Initial effects of fire and mechanical thinning on soil enzyme activity and nitrogen transformations in eight North American forest ecosystems

Ralph E.J. Boerner*, Carla Giai, Jianjun Huang, Jessica R. Miesel

Department of Evolution, Ecology and Organismal Biology, Ohio State University, 318 West 12th Avenue, Columbus, OH 43210, USA

A R T I C L E   I N F O

Article history:
Received 30 May 2008
Received in revised form 19 August 2008
Accepted 7 September 2008
Available online 12 October 2008

Keywords:
Fire
N mineralization
Net nitrification
Soil enzymes
Restoration

A B S T R A C T

This study assessed the first-year effect of three ecosystem restoration treatments (prescribed fire, mechanical thinning, and their combination) on soil enzyme activity, soil N transformations, and C:N ratios of soil organic matter and mineral soil in eight North American forested ecosystems. The ecosystems we studied were part of the larger Fire and Fire Surrogate (FFS) network, and all had a history of frequent fire that has been altered by almost a century of organized fire suppression. Across all eight sites there were no statistically significant effects of the three manipulative treatments on phosphatase activity or chitinase activity; in contrast, at the network-scale phenol oxidase activity was reduced by fire alone, relative to the control. There was no significant network-scale effect of the three treatments on net N mineralization or net nitrification. Soil C:N ratio increased modestly after mechanical thinning, but not after prescribed fire or the combination of fire and thinning. There was a statistically significant reduction in forest floor C:N ratio as a result of all three treatments. Ordination of the differences between the treated and control areas indicated that fire alone resulted in greater changes in phenol oxidase activity and net nitrification than did the other two treatments. Large-scale restoration treatments such as those utilized in this study produce modest proximate effects on soil microbial activity and N transformations.

1. Introduction

Syntheses of the literature on the effects of fire on soil properties that developed over the last half century (e.g. Ice et al., 2004; Certini, 2005; Boerner, 2006) suggest that the effects of fire vary considerably among forest ecosystem types, in relation to fire severity, and over time. The diversity of ecosystem types, soil types, fires, response parameters, and experimental designs is, in fact, so daunting that developing generalizations that are useful at the local management scale is problematical.

This is also the case for restoration-oriented mechanical treatments (e.g. tree thinning or shrub layer manipulation). Although there exists a considerable literature on the impact of high intensity timber harvest (e.g. clear-cutting and whole-tree harvesting) on forest floor and soil properties (e.g. Matson and Vitousek, 1981; Rummer et al., 1997; Klepac et al., 1999), little is known of the effects of mechanical treatments at intensities useful for ecosystem restoration and wildfire hazard mitigation. The generalizations that have been developed from the clear-cutting literature seem unlikely to be applicable to the more modest mechanical treatments involved in restoration efforts.

The Fire and Fire Surrogate Network (FFS) project was established in 2000 to evaluate the efficacy of four alternative management strategies for simultaneously reducing wildfire hazards and increasing forest ecosystem sustainability, using a common experimental design in 12 forested ecosystems ranging from California and Washington in the west to Ohio and Florida in the east. All of the forested ecosystems represented in the FFS network shared a history of frequent, low severity fires over millennia, which was altered by the organized fire suppression that began in the early 20th century (Harmon, 1982; Sutherland, 1997). As such, the FFS network presented a unique opportunity to examine commonalities and differences in the effects of fire and mechanical treatments on forests that differ in vegetation, soil characteristics, and macroclimate (McIver et al., 2008). Using the common experimental design and suite of response variables of the Fire and Fire Surrogate Network project, we sought to test the following hypotheses: (1) fire will result in decreased forest floor C:N ratio as the result of combustion of the more recalcitrant components, but soil C:N ratio will change little. Any changes in soil microbial activity (measured as soil enzyme activity), N mineralization, or net nitrification will be the result of changes in microclimate, not soil organic matter quality. (2) Mechanical treatment will result in increased forest floor C:N ratio as the result of the deposition of logging slash. Despite the increase in forest floor C:N ratio, microbial activity and N mineralization in the soil will increase as the
result of changes in surface microclimate following removal of parts of the tree and shrub canopies, and net nitrification will increase as the result of reduced plant demand for N. (3) The combination of fire and mechanical treatment will produce effects on C:N ratios that parallel those of fire alone. Microbial activity and N transformations will increase more from the combined treatment than from each of the treatments individually.

2. Methods

2.1. Study sites and sampling methods

This study employed eight of the twelve FFS study sites, ranging from California to South Carolina (Table 1). The eight sites were underlain by a range of soil types, of which Alfisols, Ultisols, and Inceptisols were the most prevalent (Table 1). Molisols were present in one site and Entisols in two. Soil textures were dominated by loams and silt loams, though sands and fine sands were present in one site (Table 1). Complete descriptions of the vegetation, climate, and soils of the study sites are given by McIver et al. (2008).

Although the designated FFS experimental design was a randomized complete block with three blocks and four treatments (control, spring prescribed fire, mechanical treatment, and the combination of mechanical treatment and prescribed fire) allocated at random to those three blocks, two of the FFS sites used in this study (CaN and CaS) implemented completely randomized designs because of local logistics, and one site (OR) established four replicates of each treatment in a completely randomized design. CaS tested only the prescribed fire treatment as it was located in one of the U.S. National Parks, which do not permit mechanical treatments. We used change in aboveground biomass and surface fuels (downed woody debris + forest floor) to assess the impact of the three treatments at each site (Table 2).

Each treatment unit consisted of a minimum of 10 ha with a buffer zone of at least 4 ha surrounding it. Both the treatment unit and the buffer received the experimental treatment designated for that unit. Among the 12 FFS sites, the treatment units ranged in size from those approximating the minimum 14 ha to as large as 80 ha.

In the designated FFS design, each treatment unit was overlain with a 50 m grid and contained a minimum of 36 grid points. Ten 0.1-ha rectangular permanent sampling plots were located at random within each treatment unit, with one corner of each plot located at a grid point. Vegetation components, soil, and fuels were sampled during the pre-treatment growing season and the growing season immediately following the completion of the treatment implementation. Two sites (CaN and OR) had incomplete pre-treatment sampling because their units and treatments had been installed before the designated FFS design was established, and logistic constraints resulted in pre-treatment sampling being incomplete in one additional site (CaS).

Table 1

<table>
<thead>
<tr>
<th>Study site</th>
<th>Location</th>
<th>Code</th>
<th>Soil orders</th>
<th>Predominant soil series</th>
<th>Soil Textural Class (es)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Southern Cascades Range, CA</td>
<td>Goosenest Adaptive Management Area</td>
<td>CA-N</td>
<td>Alfisols, Entisols</td>
<td>Belzor, Wintoner</td>
<td>Loam, silt loam</td>
</tr>
<tr>
<td>Central Sierra Nevada, CA</td>
<td>Sequoia-Kings Canyon National Park</td>
<td>CA-S</td>
<td>Inceptisols</td>
<td>Unknown</td>
<td>Loamy sand, sandy loam</td>
</tr>
<tr>
<td>Blue Mountains, OR</td>
<td>Hungry Bob (Wallowa-Whitman National Forest)</td>
<td>OR</td>
<td>Molisols, Inceptisols</td>
<td>Fivebit, Melhorn, Blocker</td>
<td>Clay loam, loam, silt loam</td>
</tr>
<tr>
<td>Northern Rocky Mountains, MT</td>
<td>Lubrecht Experimental Forest</td>
<td>MT</td>
<td>Alfisols, Inceptisols</td>
<td>Winkler, Greenough, Tevis-Mitten</td>
<td>Silt loam, loam, sandy loam</td>
</tr>
<tr>
<td>Southwestern Plateau, AZ</td>
<td>Coconino and Kaibab National Forest</td>
<td>AZ</td>
<td>Alfisols, Molisols</td>
<td>Unknown</td>
<td>Silty clay loam, clay loam, silty clay, clay</td>
</tr>
<tr>
<td>Central Appalachian Plateau, OH</td>
<td>Tar Hollow &amp; Zaleski State Forests, Vinton Furnace Experimental Forest</td>
<td>OH</td>
<td>Alfisols, Inceptisols</td>
<td>Steins, Gilpin, Shelbota</td>
<td>Silt loam, loam, sandy loam</td>
</tr>
<tr>
<td>Southeastern Piedmont, SC</td>
<td>Clemson Forest</td>
<td>SC</td>
<td>Ultisols, Inceptisols</td>
<td>Cecil, Madison, Pecolet</td>
<td>Sandy loam, clay loam</td>
</tr>
<tr>
<td>Southern Appalachians, NC</td>
<td>Green River Game Lands</td>
<td>NC</td>
<td>Ultisols, Inceptisols</td>
<td>Evard, Cowee, Clifffield</td>
<td>Loam, silt loam</td>
</tr>
</tbody>
</table>

Soils were sampled near the corners of the permanent sampling plots or in a subplot established within the permanent sampling plot. A range of 2–6 samples were taken in or around each permanent sampling plot, depending on site, yielding a total of 240–720 samples per site during the pre-treatment and post-treatment years. For the incubations used to measure N mineralization and net nitrification, samples were taken typically in June, whereas samples for enzyme activity were taken in August. Samples were kept refrigerated until analyzed, including during shipping from the field sites to the laboratory. All samples were analyzed within 5 days of arrival in the laboratory, and within 3 weeks of the sampling date, depending on site and shipping time. The organic (∆) horizon, if present, was removed prior to sampling the mineral soil. If no distinct boundary occurred between the Aa subordinate organic horizon (i.e. humic subhorizon) and the A master horizon, the Aa was included in the sampling of the surface mineral soil. In sites where a distinct horizon boundary between the A and E occurred, samples were taken either for the full depth of the A horizon or to 15 cm, whichever was less.

Organic horizons (i.e. forest floors) were sampled by clearing the forest floor around an area of 0.25 m² and removing separately each discernable organic subordinate horizon (i.e. Oi, Oe, and Oa) in that exposed 0.25 m² area.

Net nitrogen (N) mineralization and net nitrification rates were estimated from 25 to 35 days in situ incubations utilizing either the buried polyethylene bag method (Eno, 1960) or paired, intact soil cores (Raison et al., 1987).

2.2. Laboratory methods

Subsamples were passed through a 2 mm sieve to remove stones and root fragments, extracted with 0.5 M K₂SO₄ or 1.0 M KCl, then analyzed for NH₄⁺ and NO₃⁻ using the microtiter methods of Sims et al. (1995). Total inorganic N (hereafter TIN) was defined as NH₄⁺-N and NO₃⁻-N. Net N mineralization was defined as the difference in TIN between individual pre-incubation samples and TIN in either the corresponding post-incubation sample where N = 1 or the mean of the post-incubation samples where N > 1. Net nitrification was defined as the difference in NO₃⁻-N in pre-incubation and post-incubation samples. Proportional net nitrification was calculated by dividing net nitrification by the total NH₄⁺-N available to be nitrified (initial NH₄⁺-N + net N mineralized). Forest floor and soil organic carbon (C) and total N were analyzed by microDumas oxidation (Sollins et al., 1999).

For enzyme analysis, additional subsamples were passed through a 2 mm sieve to remove stones, root fragments, and particulate organic materials (i.e. remnants of the Oi, Oe, and then...
analyzed for the activity of phosphatase, chitinase, and phenol oxidase. As all roots were removed prior to analysis, enzyme activities represent only microbial contributions.

Phosphatase (EC 3.1.3.1) was chosen as an indicator of overall microbial activity as phosphatase activity is strongly correlated with microbial biomass (Kandeler and Eder, 1993; Clarholm, 1993), fungal hyphal length (Häussling and Marschner, 1989) and N mineralization (Decker et al., 1999). Chitinases sensu lato are a series of bacterial enzymes that catalyze the breakdown of chitin, a by-product of the death of both fungi and arthropods, into carbohydrates and inorganic N. The specific member of this suite of enzymes we assayed was β-1,4-N-acetylglucosaminidase (EC 3.2.1.14). As chitin is intermediate in its resistance to microbial metabolism, its synthesis is only induced when other, more labile C and N sources are absent (Hanzlíková and Jandera, 1993). Chitinases are produced principally by bacteria; thus changes in chitinase activity relative to that of other enzymes give an indication both of changes in the relative contribution of chitinolytic bacteria to microbial activity as well as changes in organic matter along the gradient from labile to recalcitrant. Phenol oxidase (EC 1.14.18.1, 1.10.3.2) is produced primarily by white rot fungi, and is specific for highly recalcitrant organic matter, such as lignin (Carlile and Watkinson, 1994). Increases in phenol oxidase activity relative to other enzymes gives another indication of changes in the relative contribution of bacteria and fungi to microbial activity as well as an additional indication of the quality of the organic matter present. Thus, as a group these three enzymes supplied insight into changes in both the microbial community and the organic matter complex. Enzyme activities were determined on field-moist soil using modifications of the standard methods developed by Tabatabai (1982). Sieved subsamples of fresh soil were suspended in 50 mM NaOAc buffer at pH 5.0 for the acidic soils of the eastern sites or NaHCO₃ buffer at pH 8.2 for the circumneutral soils of the western sites, and homogenized by rapid mechanical stirring. To minimize sand sedimentation, stirring was continued while aliquots were withdrawn for analysis.

For all sites except AZ and MT, phosphatase and chitinase activities in soil suspensions were determined using p-nitrophenol (pNP)-linked substrates. Following incubation, samples were centrifuged at 3000g for 3 min to precipitate particulates. An aliquot of 2.0 ml of the supernatant was transferred to a clean, sterile tube, and 0.1 ml of 1.0 M NaOH was added to halt enzymatic activity and facilitate color development. Prior to spectrophotometric analysis at 410 nm each sample of the supernatant was diluted with 8.0 ml of distilled, deionized water. Phosphatase and chitinase activities in samples from AZ and MT were measured in microplate wells using methylumbelliferyl (MUB)-linked substrates. Excitation was measured at 360 nm and emission at 450 nm.

Phenol oxidase (EC 1.14.18.1, 1.10.3.2) activity in soil suspensions was measured by oxidation of l-DOPA (3,4-dihydroxyphenylalanine). In all sites except AZ and MT, 1 h incubations at 20–22 °C were followed by centrifugation as above and analyzed at 460 nm without dilution. Parallel oxidations using standard horseradish peroxidase (Sigma Chemical) were used to calculate the l-DOPA extinction coefficient. In MT and AZ, absorbance was determined at 460 nm every 5 min for 1 h to determine the rate of degradation of l-DOPA.

### 2.3. Data analysis

All response variables were normally distributed. As four of the eight sites utilized randomized complete block designs and four utilized completely randomized designs, pooled responses across the eight sites were analyzed by mixed model analysis of variance for a completely randomized design with sites, treatments, and units within treatments as main effects (SAS, 1995). For individual sites, the main effects were treatment, units within treatments, and plots within units. Treatment units were considered as the unit of replication, with N = 3 (4 for OR) per treatment per site. The criterion for significance was p < 0.05.

Although all sites were expected to take both pre-treatment and post-treatment samples, only four of the eight sites generated full pre-treatment data sets. To determine whether the lack of pre-treatment data was likely to produce type I errors, we did a preliminary analysis of the data from the four sites that did have complete pre-treatment data. This involved first analyzing the post-treatment responses at those four sites by analysis of variance, as indicated above, and then again by analysis of covariance with the pre-treatment conditions as covariates (Table 3). The magnitude and significance of the main treatment effects in the analyses with the pre-treatment conditions included as covariates (ANCO-VAs) and those that included only the post-treatment data (ANOVAs) differed little, and <20% of the covariates were significant. Thus, the significance of the main post-treatment effects was relatively insensitive to the presence or absence of pre-treatment conditions.
data, at least for the four sites we could evaluate in this manner. In addition, meta-analysis of a broad range of pre-treatment soil parameters among all 12 FFS sites demonstrated no significant differences among treatment units within sites prior to treatment (Boerner et al., in press). This indicated to us that the lack of pre-treatment data from four of eight sites was not likely to lead to Type I errors.

3. Results

3.1. Soil enzyme activity

When the eight sites were pooled, there were no significant differences in phosphatase activity among treatments or sites (Table 4). In contrast, phosphatase activity was affected significantly by the treatments in five of the eight individual sites (Table 5). Among the five western sites, fire resulted in reduced phosphatase activity in six of eleven possible combinations of site and fire or fire + mechanical treatment, and in OR the fire and mechanical + fire treatments had lower phosphatase activity than did the mechanical treatment (Fig. 1). In addition, mechanical treatment alone resulted in reduced phosphatase activity at MT. Among the three eastern sites, only the mechanical + fire treatment in OH resulted in significantly reduced phosphatase activity (Fig. 1).

When the eight sites were pooled, there were no significant differences in chitinase activity among treatments or sites (Table 4). Chitinase activity was affected significantly by the treatments in five of the eight individual sites (Table 5). Chitinase activity was reduced by all three manipulative treatments in CaN and OH, by fire (with or without mechanical treatment) in OR, and by the two individual treatments but not the combined treatment in NC (Fig. 2).

When the phenol oxidase activity of the soils of the eight sites was analyzed as a group, there were significant differences both among treatments and among sites (Table 4). Phenol oxidase activity was significantly lower in the prescribed burn units than in the units given the other three treatments (Fig. 3). Among the individual sites, phenol oxidase activity was affected significantly only in CaN, OR, and OH (Table 4). Phenol oxidase activity was reduced significantly by fire alone in CaN, by fire plus mechanical treatment in OH, and by mechanical treatment alone in OR (Fig. 3).

Pre-treatment and post-treatment phosphatase activities were significantly and positively correlated in soils from the control, prescribed fire, and mechanical treatment units, and marginally correlated in the mechanical + fire treatment units (Table 6). In contrast, pre- and post-treatment chitinase activity was only significantly correlated in the control and mechanical treatments (Table 6). There were no significant correlations between pre- and post-treatment phenol oxidase activity (Table 6).

3.1.1. N mineralization and net nitrification

Overall there were no significant effects of the manipulative treatments on net N mineralization (Table 4, Fig. 4) or net nitrification (Table 4, Fig. 5). There were significant differences in net N mineralization in only three of eight sites (Table 5). In MT, the combined mechanical + fire treatment produced significantly greater N mineralization rates than the control and in OH both the mechanical treatment alone and the combined treatment had significantly greater N mineralization than did the control (Fig. 4). In NC, fire alone resulted in significantly decreased N mineralization relative to the control (Fig. 4).

The manipulative treatments resulted in significant effects on net nitrification in four of eight sites (Table 5). Net nitrification rates were enhanced by fire alone in CaS, and by fire combined with thinning in MT; in contrast, both fire treatments resulted in reduced net nitrification in OH (Fig. 5).

3.2. C:N ratios

Across the suite of eight sites, mechanical treatment resulted in a small, but statistically significant increase in soil C:N ratio (Table 4, Fig. 6). The overall relative difference between the mechanically treated units and the controls was approximately 7%. Five of the eight sites also exhibited significant effects of restoration treatments on soil C:N ratio (Table 5). In OR, SC, and NC the soil C:N ratio was significantly greater in the mechanical treatment than in the control. Fire alone resulted in increased soil C:N ratio in CaN and SC, but reduced soil C:N ratio in CaS. Again, these differences were small in absolute magnitude despite their strong statistical significance.

Over the eight sites, forest floor C:N ratio was reduced significantly by all three treatments, though once again the magnitude of the change was small (Table 4, Fig. 7). The relative difference between the controls and the three manipulative treatments averaged 8.5%. Relative to the controls, mechanical treatment

<table>
<thead>
<tr>
<th>Site</th>
<th>Phosphatase</th>
<th>Chitinase</th>
<th>Phenol oxidase</th>
<th>N mineralization</th>
<th>Net nitrification</th>
<th>Soil C:N</th>
<th>Forest floor C:N</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ca-N</td>
<td>F = 14.47, p &lt; 0.001</td>
<td>F = 9.33, p &lt; 0.001</td>
<td>F = 4.76, p &lt; 0.004</td>
<td>F = 2.24, p &lt; 0.088</td>
<td>F = 1.69, p &lt; 0.174</td>
<td>F = 10.49, p &lt; 0.001</td>
<td>F = 55.29, p &lt; 0.001</td>
</tr>
<tr>
<td>Ca-S</td>
<td>F = 23.72, p &lt; 0.001</td>
<td>F = 16.2, p &lt; 0.001</td>
<td>F = 0.46, p &lt; 0.500</td>
<td>F = 0.52, p &lt; 0.472</td>
<td>F = 6.70, p &lt; 0.011</td>
<td>F = 8.59, p &lt; 0.004</td>
<td>F = 8.12, p &lt; 0.007</td>
</tr>
<tr>
<td>OR</td>
<td>F = 4.73, p &lt; 0.005</td>
<td>F = 7.20, p &lt; 0.001</td>
<td>F = 3.56, p &lt; 0.018</td>
<td>F = 1.71, p &lt; 0.139</td>
<td>F = 5.18, p &lt; 0.003</td>
<td>F = 3.75, p &lt; 0.016</td>
<td></td>
</tr>
<tr>
<td>MT</td>
<td>F = 7.54, p &lt; 0.001</td>
<td>F = 19.0, p &lt; 0.013</td>
<td>no data</td>
<td>F = 4.69, p &lt; 0.005</td>
<td>F = 16.98, p &lt; 0.001</td>
<td>F = 0.44, p &lt; 0.728</td>
<td>F = 4.76, p &lt; 0.004</td>
</tr>
<tr>
<td>AZ</td>
<td>F = 1.89, p &lt; 0.023</td>
<td>F = 0.17, p &lt; 0.910</td>
<td>F = 1.35, p &lt; 0.344</td>
<td>F = 1.34, p &lt; 0.266</td>
<td>F = 1.71, p &lt; 0.169</td>
<td>F = 2.48, p &lt; 0.066</td>
<td>F = 14.77, p &lt; 0.001</td>
</tr>
<tr>
<td>OH</td>
<td>F = 7.21, p &lt; 0.002</td>
<td>F = 2.64, p &lt; 0.048</td>
<td>F = 2.81, p &lt; 0.044</td>
<td>F = 6.45, p &lt; 0.001</td>
<td>F = 5.89, p &lt; 0.001</td>
<td>F = 0.44, p &lt; 0.722</td>
<td>F = 3.26, p &lt; 0.025</td>
</tr>
<tr>
<td>SC</td>
<td>F = 0.83, p &lt; 0.470</td>
<td>F = 0.91, p &lt; 0.438</td>
<td>F = 0.93, p &lt; 0.431</td>
<td>F = 0.64, p &lt; 0.594</td>
<td>F = 2.19, p &lt; 0.281</td>
<td>F = 35.99, p &lt; 0.001</td>
<td>F = 61.31, p &lt; 0.001</td>
</tr>
<tr>
<td>NC</td>
<td>F = 115, p &lt; 0.032</td>
<td>F = 3.98, p &lt; 0.009</td>
<td>F = 1.30, p &lt; 0.276</td>
<td>F = 3.27, p &lt; 0.024</td>
<td>F = 3.51, p &lt; 0.018</td>
<td>F = 11.98, p &lt; 0.001</td>
<td>F = 5.89, p &lt; 0.001</td>
</tr>
</tbody>
</table>
resulted in increased forest floor C:N in only one site (SC). Fire resulted in reduced forest floor C:N in two sites (CaN and AZ) and increased C:N in one site (CaS) (Fig. 7). In six of the eight sites, the mechanical treatment resulted in significantly greater forest floor C:N than did the fire treatment (Fig. 7).

3.3. Ordination

NMS ordination of the differences between the post-treatment conditions in the control units and in the corresponding treated units in each site arrayed the site-treatment combinations along two axes which together accounted for >95% of the total variance in the matrix (Fig. 8). The large majority of the variance was represented by the first ordination axis, which was positively correlated with C:N ratios of soil and forest floor and negatively correlated with phosphatase activity, chitinase activity, and net nitrification (Fig. 8). The prescribed fire treatments were arrayed at the lower end of NMS Axis one, indicating that fire produced the greatest decreases in enzyme activity and net nitrification, relative to the corresponding controls. The mechanical and mechanical + fire units were arrayed at the center and upper portion of NMS axis one, indicating that these treatments produce greater increases in forest floor and soil C:N ratios than did the prescribed fire treatment. NMS axis two accounted for <10% of the variance and represented variance among sites within a treatment (Fig. 8).
4. Discussion

Soil microbes (*sensu lato*) release enzymes that metabolize the available soil organic matter. Some of these enzymes (e.g. phosphatase) are produced as long as the organism is metabolically active, whereas others (e.g. chitinases and phenol oxidase) are inducible and are only produced by certain microbial groups in response to the presence of specific substrates. We quantified activity rates of these three enzymes in order to gain insight into the microbial response to our restoration treatments.

When the eight study sites were considered as a group, phosphatase activity was not affected significantly by any of the three treatments; however, phosphatase activity was reduced significantly by treatments that included fire in four of the five western sites. Reductions in phosphatase activity by fire in dry coniferous forests have also been reported in *Pinus pinaster* and *Pinus sylvestris* stands in Spain (Saa et al., 1993). Among the eastern sites, phosphatase activity was reduced only by the combined treatment in the deciduous forests of the OH site. Although this result was consistent with prior studies in this particular site (Boerner and Brinkman, 2003; Boerner et al., 2004), there were no indications of significant reductions in phosphatase activity by any of the three treatments in the other two eastern sites.

**Table 6**

Correlations between pre-treatment and post-treatment enzyme activities in soils from eight Fire and Fire Surrogate Network sites. Correlation coefficients and associated probabilities are given.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Phosphatase</th>
<th>Chitinase</th>
<th>Phenol oxidase</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>0.796 (p &lt; 0.002)</td>
<td>0.709 (p &lt; 0.010)</td>
<td>0.230 (p &lt; 0.472)</td>
</tr>
<tr>
<td>Prescribed fire</td>
<td>0.726 (p &lt; 0.008)</td>
<td>0.700 (p &lt; 0.830)</td>
<td>0.188 (p &lt; 0.559)</td>
</tr>
<tr>
<td>Mechanical thinning</td>
<td>0.798 (p &lt; 0.002)</td>
<td>0.584 (p &lt; 0.047)</td>
<td>0.396 (p &lt; 0.200)</td>
</tr>
<tr>
<td>Mechanical + fire</td>
<td>0.560 (p &lt; 0.059)</td>
<td>0.467 (p &lt; 0.127)</td>
<td>0.170 (p &lt; 0.597)</td>
</tr>
</tbody>
</table>
As was the case for phosphatase, there were no significant overall effects of the three treatments on chitinase activity. In four of the eight sites chitinase activity was not significantly affected by any of the three manipulative treatments, whereas in the other four either the two treatments involving fire or all three treatments exhibited reduced chitinase activity. There was no geographic pattern to the effects on chitinase: there were two eastern and two western sites among the four in which treatments affected chitinase activity. Similarly, the sites in which chitinase activity was reduced included deciduous and coniferous forests, relatively dry and relatively humid sites, and sites with relatively low and high soil organic matter (McIver et al., 2008; Boerner et al., 2008).

Phenol oxidase activity varied greatly among sites. The soils of the OR and OH sites supported phenol oxidase activity 10- to 15-fold greater than did the soils of the AZ site. Overall, fire alone resulted in reduced phenol oxidase activity, though this difference was driven primarily by strong reductions after fire in CaN and after the combined mechanical + fire treatment in OH. We could find no previous studies that documented phenol oxidase activity in western coniferous forests following fire and/or harvesting, and prior studies of phenol oxidase activity in eastern forests have produced conflicting results (Boerner and Brinkman, 2003; Boerner et al., 2004, 2007).

Examining the correlations in the pre- and post-treatment activity of each enzyme produced several interesting contrasts. Phosphatase activity is controlled primarily by environmental factors, including temperature, moisture, and N availability. Pre- and post-treatment phosphatase activity were strongly and positively correlated in the control, prescribed fire, and mechanical treatment units, but only weakly (and not statistically significantly) correlated in the mechanical + fire plots. This suggests that the
phenol oxidase activity may often be larger than treatment effects, and this may be the cause of the conflicting effects of fire on phenol oxidase activity in previous studies (Boerner and Brinkman, 2003; Boerner et al., 2004, 2007).

Neither net N mineralization nor net nitrification was affected by any of the treatments over all eight sites, and there was no consistent pattern among the relatively few within-site differences. Furthermore, variability in N transformations among plots within treatments was considerably greater than were variations in enzyme activity. Similarly, Knoepp and Swank (1993) monitored N mineralization rates after clear-cutting of a mixed oak–pine stand in Tennessee and found that N mineralization was both greater and more variable in the harvested sites. In addition, the variation about the mean was considerably greater in the western sites than the eastern sites despite similar sampling intensity. This suggests that the interactions between spatial heterogeneity in fire severity and sampling design may have been more important in western coniferous forests than eastern forests, at least in this study.

Our results may, at first glance, seem to be in contradiction with the many previous studies that have demonstrated increases in N mineralization and net nitrification after single fires (Wagle and Kitchen, 1972; reviews by Raison, 1979; Wan et al., 2001; Boerner, 2006); however, when the results of the meta-analysis done by Wan et al. (2001) were stratified by fire intensity and ecosystem, they concluded that high intensity wildfires and slash fires in conifer forests resulted in increased N turnover whereas prescribed burning and other fires in hardwood forests did not. Whether this difference is driven by fire intensity or ecosystem type is unclear, as the many previous studies that have demonstrated increases in N mineralization following single fires (Wagle and Kitchen, 1972; reviews by Raison, 1979; Wan et al., 2001; Boerner, 2006) have most often been conducted in coniferous stands and may be the cause of the conflicting effects of fire on phenol oxidase activity in previous studies (Boerner and Brinkman, 2003; Boerner et al., 2004, 2007).

Neither net N mineralization nor net nitrification was affected by any of the treatments over all eight sites, and there was no consistent pattern among the relatively few within-site differences. Furthermore, variability in N transformations among plots within treatments was considerably greater than were variations in enzyme activity. Similarly, Knoepp and Swank (1993) monitored N mineralization rates after clear-cutting of a mixed oak–pine stand in Tennessee and found that N mineralization was both greater and more variable in the harvested sites. In addition, the variation about the mean was considerably greater in the western sites than the eastern sites despite similar sampling intensity. This suggests that the interactions between spatial heterogeneity in fire severity and sampling design may have been more important in western coniferous forests than eastern forests, at least in this study.

Our results may, at first glance, seem to be in contradiction with the many previous studies that have demonstrated increases in N mineralization and net nitrification after single fires (Wagle and Kitchen, 1972; reviews by Raison, 1979; Wan et al., 2001; Boerner, 2006); however, when the results of the meta-analysis done by Wan et al. (2001) were stratified by fire intensity and ecosystem, they concluded that high intensity wildfires and slash fires in conifer forests resulted in increased N turnover whereas prescribed burning and other fires in hardwood forests did not. Whether this difference is driven by fire intensity or ecosystem type is unclear, as most of the western coniferous forest fires in that data set were wildfires and most of the fires in the eastern deciduous forest were planned, low intensity prescribed fires. The lack of impact of a single prescribed fire on N turnover in eastern forests is further supported by studies that postdate the meta-analysis of Wan et al. (2001). For example, in an analysis of the initial effects of the FFS treatments in the OH site Boerner et al. (2000) found no significant effect of a single fire on N mineralization in an Ohio mixed-oak forest, and a similar lack of fire effect on N mineralization has been reported for oak–pine forests in Georgia and Tennessee (Hubbard et al., 2004) and oak–pine stands in North Carolina (Knoepp et al., 2004).

It is important to note that the results of single fires or relatively short term studies may not scale linearly across longer time periods. Vance and Henderson (1984) measured rates of N mineralization in a Missouri oak flatwoods that had been burned either annually or periodically (3–4 year rotation) for approximately 30 years, and found that N mineralization was reduced by long-term burning. They concluded that this change was a consequence of...
a change in organic matter quality (measured as C:N ratio), not quantity. Similarly, Eivasi and Bayan (1996) resampled the same Missouri oak flatwoods a decade later, and concluded that micro-
bial biomass and enzyme activity had been reduced in proportion
to fire frequency even though the amount of soil organic matter
present had not been significantly affected. This would be consist-
tent with what one would expect if long-term burning results in an
accumulation of recalcitrant C forms, including black carbon, over
time (Pononenko and Anderson, 2001).

Changes in soil N transformations following pre-commercial
thinning and other partial harvest practices may be the result of
additions of logging residues, which may include both relatively
low C:N ratio leaf material and relatively high C:N ratio woody
residues. In addition, opening of the canopy may produce changes
in forest floor and soil microlclimate, as well as reductions in plant
uptake resulting from the removal of tree basal area (Johnson and
Curtis, 2001). For example, clear-cutting in a Virginia hardwood
forest resulted in reduced plant uptake and increased micro-
bial biomass and N mineralization rate (Johnson et al., 1985).
Similarly, Frazer et al. (1990) reported N mineralization and net
nitrification rates in clearcuts in CA mixed conifer forests 4-fold and
2-fold greater, respectively, than in uncut stands.

A number of mechanisms may underlie the lack of apparent
effect of the FFS mechanical treatments on N turnover: (1) the
harvests that removed only 25–33% of the aboveground biomass
that characterized the FFS mechanical treatments were insufficient
to produce changes in soil micromass large enough to affect
microbial activity and N uptake, (2) the mix of logging residues was
sufficiently heterogeneous to result in no net change in organic
matter quality, and (3) spatial heterogeneity of canopy opening
and or logging slash may produce increases in variance without
changes in mean rates.

When all eight sites in this study were pooled, the C:N ratio of
both soil and forest floor was affected significantly by the treat-
ments, though the mean differences among treatments were < 10%
in both cases. Fire resulted in increased soil C:N in two sites (CaN
and SC) and decreased soil C:N in another (CaS), whereas five sites
exhibited no significant effect. Results of previous studies of the
effects of a single fire on soil C:N have been equally mixed. In
western coniferous forests Black and Harden (1995) documented a
small but significant increase in C:N ratio after fire while Antos
et al. (2003) found a significant decrease.

Forest floor C:N decreased after fire, with or without mechanical
management, in five of eight sites. This is a result that has been
commonly reported in the literature, and is primarily the result of
partial combustion removing higher C:N ratio residues from the
forest floor mix. For example, Vose and Swank (1993) reported a
reduction in forest floor C:N ratio of 10–20% following fire in an
eastern mixed forest. Similarly, Antos et al. (2003) observed a
decrease of ~30% in forest floor C:N ratio after prescribed fire in
a western coniferous forest.

Mechanical treatment resulted in increased soil C:N overall and
in three of the eight individual sites. Black and Harden (1995) noted
that soil C:N in sites clearcut 2 years prior had C:N ratio of 26–29,
compared with an average C:N of 22 in uncut sites. The primary
mechanism at work here would appear to be the mixing of forest
floor material into the mineral soil as the result of vehicle traffic or
other logging activities.

Woody residues remaining after logging typically have high C:N
ratios, and therefore we had hypothesized widespread effect of
partial harvesting on forest floor C:N ratio; thus, the observation
that the FFS mechanical treatments resulted in increased forest
floor C:N in only one site (SC) was unexpected. The mechanical
treatments were designed to be consistent with established
commercial harvesting practices at the various sites. Thus, the suite
of mechanical treatments applied across the network varied both in
the proportion of basal area removed, and perhaps more impor-
tantly, in the way the logging residues were treated. Some of the
sites removed logging residues from the site, some left the residues
in place, and others chopped the residues and mixed them with the
forest floor. The variations in the intensity of harvesting and the
residue treatment may well have obscured the somewhat higher
C:N ratio of the initial logging residues. Even in sites where residues
were left in place, the mix of foliar and woody residues may have
varied spatially, adding a strong element of spatial heterogeneity
and thereby preventing statistically significant mean effects from
occurring. For example, Olsson et al. (1996) compared coniferous
forest logging residues that were dominated by foliar materials to
those comprised of a mix of foliages and branches. The C:N ratio of
the former was 44–55 whereas the latter had C:N ratio of 74–136.

The ordination approach we used clearly separated the
prescribed fire treatment from the mechanical and mechan-
ic – fire treatment. The former had relatively greater (negative)
effects on three indicators of microbial activity (phosphatase
activity, chitinase activity, and net nitrification) whereas the latter
two treatments produced greater increases in soil and forest floor
C:N ratio. Thus, the prescribed fire treatment seemed to result in
a proximate reduction in microbial activity, whereas the two
mechanical treatments produced changes in the organic matter
complex that had potentially more persistent inhibitory effects on
microbial activity.

Only repeated sampling of these sites over time and through
several fire cycles will allow us to determine if these first-year
trends persist. As management strategies and decisions are
designed to maximize outcomes over meaningful periods of time,
so must experiments be designed to inform such decisions.

Acknowledgements

This is contribution number 63 of the National Fire and Fire
Surrogate Network Study funded by the Joint Fire Sciences Program.
We thank the following FFS site coordinators and coop-
erating scientists for field and lab assistance: Steve Hart and Carl
Edminster (AZ), Carl Skinner and Celeste Abbott (CaN), Sarah
Hamman and Eric Knapp (CaS), Tom DeLuca and Carl Fiedler (MT),
Tom Waldrop and Ross Phillips (NC, NC, Dan Yaussy (OH), and Andy
Youngblood and Jim McIver (OR). We also thank Jennifer Brinkman,
Aditi Shenoy, Phil Ruse, and Megan Cartwright for their assistance.

References

Spatial Changes in Soil Carbon and Nitrogen After Clearcutting and Burning of
an Old-growth Douglas-fir Forest. USDA Forest Service Research Paper PNW-
RP-552, Portland, OR, 19 pp.


Boerner, R.E.J., 2006. Earth, wind, water, and fire: how the elements conspire in the
forest context, USDA Forest Service General Technical Report NRS-P-1.


in soil nitrogen dynamics after prescribed burning in Ohio mixed-oak forests.
Landscape Ecol. 15, 425–439.

frequencies on nitrogen transformations and soil chemistry in a nitrogen-

restoration alternatives in eastern deciduous forests: the view from below-

effects of the FFS treatments on carbon storage and sequestration rate. For. Ecol.
Manage. 255, 3081–3097.

Boerner, R.E.J., Huang, J., Hart, S.C. Impacts of fire and fire surrogate treatments


