Microsite and Time Since Prescribed Fire's Influence on Soil Microbiology in a Pinyon Woodland

Benjamin M. Rau, Robert R. Blank, and Tye Morgan

Abstract—Pinyon-juniper (*Pinus monophylla* Torr. & Frém.—*Juniperus osteosperma* Torr.) encroachment into sagebrush grasslands is a continuing problem in the Western United States. Prescribed burning has been suggested to slow woodland encroachment. We examined surface soil microbial community structure using Phospholipid Fatty Acid (PLFA) analyses to determine differences between burned and unburned woodlands at two microsites. Tree canopy interspace microsites had a greater total PLFA and percentage Eukaryotes. Conversely, under tree canopy microsites had a greater percentage of fermicutes, anaerobic metal reducers, and higher PLFA Cis/Trans fatty acid ratio. Time since burning increased Eukaryote PLFA in both microsites.

Introduction

Pinyon-juniper (*Pinus monophylla* Torr. & Frém.—*Juniperus osteosperma* Torr.) woodland encroachment into sagebrush-grasslands is a large scale ongoing problem in the Great Basin and other arid regions in the Western United States (Miller and Tausch 2001). Woodland encroachment can lead to exotic invasion by competition with perennial understory vegetation for resources, elimination of perennial understory seedbanks over time, and increasing the risk of catastrophic wildfire due to woody fuel accumulation (Tausch 1999a,b). Also litter produced by pinyon pine and juniper contains high concentrations of monoterpenes and could be allelopathic to native perennial vegetation (Everett 1987; Wilt and others 1993). We hypothesize that pinyon litter and burning influences the microbial community possibly through the changes in carbon substrate, pH, monoterpene concentration, or other chemical parameters.

Methods

We collected soils from a Great Basin pinyon woodland in Nevada, U.S.A. (39°15'11" N, 117°35'83" W). Soils in this study are classified as coarse loamy mixed frigid Typic Haploxerolls derived from silica ash flow tuffs (Rau 2005). Surface (0 to 15 cm) soil was collected from under pinyon tree canopies after removing the O horizon (litter) and from between tree canopies where healthy perennial understory vegetation occurred. Soils were collected from three adjacent tree plots in May 2005. The first plot had not been burned in greater than 80 years, the second was prescription burned in May of 2002, and the third was prescription burned in May of 2004. Three sub-samples were collected from three individual tree canopies and canopy interspaces at all three burn treatments using a 10 cm diameter sample auger. Sub-samples were homogenized in a bucket in the field then placed on ice and returned to the lab (N = 3 burn treatments x 2 microsites x 3 replicates = 18). Samples were packed in ice and sent to Microbial insights® (Rockford, TN, U.S.A.) for PLFA analyses. Microbial Insights calculates microbial biomass as cells g^{-1} based on a conversion factor (20,000 cells mol^{-1} PLFA). Results of the PLFA analyses were reported as total cells g^{-1} soil, PLFA Cis/Trans fatty acid ratio, percent fermicutes, percent anaerobic and metal reducing bacteria, percent proteobacteria, percent actinomycetes, percent eukaryotes, and the fungus to bacteria ratio. A two way ANOVA was used to compare the results from the PLFA analyses with burn treatment and microsite considered as main effects, and group means were compared using Duncan's test (SAS Institute 2004).

Results and Discussion

Results from the ANOVA are presented in table 1. Canopy interspaces had a greater total concentration of PLFA than soil from beneath tree canopies, and the PLFA beneath tree canopies had a higher Cis/Trans fatty acid ratio (fig. 1). The Cis/Trans fatty acid ratio is a measure of cell wall permeability and cell growth rate. A higher Cis/Trans fatty acid ratio indicates that the microbial community was possibly under more environmental stress and its cell membrane is less permeable and growth rate has slowed (Guckert and others 1985). Microbial stress could be related to carbon substrate quality, low pH, and monoterpene content of pinyon litter and the soil beneath it (Guckert and others 1985; Pietikainen and others 2000; Rau 2005; Wilt and others 1993) Fermicutes and anaerobic reducers were a higher proportion of the microbial community under tree canopies (fig. 2). Soil beneath pinyon canopies is typically much higher in organic carbon than canopy interspace soil which may facilitate reducing conditions (McDaniel and Graham 1992). Anaerobic reducers were generally a higher portion of the community in areas burned in 2002 than in unburned areas (fig. 2). This seems counter intuitive since some organic carbon was inevitably

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Table 1—Result for the ANOVA for burn treatment and microsite differences.

	DF	# Cells/g Soil		Cis/Trans FA		Actinomycetes		Proteobacteria	
		F	P	F	P	F	P	F	P
Treatment (T)	2, 12	0.3	0.7488	0.14	0.8712	3.29	0.0725	3.49	0.0638
Vegetation type (V)	1, 12	28.97	0.0002	18.74	0.001	1.85	0.1986	0.1	0.7609
T x V	2, 12	0.95	0.4159	0.83	0.4614	0.85	0.4507	0.6	0.5619

	DF	Fermicutes		Reducing bacteria		Eukaryotes		Fungi/Bacteria	
		F	P	F	P	F	P	F	P
Treatment (T)	2, 12	3.87	0.0504	7.17	0.0089	52.39	<0.0001	51	<0.0001
Vegetation type (V)	1, 12	5.66	0.0348	26.91	0.0002	42.27	<0.0001	36.7	<0.0001
T x V	2, 12	0.06	0.9467	9.86	0.0029	5.53	0.0198	5.26	0.0229

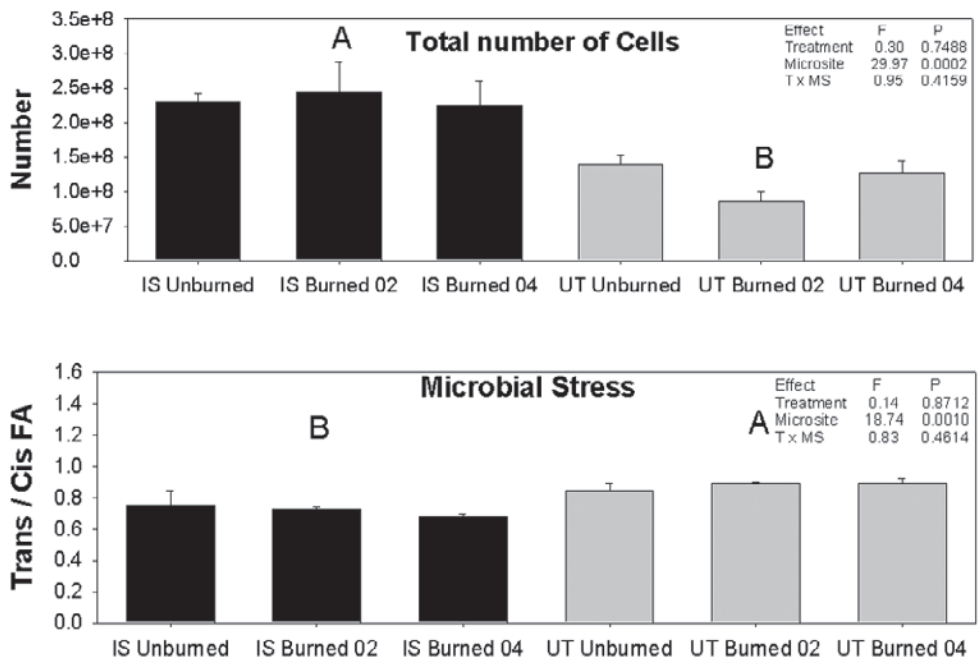

Figure 1—Mean number of microbes per gram of soil, Cis/Trans fatty acid ratio of PLFA, and standard errors for burn treatment and microsite. Upper case letters indicate microsite differences. IS = canopy interspace, UT = Under tree canopy.

consumed by combustion. Likewise the results for treatment microsite interactions do not provide a logical pattern of anaerobic reducer distribution (fig. 2). Eukaryotes were a significantly greater portion of the microbial community in the canopy interspace microsites than under tree canopies (fig. 3). Eukaryotes increased their proportion steadily with time following fire in canopy interspaces, but immediate recovery was delayed under tree canopies (fig. 3). Eukaryote PLFA is indicative of fungi and may represent not only decomposers but also mycorrhizae, which are essential for many perennials in the Great Basin (Grayston and others 2001; Wicklow 1994). Identical results were observed for the fungi to bacteria ratio because no differences in bacterial PLFA were observed during this study (fig. 3).

The results of our limited study contrast and confirm results observed by other researchers. In a study from pinyon-juniper woodlands in Arizona the authors suggest that bacterial populations are more diverse under tree canopies than inter-canopy, although relative biomass and fungi estimates were not compared (Dunbar and others 1999). Results from a lab study where soil and litter from pinyon-juniper woodlands was burned in microcosms showed soils

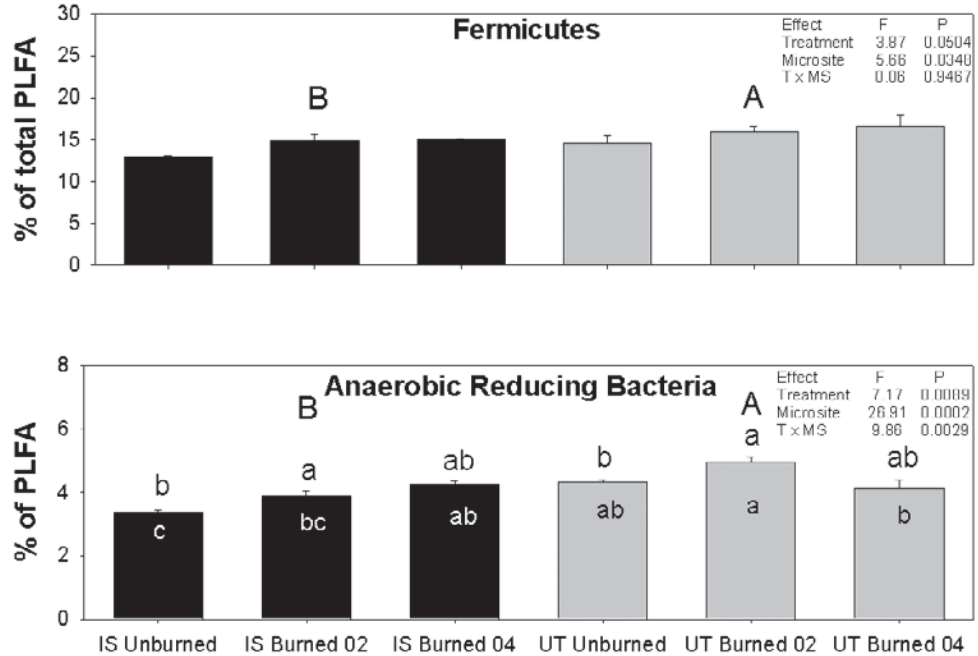

Figure 2—Mean percent of fermicute PLFA, anaerobic reducing bacteria PLFA, and standard errors for burn treatment and microsite. Upper case letters indicate microsite differences, lower case letters above each bar indicate burn treatment differences within microsite, and lowercase letters within bars indicate differences between all burn treatments and microsites. IS = canopy interspace, UT = Under tree canopy.

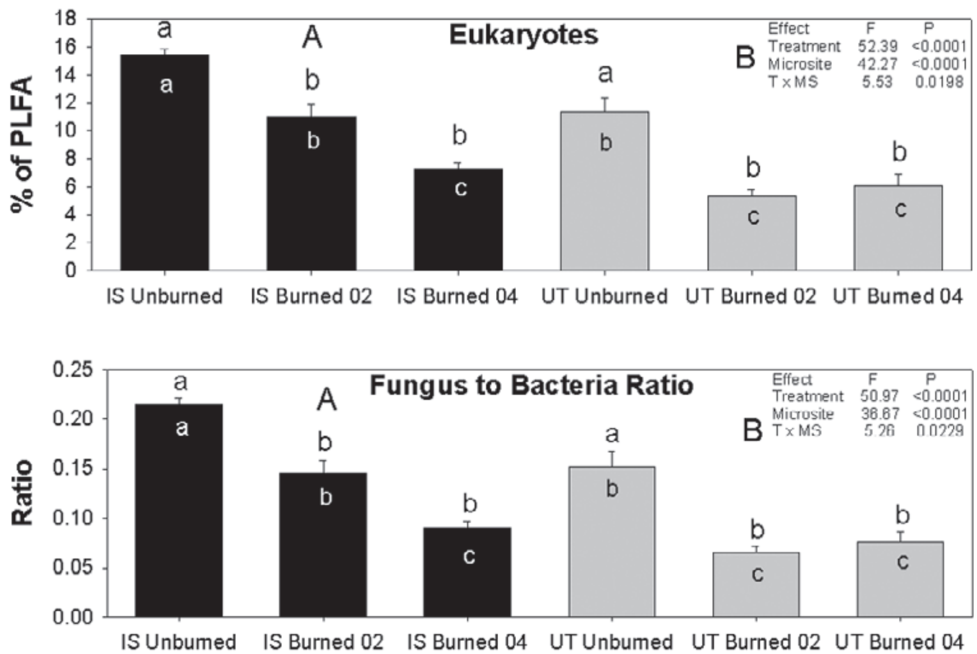

Figure 3—Mean percent of Eukaryote PLFA, fungus to bacteria ratio, and standard errors for burn treatment and microsite. Upper case letters indicate microsite differences, lower case letters above each bar indicate burn treatment differences within microsite, and lowercase letters within bars indicate differences between all burn treatments and microsites. IS = canopy interspace, UT = Under tree canopy.

beneath burned pinyon litter supported less mycorrhizal colonization than unburned controls (Klopatek and others 1988). A prescribed burn and wood ash treatment study in Scots pine (*Pinus sylvestris* L.) indicate that microbial biomass particularly fungi are reduced by burning and raising pH (Baath and others 1995). Prieto and others (1998) also observed that although microbial biomass decreases immediately after fire, it rebounds, but may take up to 4 years to reach pre-burn levels. Pietikainen and others (2000) found that heating forest floor material to 230 °C changed the carbon substrate and raised the pH in Scots pine litter and subsequently altered the microbial community.

The changes observed in the microbial community caused by pinyon encroachment and burning may have significant effects on perennial understory vegetation recovery potential. Most native perennial vegetation in the Great Basin is dependent on mycorrhizal associations, and although the relationships may not be species specific, each plant species responds differently to various mycorrhizae species (Bever 2002; Wicklow 1994). It has also been observed that plants have positive feedback loops with certain mycorrhizae. A plant alters its soil environment after establishment through litter chemistry or root exudates so that particular mycorrhizae are favored; the preferred micorrhizae then optimally assists the plant with nutrient uptake (Bever 2002). It is known that pinyon pines are one of the only species in arid woodlands to host ectomycorrhizal species (Haskins and Gehring 2004). Most other perennial shrubs, herbaceous, and tree species, including *Juniperus* spp., are arbuscular mycorrhizal hosts (Haskins and Gehring 2004). If pinyons have altered the microbial communities under their canopies (especially mycorrhizae) and fire has reduced microbial numbers, then it may be difficult for native perennial understory vegetation to recover following fire on sites previously occupied by pinyon trees. Klironomos (2002) showed that native vegetation grows significantly better on soil that was previously occupied by the same species than on soil previously occupied by a different species.

Conclusions

From our study we cannot say with certainty which species of fungus or mycorrhizae inhabit the under tree canopy and tree canopy interspace microsites. However, we know that these microsites differ in there microbiological properties. Changes in soil biota caused by woodland encroachment along with monoterpene production by pinyon, and the harsh physical environment produced after burning may inhibit native perennial understory vegetation recovery in pinyon canopy microsites following burning and lead to exotic invasion by cheatgrass (*Bromus tectorum*). Studies designed to identify the magnitude of impact each of these factors has on perennial vegetation establishment are needed to improve the likelihood of successful prescribed burning in pinyon woodlands.

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