SOM dynamics based on turnover time because of the possibility of separating newly incorporated, partially decomposed plant debris (light fraction) from organic matter adsorbed on mineral surfaces or sequestered within soil aggregates (heavy fraction) (Sollins et al., 1984; Christensen, 1992). The light fraction (LF) of SOM has a wider C-to-N ratio than the heavy fraction (HF), and tends to decompose more quickly than the HF, although the relative contribution of LF and HF to N mineralization varies with soil texture and land use (Strickland and Sollins, 1987; Hassink, 1995; Barrios et al., 1996).

Dynamics of the LF pool can be related to changes in total SOM, and it has been suggested that LF may be a labile source of mineralizable C and N. In cultivated agricultural soils, 1–25% of SOM may be composed of LF organic matter (Greenland and Ford 1964; Janzen et al., 1992; Boone, 1994), whereas SOM in forest soils may contain as much as 63% of LF organic matter (Sollins et al., 1983, 1984; Strickland and Sollins, 1987; Boone, 1994). Mineralization of C and N from LF has been correlated with the C and N content of LF material (Janzen et al., 1992; Hassink, 1995; Barrios et al., 1996) and with active microbial biomass (Hassink, 1995). In most studies, however, LF has been shown to contribute less to net N mineralization than HF mainly because it comprises a relatively small portion of total SOM (Sollins et al., 1984; Strickland and Sollins, 1987; Boone, 1994). In addition, N mineralization from LF varies temporally due to the seasonality of litter inputs to the LF pool (Boone, 1994).

Our purpose was to determine how C and N dynamics from whole soil (WS) and isolated HF and LF are influenced by different management practices in agricultural and forest sites. Samples were obtained from cultivated, cropped and undisturbed, grass-covered plots at two agricultural sites, and from clear-cut and old-growth Douglas fir plots at the forest site. Net C and N mineralization from WS and WS with added HF (S + HF) or LF (S + LF) were measured. The proportion of C and N from WS, HF and LF that was potentially mineralizable was calculated for agricultural and forest soils with different management practices.

## 2. Materials and methods

### 2.1. Soil

Soil samples were collected from the upper layers (0–15 cm) of three long-term experimental sites in Oregon, USA in June 1997 (Table 1). One of the sites was at the Columbia Basin Agricultural Research Center near Pendleton in eastern Oregon, USA, a region characterized by a semi-arid Mediterranean climate (mean annual precipitation of approximately 400 mm/year). The other two sites were in western Oregon, USA, a region characterized by a humid Mediterranean climate. One of the sites was located in the Willamette Valley at the North Willamette Research Extension Center (mean annual precipitation of approximately 1200 mm/year) and the other in the Cascade Mountain range at the H.J. Andrews Experimental Forest (elevation of 850 m, mean annual precipitation of approximately 2150 mm/year). At each site, soil was collected from replicated field plots that have a history of management-induced disturbance or minimal-disturbance, including wheat–fallow and unimproved grassland plots (Columbia Basin), annually cropped vegetable and fescue-covered plots (North Willamette), and replanted and old-growth forest plots (H.J. Andrews) (Table 1). Field-moist soils from replicated plots were composited, sieved (1.25 cm mesh), and stored at 4°C until analysis. Soil water contents at the time of sampling were 8% and 5% (w/w) for the prairie and wheat treatments, 6% and 5% for the fescue sod and vegetable treatments, and 13% and 22% for the old-growth and replanted forest treatments. Unless otherwise indicated, soil weights are expressed on an oven-dry (105°C for 48 h) basis.

### 2.2. Soil organic matter fractionation

Soil organic matter was separated by density into LF and HF following the procedure of Swanston and Myrold (1997) modified from Strickland and Sollins (1987). Briefly, 100 g of coarsely sieved (<1.25 cm mesh) soil was mixed with 100 ml of sodium polytungstate (SPT) solution (density = 1.6 g cm⁻³; Sometu-US, Van Nuys, CA, USA) for 30 s in a soil mixer at 4000 × 9 g to disrupt soil aggregates. Soil aggregates were then dispersed for 2 min with a probe-type sonic disrupter (Model 350, Branson Sonic Power Company, Danbury, CT, USA) using an energy input of 2000 J. After settling overnight, suspended LF material and the upper 1 cm of SPT solution were aspirated, transferred to Whatman No. 1 filter paper, and rinsed thoroughly with distilled, deionized water. Soil was re-suspended in an additional 100 ml of SPT solution, mixed thoroughly, and allowed to settle overnight. Light fraction material and SPT solution were aspi-
rated, rinsed thoroughly, and added to previously collected LF. The remaining HF was rinsed several times to remove SPT by mixing with distilled, deionized water, settling overnight, and aspirating the supernatant. Light and heavy fractions and whole soil were oven-dried (105°C for 48 h), ground, weighed, and analyzed for total C and N using a Carlo-Erba C and N analyzer (Milano, Italy) (Table 2).

2.3. Aerobic mineralization study

Soil C and N mineralization was determined on 20 g (oven-dry basis, sieved (<2 mm mesh)) WS, 15 g WS mixed with 5 g (oven-dry) HF (S+HF), or 19.5 g WS mixed with 0.5 g (oven-dry) LF (S+LF). Amounts of HF and LF added to WS were chosen to provide sufficient C so that quantities of C and N mineralized from HF and LF could be detected. The total C content of S+HF mixtures was 4–9% lower than WS due to the lower C content of HF than WS, and the total C content of S+LF mixtures was 7–37% greater than WS, depending on the treatment. The quantity of LF added to soil was constrained by the limited quantity of LF collected from SOM fractionation. The WS, S+HF, and S+LF mixtures were moistened to 30% (w/w) gravimetric water content, placed immediately in sealed 950-ml jars, and incubated in the dark at 25 ± 1°C for 28 days. Approximately 10 ml of water was placed in the bottom of each jar to maintain soil humidity. Jar lids were equipped with an air-tight rubber septum for measurement of soil respiration. Jar headspace was sampled every seventh day and, after sampling, jars were aerated by removing lids for 10–15 min to re-establish ambient conditions. Gas samples were injected into vacutainers for storage and CO2–C production was measured with a Hach Carle Series 100 gas chromatograph equipped with a thermal conductivity detector (Loveland, CO, USA). Net C mineralization was the cumulative mg CO2–C produced during the 28-day incubation per gram of C in the WS, S+HF or S+LF mixture. Calculations of the proportion of C mineralized from WS, HF or LF are outlined in Table 3.

Extractable soil C (dissolved organic C, microbial biomass C) and soil N (NH4+-N, NO3–N, dissolved organic N, microbial biomass N) concentrations were determined on non-incubated samples and 28 days after the aerobic incubation study was initiated. All analyses were run on triplicate 10 g samples of non-incubated and incubated whole soil and soil–SOM mixtures. Dissolved organic C (DOC) was measured in 0.5 M K2SO4 extracts (1:5, soil:extractant) using a
## Table 3
Proportion of total C and N in WS, HF, and LF mineralized during 28-day incubation

<table>
<thead>
<tr>
<th>Site</th>
<th>Land management</th>
<th>Treatment</th>
<th>Incubated soil (g C)</th>
<th>Incubated SOM (g C)</th>
<th>C mineralized</th>
<th>Soil C_{min}</th>
<th>SOM C_{min}</th>
<th>% C_{min} (%)</th>
<th>% N_{min} (%)</th>
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</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>mg CO_2–C g^{-1}</td>
<td>mg CO_2–C</td>
<td>(mg CO_2–C)</td>
<td>(mg CO_2–C)</td>
<td>WS</td>
</tr>
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<td>Columbia Basin</td>
<td>Prairie</td>
<td>WS</td>
<td>0.53</td>
<td>18.4</td>
<td>9.8</td>
<td>9.8</td>
<td>1.8</td>
<td>−3.0</td>
<td>0.4</td>
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<tr>
<td></td>
<td></td>
<td>S + HF</td>
<td>0.40</td>
<td>9.2</td>
<td>4.4</td>
<td>7.4</td>
<td>−3.0</td>
<td>1.7</td>
<td>−1.2</td>
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<tr>
<td></td>
<td></td>
<td>S + LF</td>
<td>0.52</td>
<td>15.6</td>
<td>10.1</td>
<td>9.6</td>
<td>0.5</td>
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<td></td>
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<tr>
<td></td>
<td>Wheat</td>
<td>WS</td>
<td>0.34</td>
<td>17.3</td>
<td>5.9</td>
<td>5.9</td>
<td>1.7</td>
<td>−1.2</td>
<td>3.2</td>
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<tr>
<td></td>
<td></td>
<td>S + HF</td>
<td>0.25</td>
<td>11.6</td>
<td>3.6</td>
<td>4.3</td>
<td>−0.7</td>
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<tr>
<td></td>
<td></td>
<td>S + LF</td>
<td>0.33</td>
<td>21.0</td>
<td>9.5</td>
<td>5.7</td>
<td>3.8</td>
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<td></td>
</tr>
<tr>
<td></td>
<td>Wheat</td>
<td>WS</td>
<td>0.36</td>
<td>13.2</td>
<td>4.8</td>
<td>4.8</td>
<td>1.3</td>
<td>−0.7</td>
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<td></td>
<td></td>
<td>S + HF</td>
<td>0.27</td>
<td>9.0</td>
<td>3.1</td>
<td>3.6</td>
<td>−0.5</td>
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<tr>
<td></td>
<td></td>
<td>S + LF</td>
<td>0.35</td>
<td>10.5</td>
<td>5.3</td>
<td>4.7</td>
<td>0.6</td>
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<tr>
<td></td>
<td>Vegetable</td>
<td>WS</td>
<td>0.32</td>
<td>8.5</td>
<td>2.7</td>
<td>2.7</td>
<td>0.8</td>
<td>2.4</td>
<td>−0.2</td>
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<tr>
<td></td>
<td></td>
<td>S + HF</td>
<td>0.24</td>
<td>12.0</td>
<td>3.7</td>
<td>2.0</td>
<td>1.7</td>
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<tr>
<td></td>
<td></td>
<td>S + LF</td>
<td>0.31</td>
<td>5.5</td>
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<td>2.6</td>
<td>−0.2</td>
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<td></td>
<td>North Fescue sod</td>
<td>WS</td>
<td>0.87</td>
<td>10.7</td>
<td>9.3</td>
<td>9.3</td>
<td>1.1</td>
<td>−1.3</td>
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<td></td>
<td></td>
<td>S + HF</td>
<td>0.65</td>
<td>6.1</td>
<td>4.9</td>
<td>6.9</td>
<td>−2.0</td>
<td></td>
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<td></td>
<td>S + LF</td>
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<td>10.2</td>
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<td>9.0</td>
<td>1.0</td>
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<td></td>
</tr>
<tr>
<td></td>
<td>H.J. Andrews Old-growth</td>
<td>WS</td>
<td>1.62</td>
<td>7.7</td>
<td>12.5</td>
<td>12.5</td>
<td>0.8</td>
<td>−0.4</td>
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<tr>
<td></td>
<td></td>
<td>S + HF</td>
<td>1.21</td>
<td>5.4</td>
<td>8.2</td>
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<td>−1.1</td>
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<tr>
<td></td>
<td></td>
<td>S + LF</td>
<td>1.58</td>
<td>6.7</td>
<td>11.7</td>
<td>12.2</td>
<td>−0.5</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* Quantity of C mineralized from soil in WS, S + HF and S + LF mixtures; Soil C_{min} = [(mg CO_2–C mineralized)_{WS} × (g soil C)_{S + SOM mixture}]/(g soil C)_{WS}.

* Quantity of C mineralized from HF or LF; SOM C_{min} = [(mg CO_2–C mineralized)_{S + SOM mixture} − (mg CO_2–C mineralized)_{WS}]/(g soil C)_{WS}.

* Proportion of total C in WS, HF or LF mineralized during incubation; %C_{min} = [(Soil C_{min})/1000]/(g soil C) × 100; %C_{min} = [(Soil C_{min})/1000]/(g soil C) × 100.

* Proportion of total N in WS, HF or LF mineralized during incubation. Values for incubated soil (g N), incubated SOM (g N) and N mineralization (mg N g^{-1} N) were substituted into the equations above to calculate % N_{min}.
Dohrman DC-190 carbon analyzer (Rosemount Analytical, Santa Clara, CA, USA). Microbial biomass C (MBC) was determined using the chloroform fumigation–direct extraction method and calculated as: (total extractable C after fumigation – total extractable C before fumigation)/0.45 (Joergensen, 1996).

Inorganic N (NH₄⁺–N, NO₃⁻–N) was determined in 0.5 M K₂SO₄ soil extracts (1:5 soil:extractant) colorimetrically using the modified indophenol blue technique (Sims et al., 1995) and measured at 650 nm using Titertek Multiscan MCC/340 automated microplate reader (Huntsville, AL, USA). Dissolved organic N (DON) was calculated as the difference between the NO₃⁻–N concentration in an alkaline persulfate digestion of the soil extract and the inorganic N (NH₄⁺ and NO₃⁻) concentration of the soil extract (Cabrera and Beare, 1993). Microbial biomass N (MBN) was determined using the chloroform fumigation–direct extraction method followed by persulfate digestion (Brookes et al., 1985) and calculated as: (total extractable N after fumigation – total extractable N before fumigation)/0.54 (Joergensen and Mueller, 1996). Net N mineralization was the difference in inorganic N (NH₄⁺–N + NO₃⁻–N) between incubated (28 days) and non-incubated samples. Net N mineralization was the mg of inorganic N mineralized during incubation per gram of N in WS, S+HF, or S+LF mixtures. The proportion of N mineralized from WS, HF or LF was calculated by substituting values for incubated soil (g N), incubated SOM (g N) and N mineralization (mg N g⁻¹ N) in the equations outlined in Table 3.

2.4. Statistical analysis

Data were log transformed to equalize variance and evaluated statistically by ANOVA in a general linear model (GLM) using SAS software (SAS Institute, 1990). Nutrient dynamics in WS, S+HF and S+LF treatments were evaluated using a randomized complete block design where the random variables were the six field plots (blocks) from which composite soil samples were collected. Treatment means were compared statistically with an LSD test at the 95% confidence level (Steel and Torrie, 1980).

3. Results

Concentrations of extractable C and N in soils collected from the Columbia Basin, North Willamette, and H.J. Andrews sites were assessed before the mineralization study began (Table 2). The extractable C and N content of WS, S+HF and S+LF treatments did not differ significantly, and values presented in Table 2 are averaged across these treatments. The C to N ratios of dissolved organic matter ranged from 2.4 to 9.1 and tended to be lower in disturbed than minimally-disturbed soils. The DOC pool contained 0.5–0.7% of total soil C and DON pool contained between 1.8% and 3.4% of total soil N for the sites examined. The C-to-N ratio of microbial biomass was greater in disturbed than minimally-disturbed soils from the Columbia Basin and H.J. Andrews sites, and lower in disturbed than minimally-disturbed soils from the North Willamette site. In the agricultural sites, the MBC pool contained 1.5–3.2% of total soil C, while in the forest site, about 0.5% of total soil C was in microbial biomass. In agricultural and forest sites, between 1.6% and 3.3% of the total soil N was in the MBN pool.

The C content of WS from the forest sites was greater than the agricultural sites, regardless of site history (Table 2). Soil C-to-N ratios were 24 and 32 for old-growth and replanted forest sites and between 13 and 17 for agricultural sites. The C and N content of HF tended to be lower than WS and the C-to-N ratios of HF were similar to the C-to-N ratios of WS. The LF had 10–16 times more C than WS from agricultural sites and 4–6 times more C than WS from forest sites (Table 2). There was 2–5 times more N in LF...
than WS from agricultural sites. The C-to-N ratios of LF ranged from 40 to 105 in agricultural and forest sites (Table 2).

3.1. Carbon mineralization

Cumulative C mineralization during soil incubation was often lower in WS and soil–SOM mixtures from the H.J. Andrews site than the Columbia Basin and North Willamette sites (Fig. 1). C mineralization was significantly \( P < 0.05, \text{LSD} \) greater in WS and S+LF than the S+HF treatment in disturbed soil from the Columbia Basin site (Fig. 1(a)), but C mineralization did not differ among treatments in disturbed soils from the North Willamette and H.J. Andrews sites (Fig. 1(b) and (c)). Significantly \( P < 0.05, \text{LSD} \) less C was mineralized from S+HF than WS in both minimally-disturbed soils from agricultural and forest sites, but there was no difference in the quantity of C mineralized from S+LF and WS for any site (Fig. 1(a)–(c)). In agricultural sites, between 0.8% and 1.8% of soil C was respired from WS, while in the forest sites, 0.8–1.1% of soil C was respired from WS during incubation (Table 3). The amount of C mineralized from HF was negligible for most sites examined, while −0.3 to 3.2% of the C in LF was mineralized during the 28-day incubation (Table 3).

3.2. Net N mineralization

Net N mineralization tended to be greater in minimally-disturbed WS than disturbed WS in agricultural and forest sites (Fig. 2). In minimally-disturbed agricultural and forest sites, between 1.1% and 1.2% of soil N was mineralized from WS, whereas 0.3% to 0.5% of soil N was mineralized from WS in disturbed sites (Table 3). Net N mineralization from S + HF did not differ significantly from WS for any site examined (Fig. 2(a)–(c)). Larger proportions of N from HF were mineralized in S + HF mixtures from disturbed than minimally-disturbed soils (Table 3). In most treatments examined, N mineralization was depressed in the S + LF treatment relative to WS (Fig. 2(a)–(c)), but was significantly lower \( P < 0.05 \) than WS in S + LF mixtures from the prairie and fescue sod treatments (Fig. 2(a) and (b)). In the agricultural sites, the quantity of N mineralized from LF ranged from −4.7% to −19.6%, indicating N immobilization in S + LF mixtures (Table 3).

4. Discussion

The C-to-N ratio of LF in agricultural soils has been reported to range from 13 to 36 (Janzen et al. 1992; Bremmer et al. 1994; Hassink 1995). In forest soils, the C-to-N ratio of LF may be considerably higher, and values of 24–86 have been reported (Strickland and Sollins, 1987; Swanston and Myrold, 1998). The C-to-N ratios of the LF collected in this study are greater than those reported previously, and may be due to our method of collecting LF. In other studies that we are aware of, soils were finely sieved (<2 mm) prior to density fractionation. However, we considered large organic debris in coarsely-sieved soils (<1.25 cm) to be a component of LF because, by definition, LF consists of newly incorporated, partially-decomposed plant residues. The C-to-N ratio of LF was three- to seven-fold higher than WS from the agricultural sites and 3-fold higher than WS from the forest sites.

4.1. Carbon mineralization from soil and SOM fractions

The initial C-to-N ratio of WS and soluble organic matter were two- to three-fold higher in forest than agricultural soils, suggesting differences in SOM quality among the sites. C mineralization tended to be greater in WS from minimally-disturbed than disturbed plots in the agricultural and forest sites, but relatively small proportions of soil C were mineralized during
the study. Only 0.8–1.8% of soil C was mineralized, which is considerably lower than the 3–21% of soil organic C mineralized during laboratory incubation that has been reported (Hassink, 1994; Franzluebbers et al., 1996). The MBC pool in WS declined during incubation (data not shown), suggesting insufficient quantities of readily-available C substrates for maintenance of microbial biomass.

Carbon mineralization was lower in the S+HF treatment than WS from minimally-disturbed sites and from the wheat plots at the Columbia Basin site, and the proportion of C mineralized from HF during incubation was negligible for most sites. While the C-to-N ratios of WS and HF are similar, the low mineralization of C from HF suggests differences in the quality of SOM in WS and HF. The HF component of soils may contain C that is chemically protected from mineralization, which suggests that the HF could be a major sink for C storage in soils.

Carbon mineralization was not affected significantly by the addition of LF relative to WS at any site examined. The proportion of C mineralized from LF was greater from wheat than prairie treatments of the Columbia Basin site. Since the quality of SOM from these two sites is similar (C-to-N ratios of LF are between 40 and 49), these results suggest that LF would be more physically protected in the disturbed than minimally-disturbed plots at this site. In the North Willamette and H.J. Andrews sites, the proportion of C mineralized from LF was greater in the minimally-disturbed than disturbed plots. The C-to-N ratios of LF were greater in disturbed than minimally-disturbed plots from these sites, suggesting more recalcitrant C in LF from soils of disturbed than minimally-disturbed plots. While the LF component of soils may be chemically and, to a lesser extent, physically protected from decomposition, it tends to contain more C that is potentially mineralizable than the HF component of soils.

4.2. Net N mineralization from soil and SOM fractions

In agricultural and forest sites, N mineralization tended to be lower in WS from disturbed than minimally-disturbed plots. N mineralized from HF during a 7-day anaerobic incubation in forest soils has been found to range from 2.0% to 3.5% (Sollins et al., 1984; Barrios et al., 1996; Imhof et al., 1996), and it has been calculated that N from the LF component contributes between 2% and 13% of soil N mineralization (Boone, 1994). In agroecosystems, LF likely has a minor role in N mineralization because LF often represents a small fraction of total OM (<25%), although in forest systems, where a greater proportion of total OM may be soil-free plant material, LF may make a greater contribution to nutrient cycling. The LF collected in this study had a higher C-to-N ratio than LF collected in other studies, however, which we believe to be due to the size of soil-free macroorganic matter we included in the LF pool. In natural systems where LF consists of newly-incorporated to partially-decomposed plant residues and macroorganic matter, it seems likely that net N immobilization rather than mineralization would be the initial fate of N in the LF component of soils.

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