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Post-Fire Erosion Control Mulches Alter Belowground Processes and Nitrate Reductase Activity of a Perennial Forb, Heartleaf Arnica (*Arnica cordifolia*)

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Abstract

Four years post-wildfire, we measured soil and plant properties on hillslopes treated with two different mulches (agricultural wheat straw and wood strands) and a control (unmulched, but burned). Soil total N was about 40% higher and microbial respiration of a standard wood substrate was nearly twice as high in the mulched plots compared to the unmulched plots. Greater respiration was tied to increased substrate moisture underneath mulch compared to bare soil. Nitrate reductase activity of a common forb (Arnica cordifolia) was about 30% higher on the wood strand plots than either the wheat straw or the unmulched plots. Mulch applications after wildfire may enhance N availability by increasing soil moisture, promoting microbial N mineralization, or by increasing biological nitrogen fixation. Because inference is limited for this case study, we call for additional replicated experiments investigating effects of mulch treatments on soil carbon and nitrogen cycling with links to plant regeneration.

Keywords: *soil rehabilitation, restoration, fire effects, respiration, nitrogen*

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Introduction

Following severe wildfire, rehabilitation efforts often focus on reducing soil erosion, protecting downstream water quality, and maintaining soil productivity, particularly where there are ecosystem values at risk (Robichaud et al. 2013a). Erosion reduction and water quality objectives can be successfully addressed using erosion control techniques, such as aerially applied mulch (Robichaud et al. 2013b). These treatments also likely affect soil nutrient cycling and other belowground processes, such as decomposition, with implications for ecosystem recovery, but few studies have examined such non-target effects. Thus, forest managers who develop post-fire rehabilitation plans are less informed than they should be to fully consider ecosystem effects of applying mulch following a wildfire.

Mulching may have both positive and negative effects on soil nutrient cycling in the context of post-fire plant regeneration. Because temperate conifer forests are usually nitrogen (N)-limited (Hunt et al. 1988), regenerating vegetation may benefit from the ephemeral pulse of soil soluble N that often results from wildfire (Hart et al. 2005; Rodríguez et al. 2009; Stephan et al. 2012). Surface mulches may increase substrate for N mineralization by preventing erosional losses of organic matter or by serving as a substrate itself. In contrast, decomposition of added shredded wood or wheat straw mulch can immobilize soil N (Mary et al. 1996; Rhoades et al. 2012). These direct effects on N would likely interact with mulch effects on soil properties governing organic matter (OM) turnover rates, such as changes in soil moisture. It is difficult to predict the net effect of these mechanisms on soil N availability to plants.

To date, no studies have examined nutrient cycling consequences of post-fire mulching in dry, mixed conifer forests (Robichaud et al. 2009). Four years following a wildfire and mulch application in southeastern Washington, USA, we tested mulch effects on soil total N and OM, OM turnover rates, and NO_3^- assimilation by a common perennial forb, heartleaf arnica.

Methods

Study Area

Study plots were located within an area burned by the School Fire in July and August 2005 on the Umatilla National Forest in the Tucannon River drainage of the Blue Mountains, southeastern Washington, USA (latitude 46.22°, longitude -117.67°). Sites were located in high severity burned areas (>70% tree mortality, >50% bare mineral soil exposure; Parsons et al. 2010) on steep (51% to 70%) slopes. Pre-fire overstory vegetation was mixed-conifer, dominated by Douglas-fir (*Pseudotsuga menziesii* (Mirb) Franco) and grand fir (*Abies grandis* (Dougl) Lindl). Four years post-fire, dominant understory vegetation consisted of bluebunch wheatgrass (*Pseudoroegneria spicata* (Pursh) Å. Löve), pinegrass (*Calamagrostis rubescens* Buckley), and Geyer's sedge (*Carex geyeri* (Boott)) with many shrubs and forbs (Moy 2010). The dominant soil is an ashy silt loam (Limberjim series; ashy over loamy-skeletal, amorphic over isotic, frigid *Alfic Udivitrand*; NRCS 2013) derived from basalt.

Average annual precipitation from the time period between the wildfire and 1 year following our sampling date (2005-2010) was 1458 mm whereas average annual daily maximum and minimum temperatures were 10.9 and 2.3 °C, respectively (data from nearest weather station, 21 km from study site: Touchet SNOTEL 1686 station, latitude 46.11° longitude -117.85°, elevation 1681 m). This was slightly wetter than the average precipitation of 1434 mm and the same average annual daily maximum and minimum temperatures of 10.1 and 1.5 °C (1989-2010). Annual precipitation at the SNOTEL site was 1135, 1671, 1285, 1631, 1572 mm in the year of the fire (2005) and the subsequent 4 years (2006-2009), respectively.

In August 2005 soon after the fire, two individual but similar northwest-facing hillslopes were selected and each hillslope received an aerial application of one of two types of mulch: wheat straw (*Triticum aestivum* L.) and wood strands which were Forest Concepts' WoodStraw® (Federal Way, WA). As described by Robichaud et al. (2013b), wheat straw (C:N ~140) was applied at a rate of 2.2 Mg ha^{-1} on a steep 65% slope hillslope (NW aspect), resulting in an average of 57% ground cover. Wood strands (C:N ~530) were applied on an adjacent hillslope (62% slope, NW aspect) at a rate of 4.5 Mg ha^{-1}, resulting in an average of 54% ground cover. High burn severity was assessed using the Burned Area Reflectance Classification maps and was confirmed on-site. One similarly burned hillslope (65% slope, W aspect) was selected and retained as the study's control. All hillslopes were at similar elevations (1500 m +/- 50 m) and had similar pre-fire vegetation. We report measurements conducted 4 years following treatment, in July and August 2009.

To monitor mulch effects on soil N available for plant uptake, three plots, separated by ~ 20 m to adequately capture spatial variation, were selected on each hillslope for collection of plant material for plant N-use assays (see *Plant N Use* below). Locations of plots represented three different vertical sections of the hillslopes, to correspond with measurements of plant response and sediment transport from the hillslopes (Robichaud et al. 2013b; Moy 2010). To monitor mulch impacts on organic matter turnover, microbial respiration was measured on common substrates buried in three subplots per hillslope (see *Microbial Respiration* below).

Soil Properties

On 1 August 2009, we collected five each of surface mulch and mineral soil samples (0 to 10 cm depth) from each plot and composited them into three bulk samples for each hillslope. Samples were dried at 60 °C until their weight stabilized (overnight), ground, and analyzed for soil pH on a 2:1 water:soil paste. Soil and mulch percent C and N were analyzed with a LECO® Induction Furnace. Soil percent organic matter was determined after loss-on-ignition in a 475 °C oven for 6 hr, as recommended by Ball (1964).

Forb N Use

Availability of soil N for plant use was estimated by measuring nitrate (NO_3^-) reductase activity (NRA), which measures assimilation of nitrate (Kelker and Filner 1971; Högberg et al. 1986). On 18 August 2009, two separate heartleaf arnica (*Arnica cordifolia* (Hook.)) plants were sampled in each of three plots per hillslope. This particular species was selected because 1) it is a perennial and its nutrient status likely reflects the cumulative nutrient availability of the past several years since the fire, 2) it was abundant across most plots following the fire (Moy 2010), and 3) it is not an N-fixer and thus would reflect only soil N status. Squares (1 cm^2) were cut from each of two leaves from each plant; both squares were placed together immediately in a vial containing a pH 7.5 buffered solution of potassium nitrate, potassium phosphate and propanol, and reacted for 1 hour in the field in darkness at 22 °C (in a portable cooler). After incubation, we stopped the nitrate reduction reaction and developed color of the resulting nitrite by pipetting 0.5 mL of the incubation medium into a 2 mL centrifuge vial, to which 0.5 mL of 1% sulfanilamide in 3 N hydrochloric acid HCl (Su - HCl) and 0.5 mL of 0.02% N-(1-naphtyl) ethylene diamine dihydrochloride (NED) in distilled, deionized water (DDI) were added. We removed leaf samples from the buffer vials and dried them at 70 °C for 24 hours to determine their dry weight. The reacted solutions were kept at 4 °C until absorbance was measured at 540 nm using a Beckman Coulter Spectrophotometer (Fullerton, CA). We took these measurements within 2 days of collecting samples. The blank consisted of the buffer and Su - HCl and DDI water. We computed nitrite concentration from absorbance using standard curves; NRA was expressed in terms of nmol nitrite production per hour per gram (dry weight) of the leaf samples.

Microbial Respiration

In each subplot, twenty-five loblolly pine (*Pinus taeda*) wood stakes (free from defect) (2.2 cm x 2.2 cm x 20 cm) were buried vertically at the mineral soil and mulch interface following the fire just after mulch application (for details on installation method, see Jurgensen et al. 2006). We retrieved buried loblolly pine stakes (n = 5 per subplot) on 17 June 2009 for measurement of microbial respiration of a common substrate, which we used as a short-term index of soil organic matter turnover. Immediately upon stake retrieval, we weighed the stakes in the field, then placed each stake into a modified Li-Cor respiration chamber (LI-6000-09, Lincoln, NE) attached to a portable respiration system (LI-6400). We measured respiration rate at ambient CO_2 for two cycles (each lasting from 0.5 to 1 minutes per stake), recording the average of the two cycles. Stakes were dried at 70 °C for about 48 hr until weight stabilized, then weighed to obtain field moisture content as percent of dry weight. Stake respiration is expressed as mmol C dry g^{-1} s^{-1}.

Data Analysis

We determined hillslope differences in soil percent nitrogen (N), substrate moisture, and plant NRA with analysis of variance (ANOVA) using functions *aov* and *lm* in R (R Core Development Team, 2011). Differences in stake respiration by hillslope (and the covariate of stake moisture) were determined with analysis of covariance (ANCOVA) using functions *Anova* (package *car*) and *lm* in R. Data from each plot (3 per hillslope) were treated as independent measurements, acknowledging that true replication and independence was sacrificed in favor of the need to work with large-scale mulching treatments (Oksanen 2001). Therefore, these results represent the outcome of a case study and caution should be used when extrapolating outside of the treated hillslopes studied here. We tested data for assumptions of homoscedasticity and model residual normality using Levene's test for one-way ANOVA and the Shapiro-Wilks test, respectively. If assumptions were not met, then appropriate power transformations were applied until assumptions were met. Means were separated using Tukey's Honest Significant Difference. Data are reported with mean +/- 1 standard error, and statistical significance is reported at $\alpha = 0.05$.

Results and Discussion

Four years following wildfire and aerial mulch applications to the School Fire, total surface (0 to 10 cm) soil N was up to 40% higher underneath wood strands and wheat straw compared to unmulched areas ($P < 0.01$; Table 1). These results may not be fully explained by increased retention of organic matter and sediment by mulch treatments that otherwise erode during post-fire rain events. This is because soil loss from the control hillslopes was low, with mulch treatment having little impact on soil loss compared to the control (Robichaud et al. 2013b). There are two possible causes for increased soil N underneath mulch. One possibility is that mulch altered rates of biological nitrogen fixation (BNF) by free-living (asymbiotic) bacteria from the atmosphere into the soil, an important mechanism controlling N availability to plants in natural, N-limited forests (Granhall and Lindberg 1980; Jurgensen et al. 1987). In Inland Northwest forests, mineral soil BNF by free-living bacteria may account for 0.01 to 0.05 g N m^{-2}, and BNF on woody residue accounts for about 0.014 g N m^{-2} whereas additional woody residue, such as that following harvest, may increase that amount by two-thirds (Jurgensen et al. 1992). Thus, mulch could provide additional surface area to support higher rates of BNF, increasing soil N concentrations. In addition, BNF responds strongly to changes in soil moisture (Belnap 2002); higher soil moisture underneath mulched areas may have increased BNF. Second, some of the organic N in the original mulch material may have become incorporated into the soil over the 4 years between application and sampling. Visual observations suggested that smaller pieces of decomposed straw mulch was contained in the surface soil layers that were sampled for C and N, despite efforts to remove intact mulch pieces from the soil surface. In areas where wildfire completely consumed the organic litter and duff layers,

Table 1—Average soil characteristics under the two mulch treatments and the unmulched control 4 years following wildfire and mulch application on the School Fire, eastern Washington.

Site treatment	pH	Organic Matter	Carbon	Nitrogen
		--------------------------------*Percent*---------------------------		
Wheat straw	6.6 (0.18)[a]	7.5 (1.8)	3.07 (0.91)	0.184 (0.036)a
Wood strands	6.4 (0.03)	8.3 (0.4)	3.40 (0.27)	0.224 (0.020)a
Burned control	6.8 (0.13)	5.7 (0.1)	2.39 (0.12)	0.158 (0.011)b

[a] Values in parentheses are standard error of the mean. Different letters within a single column indicate significantly different treatment effects ($P < 0.01$; n = 3 composite samples).

the amount of N added by mulch may have exceeded that lost during the fire. Original aerial mulch application rates for the School Fire are estimated at 2.2 Mg ha^{-1} for wheat straw and 4.5 Mg ha^{-1} for wood straw, which would have potentially added 1.89 and 0.75 g N m^{-2} to the soil surface. Comparatively, forest floor N loss from burned forests in the Inland Northwest may range from 7.7 to 20.9 g N m^{-2} (Page-Dumroese and Jurgensen 2006), suggesting that organic N delivered via mulch may offset 10 to 25% of wildfire losses from the forest floor. However, this effect depends strongly on mulch decomposition and incorporation rates of N into the mineral soil. Future work needs to characterize decomposition rates of mulch in order to understand the potential contribution of mulch-derived N to mineral soil N. Replacing fire-induced losses of organic N via mulch may increase substrate for N mineralization, aiding forest productivity, influencing plant regeneration and ecosystem recovery (Pastor et al. 1984; Chapin and Matson 2011). The fate of mulch-delivered N in post-fire ecosystems needs to be fully assessed, together with the erosional mitigation impact of mulching on post-fire N retention and potential effects of mulch on BNF inputs.

Mulch cover reduces soil water evaporation and affects soil aggregate stability and porosity, thereby changing nutrient and water relations within the soil profile (Mulumba and Lal 2008). By increasing substrate moisture at a time when it would otherwise be low (typical of late summers in the Inland Northwest), surface mulch applications create an environment that promotes soil microbial activity, thereby altering long-term soil sustainability through the breakdown of OM, nutrient flux control, soil C sequestration, decomposition, mineralization, and immobilization (Nannipieri et al. 2003). Microbial turnover of soil C, measured as respiration from buried loblolly pine stakes, strongly co-varied with stake moisture content (Figure 1, Table 2). All stakes with moisture contents in the top 50th percentile were harvested from mulched areas rather than from the control. Thus, lower moisture in the control plots probably restricted microbial respiration there. Our results suggest that by altering the soil environment surrounding microorganisms, surface mulch applications may alter soil nutrient transformations with potential feedbacks to vegetation. Future work should resolve mulch effects on soil temperature as well as moisture effects in areas with different climate regimes.

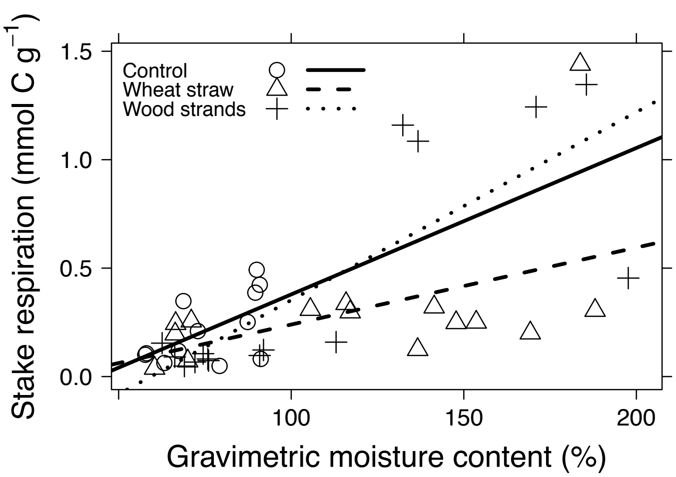

Figure 1—Stake respiration (mmol C g^{-1}) as related to stake moisture content (% w/w) at the time of stake retrieval 17 June 2009, with least squares linear regression lines for each mulched hillslope.

Table 2—ANCOVA results showing effect of stake moisture on stake respiration and no effect of treatment on stake respiration neither before (Type II SS) nor after (Type III SS) accounting for treatment effect on stake moisture content (g H_2O g^{-1} dry stake). Analyses performed on power-transformed stake respiration rates (power = 0.0158).

Source	DF	Type II SS	MS	F	P value
Treatment	2	0.000247		1.12	0.34
Stake moisture content	1	0.00411	0.00411	37.1	<0.0001
Stake moisture content*Treatment	2	0.000589		2.66	0.084
Source	**DF**	**Type III SS**	**MS**	**F**	**P value**
Treatment	2	0.00041		1.85	0.17
Stake moisture content	1	0.00157	0.00157	14.1	<0.001
Stake moisture content*Treatment	2	0.000589		2.66	0.084

The effect of stake moisture on respiration may have varied by hillslope (P = 0.08), suggesting that differences in mulch type, such as mulch chemistry, may have been a secondary control over microbial respiration. At similar moisture levels, stake respiration underneath wood strands was typically higher than underneath wheat straw. Different substrate C:N derived from different mulches may result in divergent decomposer communities at the two hillslopes, which may control decomposition rates (Lauber et al. 2008); however, this effect is difficult to separate from existing edaphic or climate differences between the two hillslopes. The possibility of mulch type to act as a secondary control over microbial nutrient cycling should be further explored in the post-fire environment.

Nitrate reductase activity of heartleaf arnica leaves was significantly higher in the plots mulched with wood strands than in those either mulched with the wheat straw or control (P < 0.01, Figure 2). Plant leaf NRA is a good indicator of soil nitrate availability in systems where NO_3^- is the dominant form of N available to plants (Högberg et al. 1986; Koba et al. 2003), as is often the case in post-fire ecosystems (DeLuca and Sala 2006; Koyama et al. 2010; Smithwick et al. 2005). Our data suggest that wood strand mulch results in elevated soil NO_3^- compared to the control after 4 years. This is supported by findings of elevated inorganic soil N in unburned forests 3 years after wood chip application (Miller and Seastedt 2009) and may have resulted from increased microbial mineralization or by increased BNF triggered by the higher soil moisture underneath mulch (Cassman and Munns 1980). However, it remains unclear why the wheat straw mulch was not associated with higher NRA despite the increased microbial respiration and similar soil moisture as the wood strands. More work is needed to understand whether effects of mulch on soil N processing are mediated more by abiotic effects (i.e., elevated soil moisture or temperature) or by substrate quality of mulch (i.e., C:N, lignin content).

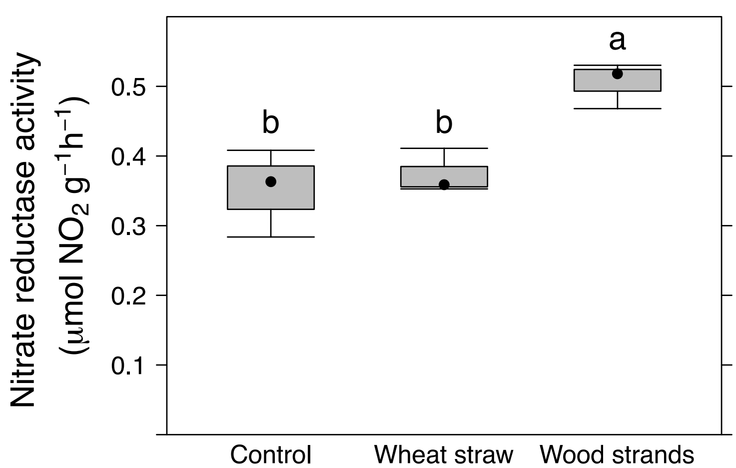

Figure 2—Box-and-whisker plots of heartleaf arnica leaf nitrate reductase activity collected on 18 August 2009. Different letters indicate significantly different treatment effects (P < 0.05). Dots represent sample median, boxes represent the 25th and 75th percentiles, and umbrellas represent the full data range.

Table 3—Mulch C:N in original mulch material and in material collected from soil surface 4 years following wildfire and mulch application. Values in parentheses represent standard error (n = 3).

Mulch type	C:N
Wheat straw – original	142[a]
Wheat straw – 4 yr old	50.8 (3.87)
Wood strands - original	532[a]
Wood strands – 4 yr old	214 (27.6)

[a] Original values were obtained from a single large (~5 L) sample of original mulch material.

Both mulch types increased in N during the 4 years they were present on the soil surface (Table 3). Decreased mulch C:N is consistent with net N immobilization measured during decomposition of residues in unburned forests and agricultural soil (Jensen, 1996; Li et al., 2013; Rhoades et al. 2012). However, there was no evidence that N immobilization into mulch reduced nitrate availability to plants, estimated by arnica NRA. This result may have been due to the low mulch application rate (compared to forest fuel mitigation and agricultural applications) and the long delay (4 years) in measurement following mulch application. Microbial immobilization of N in organic amendments is typically highest at high initial C:N and high C:N substrates such as sugar and sawdust are often used to immobilize soil N as part of ecosystem restoration (Paustian et al. 1992; Perry et al. 2010). Our wheat straw application rate of 2.2 Mg ha^{-1} is on the lower range of that remaining after agricultural practices (3 to 10 Mg ha^{-1}) shown to immobilize soil N (Mary et al. 1996); however, it should be pointed out that, unlike surface aerial mulching, previous research on wheat straw amendments involved physical incorporation into the soil, which would have maximized microbial access to C. Thus, the low application rates of the wheat straw or the lack of wheat straw incorporation could have minimized N immobilization. It is important to better resolve the question of N immobilization following application of post-fire mulch, especially the effects of amount and timing of mulch application. Mulch applications may fuel the post-fire microbial N sink that Turner et al. (2007) described, with mulch conserving soluble N that would otherwise be lost due to post-fire leaching. Further research is needed to quantify short-term N immobilization across post-fire mulches of different C:N and application rates.

Wildfire can result in large losses of N from forest ecosystems (Carreira et al. 1994; Turner et al. 2007; Stephan et al. 2012), increasing N limitation of primary productivity. In the long term, mulching may sequester N through erosion reduction or immobilization, or increase N inputs via biological nitrogen fixation, and lead to higher post-fire N pools, ultimately increasing ecosystem N compared to similarly burned but unmulched areas. Higher post-fire N has important implications for vegetative recovery and, consequently, for managers meeting ecological restoration or C sequestration objectives. Besides serving as a valuable technique for reducing post-fire erosion and runoff, our data suggest that post-fire mulching increases total soil N, soil microbial activity and potentially soil N availability to regenerating vegetation. However, these effects are based on a one-time sampling 4 years post fire. Thus, our results only represent net effects on soil N processing that may be attributable to a combination of multiple processes, such as increased soil moisture combined with increased sediment retention, with an unknown role for N immobilization. There remains a further need for monitoring of soil processes including immediately before and after mulch application and continuing throughout at least 5 years, at a temporal resolution high enough to capture the full trajectory of mulch decomposition on the soil surface. Ideally, this should be done using a controlled, replicated experiment that includes a non-decomposing mulch to help differentiate the effects of mulch on the physical environment (e.g. in increasing soil moisture) from effects as a substrate for soil microorganisms. In addition, these effects are likely to change depending on climate, soil and forest type. For example, in areas with coarser-textured soil than those studied here, moisture differences with mulch application may not be as pronounced due to the lower water-holding capacity of the soil. Thus, future experiments at multiple sites capturing a range of fire-affected ecosystems will help us fully assess whether post-fire mulching provides additional ecological benefits or drawbacks both in the short- and long-term.

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