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Emma Jones  
*University of Louisville*

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**The effects of prescribed fire on ant community composition in a temperate deciduous forest**

Emma Jones

Department of Biology  
University of Louisville

Submitted in partial fulfillment of the requirements  
for Graduation *summa cum laude*  
and  
for Graduation with Honors from the Department of Biology

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**Keywords:** disturbance, forest management, Formicidae, Kentucky, pitfall trap

24 **Abstract**

25 Prescribed fire is a tool commonly used in land management to decrease wildfire  
26 frequency and promote plant diversity. However, the effects of prescribed fire on  
27 invertebrate communities, especially those within temperate deciduous forest, are  
28 poorly understood. I measured the response of epigeic ant communities in mixed  
29 mesophytic forest in Berea, Kentucky following prescribed burning. I used pitfall traps  
30 to repeatedly sample epigeic ants in replicate burned and unburned plots for up to 21  
31 months postburn following two separate (2021 and 2022) prescribed fires. Ant species  
32 richness was similar between treatments (burn vs. control) and by burn year. Ant  
33 community composition generally differed between treatments and across years but was  
34 similar between the paired 2022 burned and unburned plots, probably due to the low  
35 intensity of that burn. The results of this study indicate that epigeic ant communities in  
36 an eastern deciduous forest are altered by prescribed burning, and do not return to  
37 normal activity levels after 1-year post-burn. Additional experimental studies are needed  
38 to determine the effects of fire intensity and frequency on ant assemblages in this  
39 setting.

40

## 41 **Introduction**

42 Ecological disturbance is any relatively discrete event in time that disrupts  
43 ecosystem, community, or population structure and alters resources or habitat (Pickett  
44 and White 1985). Fire is a natural disturbance in many terrestrial ecosystems and can  
45 vary widely in intensity and frequency (Swetnam and Betancourt 1990, Delcourt and  
46 Delcourt 1997). Fire in forested ecosystems often promotes plant biodiversity (Richter et  
47 al. 1982, Mitchell et al. 2006), which generates a bottom-up effect for organisms at  
48 higher trophic levels (Scherber et al. 2010). Historical suppression of wildfires in  
49 temperate forest of North America altered natural forest turnover (Ryan et al. 2013),  
50 leading to a decrease in biodiversity and habitat heterogeneity (Baker 1992, 1993).  
51 Consequently, intentional burning (i.e., “prescribed burning”) of forests has become  
52 increasingly common in forestry management operations to promote biodiversity,  
53 create habitat heterogeneity, and mitigate extreme wildfire events. (Agee and Skinner  
54 2005, Finney et al. 2005, Prichard et al. 2010, Cochrane et al. 2012).

55 Fire regimes can vary in frequency (i.e., number of fires per unit time) and burn  
56 intensity (i.e., heat transfer per unit duration of fire, Hiers et al. 2009, Perry et al.  
57 2011.). Most studies evaluating how different fire regimes influence forest components  
58 focus on plant communities (Pausas 1999, Franklin et al. 2001, Bond et al. 2004, Keeley  
59 2006, Miller et al. 2019), but data on ecologically important groups, like epigeic (i.e.,  
60 ground dwelling) arthropods, are lacking.

61 Ants are ecologically important components of forested ecosystems. They  
62 function as ecosystem engineers (Folgarait 1998, Jouquet et al. 2006), keystone seed  
63 dispersers (Lengyel et al. 2010, Gorb and Gorb 2013, Warren and Giladi 2014), and  
64 predators of many primary consumers (Folgarait 1998, Philpott and Armbrecht 2006).  
65 Fire can negatively impact ants directly through mortality and indirectly by reducing  
66 available nesting sites (Graham et al. 2009). Given that ants are numerically abundant,  
67 good bioindicators of disturbance (Anderson et al. 2002), and easily sampled, they may  
68 provide insight on how similar epigeic arthropods respond to prescribed fire in  
69 temperate deciduous forests.

70 Previous studies have examined the effect of prescribed fire on ant species  
71 richness and community composition in a variety of ecosystems, including prairies  
72 (Underwood and Christian 2009, Menke et al. 2015, Bonoan and McCarthy 2022),  
73 eucalypt forests (Andersen et al. 2009, Beaumont et al. 2012), and savannas (Izhaki et  
74 al. 2003, Houdeshell et al. 2011). Temperate deciduous forest cover 7.8 million km<sup>2</sup>  
75 worldwide and dominate most of the eastern United States (Allaby 2006). The few  
76 studies that were conducted in temperate deciduous forest habitats tended to examine  
77 only the short-term effects of prescribed burning on epigeic ant communities (Verble  
78 and Yanoviak 2013) or analyzed a general response of soil invertebrates to prescribed  
79 burning (Kalisz and Powell 2000).

80 Here, I investigate the effects of prescribed fire on the structure of epigeic ant  
81 assemblages (species composition and species richness) up to 1 year following a burn in  
82 an eastern deciduous forest in Kentucky, USA. I expected ant species richness to be  
83 lower in burned forest (Underwood and Quinn 2010, Verble and Yanoviak 2013). I also  
84 expected ant species composition in burned forest to be distinct from that of unburned  
85 forest (Beaumont et al. 2012).

## 86 **Methods**

### 87 *Study Site*

88 Field work was conducted at Berea College Forest, a 3642-hectare mixed  
89 mesophytic forest in Berea, Kentucky (37.5687° N, 84.2963° W). Berea Forest is  
90 managed in part via an annual prescribed burn program (Patterson and Singleton  
91 2019). This study focused on the effects of two annual burns conducted in April (2021  
92 and 2022). In each case, only one section of forest was burned, an adjacent unburned  
93 section of equal area was used as a control site and burn, and control sites did not  
94 overlap between years.

### 95 *Experimental Design*

96 Following each prescribed burn, I established three transects in the burned site  
97 and three similar transects in the adjacent control site. Each transect was 10 m long and

98 separated from other transects by > 100 m. Hereafter, *treatment* refers to burned vs.  
99 unburned (control) forest and *year* refers to 2021 vs. 2022.

100 I used pitfall traps to sample epigeic ants along each transect. Each trap  
101 consisted of a 475 ml plastic cup (model No. S-22770, Uline, Pleasant Prairie, WI, USA),  
102 buried such that the cup opening was flush with the soil surface. A second cup was  
103 placed inside the buried cup to facilitate sample collection. Each buried cup assembly  
104 was sheltered with a roof of acrylic sheeting (ca. 0.6 x 15.2 x 15.2 cm) suspended 2.5 cm  
105 above the cup opening with landscape staples (Amagabeli, Beijing, China). A small  
106 amount of antifreeze (Model: RV and Marine antifreeze -50 °F, Splash, St. Paul, MN,  
107 USA) mixed with dish soap added to each trap served as the killing agent (Fig. 1).

108 In 2021, pitfalls traps were placed every meter on alternating sides of the transect  
109 for all six transects (Fig. 2). I placed 30 pitfall traps in burned forest and 30 pitfall traps  
110 in unburned forest, which were sampled weekly. In 2022, I sampled burned and  
111 unburned plots of forest following the 2022 burn along with the burned and unburned  
112 plots established in 2021. I reduced sampling intensity to 10 pitfalls (3, 3, and 4 traps for  
113 each transect) in each plot, resulting in a total of 40 pitfalls across 4 different plots in  
114 2022 (Fig. 2), which were sampled monthly.

### 115 *Statistical Analyses*

116 Analyses include only trap data where at least one ant was present. I did not use  
117 raw abundance as a response due to the social nature of ants (Wilson and Hölldobler  
118 2005). Instead, I used the total number of unique occurrences (i.e., the number of times  
119 a species was recorded in a pitfall trap through the entire sampling period.) The total  
120 number of unique occurrences for each species was pooled over time for each pitfall  
121 trap.

122 To determine whether the number of unique ant occurrences differed between  
123 treatments and year, I constructed a generalized linear mixed effect model (GLMM)  
124 with a negative binomial distribution to account for the extreme right-skew in the data. I  
125 included the interaction term between the main effects of treatment and year and  
126 included plot as a random effect (Table 1). I did not include days post-fire as an  
127 explanatory effect because pitfall data were pooled across samples within a year.

128 I used non-metric multidimensional scaling (NMDS) ordination to determine  
129 whether ant assemblages in burned forest were compositionally distinct from those in  
130 unburned forest (Fig. 3; PRIMER Software Version 7.0.21). I created a Bray-Curtis  
131 dissimilarity matrix using the number of unique occurrences for each species within  
132 each pitfall trap as the response. To determine whether forest status, year, and their  
133 interactions had a significant effect on ant communities, I used permutational  
134 multivariate analysis of variance (PERMANOVA; Table 2). I then ran a permutational  
135 test of dispersion (PERMDISP) to determine whether the potential differences were  
136 attributable to the main effects or to dispersion within the data (Table 3).

137 I conducted a post-hoc test to determine which treatment and year combinations  
138 were significantly different from each other (Table 4). I used Bonferroni adjusted p-  
139 values to account for multiple comparisons.

## 140 **Results**

141 I collected 4,947 ants comprised of 23 species and 13 genera over the 21 months  
142 of sampling. The total number of ants collected was distributed among 100 trap  
143 occurrences, for an average of 49.5 ants per trap collection over the course of the study.  
144 In most cases, each trap contained relatively few species. The most frequently collected  
145 species was *Aphaenogaster carolinensis* (n = 986 occurrences), and many species were  
146 represented by a single individual (e.g., *Camponotus castaneus*, *Formica pallidefulva*,  
147 *Proceratium chickasaw*, *Strumigenys ohioensis*, *Temnothorax curvispinosus*, and  
148 *Temnothorax schaumii*; Fig. 4)

149 Most species were collected in both burned and unburned plots. *Aphaenogaster*  
150 *carolinensis*, the most abundant species, was collected approximately equally between  
151 burned and unburned plots. However, some “common” species (those represented by >  
152 1 occurrence) were collected exclusively from one treatment (e.g., *Lasius aphidicola* was  
153 collected only in unburned plots, while *Formica subcericea* was collected only in burned  
154 plots).

155 The sampling year had a significant effect on ant species richness (p-value = <  
156 0.001; Table 1; Fig. 5.), with the first year of sampling yielding a greater number of  
157 unique occurrences than a second year of sampling of a plot. However, treatment did

158 not have a significant effect on unique occurrences ( $p = 0.46$ ; Fig. 6) and there was no  
159 treatment x year interaction ( $p = 0.27$ ; Table 1).

160 Ant communities in burned plots were significantly different from communities  
161 in unburned plots ( $F = 23.94$ ,  $df = 1$ ,  $P = 0.001$  and  $F = 2.25$ ,  $df = 1$ ,  $P = 0.001$ ,  
162 respectively; Table 2; Fig. 3). Sampling year also resulted in a distinction between  
163 communities that were sampled during the first year of collection and communities  
164 sampled a year following the initial burn (Table 2; Fig. 3). Year and treatment had no  
165 significant effect in the PERMDISP test ( $F = 0.22$ ,  $P = 0.66$ ;  $F = 0.12$ ,  $P = 0.72$ ; Table 3),  
166 indicating that observed differences were attributable to the main effects, not dispersion  
167 within the data.

168 Ant community composition in the burned plot was significantly different from  
169 control site in 2021, but there was no treatment effect on composition in 2022 (Table 4).  
170 Ant composition also differed between the two burn plots (Table 4). Ant composition  
171 also differed between the 2021 and 2022 samples from a site that was burned in 2021,  
172 and between its associated control samples (Table 4).

## 173 **Discussion**

174 The result of this study generally agrees with those of similar studies, specifically,  
175 that prescribed fire alters epigeic ant communities (Izhaki 2003, Underwood and Quinn  
176 2010). However, it contrasts with other studies indicating that ant species composition  
177 does not differ between burned and unburned plots (Verble and Yanoviak 2013). Pitfall  
178 traps measure ant activity, so these results can also be explained by a decrease in ant  
179 activity, not necessarily extermination of local species.

180 I suspect that the differences in year are due to differences in burn intensity  
181 between 2021 and 2022 (Table 4). The 2022 burn appeared to be of a much lower  
182 intensity. Specifically, the vegetation appeared less burnt and there was more leaf litter  
183 present following the 2022 burn in vs. the 2021 burn. However, these differences in  
184 anecdotal and speculative at this point because supporting data regarding fire intensity  
185 are lacking. Previous studies determined that prescribed burn intensity influences ant  
186 ecology (Underwood and Quinn 2010, Verble 2012).



187           The overall negative effect of prescribed burning on ant species richness has been  
188 illustrated in similar systems (Izhaki et al. 2003, Underwood and Quinn 2010).  
189 Contrasting studies, however, indicate that prescribed burning can increase ant  
190 occurrences and species richness (Beaumont et al. 2012, Banschbach and Ogilvy 2014)  
191 or have temporary effects on ant species composition (Verble and Yanoviak 2013). The  
192 variety of responses of epigeic ants to prescribed fire might reflect ecosystem-level  
193 differences (Vasconcelos et al. 2016). The results of this study suggest that most ant  
194 species are resilient to prescribed burns. However, pitfall traps do not provide a  
195 complete picture on the effects of fire on ants, thus further study of individual species  
196 responses is needed to clarify the short- and long-term ecological effects of fire.

197           The differences between communities at the 2021 burned and unburned plots  
198 one year after the initial burn suggest that ant assemblages require more than a year to  
199 recover from fire. Communities within the 2021 burn plot are not resilient enough to  
200 rebound within a year. This finding contrasts with a similar study that suggests epigeic  
201 ant communities in deciduous forest are relatively resilient and can return to normal  
202 activity levels less than a year after the initial prescribed burn (Verble and Yanoviak  
203 2013). The significant difference in the 2021 control plot from 2021 to 2022 could be  
204 due to a variety of factors, including source/sink dynamics from the adjacent burn plots,  
205 natural turnover, or environmental differences between the years (Crist 2009).

206           In summary, fire intensity likely influences ant activity levels, but future  
207 measurements of the intensity of prescribed burns is necessary to confirm this. As  
208 prescribed burning is increasingly being added to forestry management to promote  
209 plant diversity (Richter et al. 1982, Mitchell et al. 2006), it is important to document its  
210 effects on common consumer taxa like ants. Although the effect of prescribed burning  
211 on ants has been studied in many other ecosystems (Izhaki et al. 2003, Andersen et al.  
212 2009, Underwood and Christian 2009, Houdeshell et al. 2011, Beaumont et al. 2012,  
213 Menke et al. 2015, Bonoan and McCarthy 2022), additional data are needed for  
214 temperate deciduous forests, as they are expansive but steadily declining ecosystems  
215 (Loucks 1998).

216

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368 **Tables**

369 **Table 1.** Output of generalized linear mixed effect model with a negative binomial  
 370 distribution.

<b>Effect</b>	<b>Term</b>	<b>Estimate</b>	<b>Std. Error</b>	<b>Z-value</b>	<b>p-value</b>
Intercept	Fixed	2.21	0.19	11.36	< <b>0.001</b>
Year	Fixed	-0.52	0.15	-3.52	< <b>0.001</b>
Status	Fixed	0.20	0.27	0.75	0.46
Year:Status	Interaction	-0.23	0.21	-1.11	0.27
Site	Random	0.04	NA	NA	NA

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374 **Table 2.** Permutational multivariate analysis of variance (PERMANOVA) output.

<b>Effect</b>	<b>Term</b>	<b>df</b>	<b>Sums Squares</b>	<b>F-value</b>	<b>P(perm)</b>
Year	Fixed	1	2.83	23.94	<b>0.001</b>
Status	Fixed	1	0.27	2.25	<b>0.001</b>
Year:Status	Interaction	1	0.14	1.16	0.25
Site	Random	NA	NA	NA	NA
Residuals	NA	95	11.22	NA	NA
Total	NA	98	14.45	NA	NA

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378 **Table 3.** Permutational multivariate analysis of dispersion (PERMDISP) output.

<b>Effect</b>	<b>Term</b>	<b>Sums Squares</b>	<b>F-value</b>	<b>P(perm)</b>
Year	Fixed	0.005	0.22	0.66
Residuals	NA	2.04	NA	NA
Status	Fixed	0.002	0.12	0.72
Residuals	NA	0.02	NA	NA

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381 **Table 4.** Output of post-hoc test with a Bonferroni correction comparing burned and  
 382 unburned communities with the 2021 and 2022 prescribed burns. B1 refers to the 2021  
 383 burn and B2 refers to the 2022 burn. Yr1 refers to the first sampling period and Yr2  
 384 refers to the second sampling period. B refers to burned forest and U refers to unburned  
 385 forest.

Pairs	df	Sums Squares	F-value	R <sup>2</sup>	p-value	adjusted p-value
B1_Yr1_B vs. B1_Yr2_B	1	2.36	34.05	0.47	0.001	0.005
B1_Yr1_B vs. B2_Yr1_B	1	1.87	27.41	0.42	0.001	0.005
B1_Yr1_B vs. B1_Yr1_U	1	0.31	5.58	0.09	0.001	0.005
B2_Yr1_B vs. B2_Yr1_U	1	0.25	2.58	0.13	0.034	0.17
B1_Yr1_U vs. B1_Yr2_U	1	2.25	33.84	0.47	0.001	0.005

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388 **Table 5.** Summary of taxa collected and their corresponding occurrences in pitfall traps  
 389 in burned or unburned forest.

Taxon	Burned	Unburned
<b>Amblyoponinae</b>		
Amblyoponini		
<i>Stigmatomma pallipes</i>	4	3
<b>Formicinae</b>		
Camponotini		
Camponotus americanus	129	54
Camponotus pennsylvanicus	78	127
Camponotus chromaiodes	124	81
Camponotus subbarbatus	8	16
Camponotus castaneus	1	0
Formicini		
Formica pallidefulva	1	0

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405	<i>Formica subsericea</i>	33	2
406	<i>Formica subintegra</i>	1	0
407			
408	Lasiini		
409	<i>Lasius aphidicola</i>	0	19
410	<i>Prenolepis imparis</i>	2	0
411			
412	Plagiolepidini		
413	<i>Brachymyrmex depilis</i>	2	7
414			
415	<b>Myrmicinae</b>		
416	Attini		
417	<i>Strumigenys ohioensis</i>	0	1
418	<i>Strumigenys talpa</i>	1	1
419			
420	Crematogastrini		
421	<i>Crematogaster cerasi</i>	60	52
422	<i>Crematogaster pilosa</i>	25	19
423			
424	Formicoxenini		
425	<i>Temnothorax curvispinosus</i>	1	0
426	<i>Temnothorax schaumii</i>	0	1
427			
428	Stenammini		
429	<i>Aphaenogaster carolinensis</i>	497	487
430	<i>Aphaenogaster fulva</i>	24	43
431			
432	<b>Ponerinae</b>		

433	Ponerini		
434	<i>Ponera pennsylvanica</i>	7	6
435			
436	<b>Proceratiinae</b>		
437	Proceratiinae		
438	<i>Proceratium chickasaw</i>	1	0
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444 **Figures**

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451 **Fig. 1.** Pitfall trap placed in burned portion of forest.

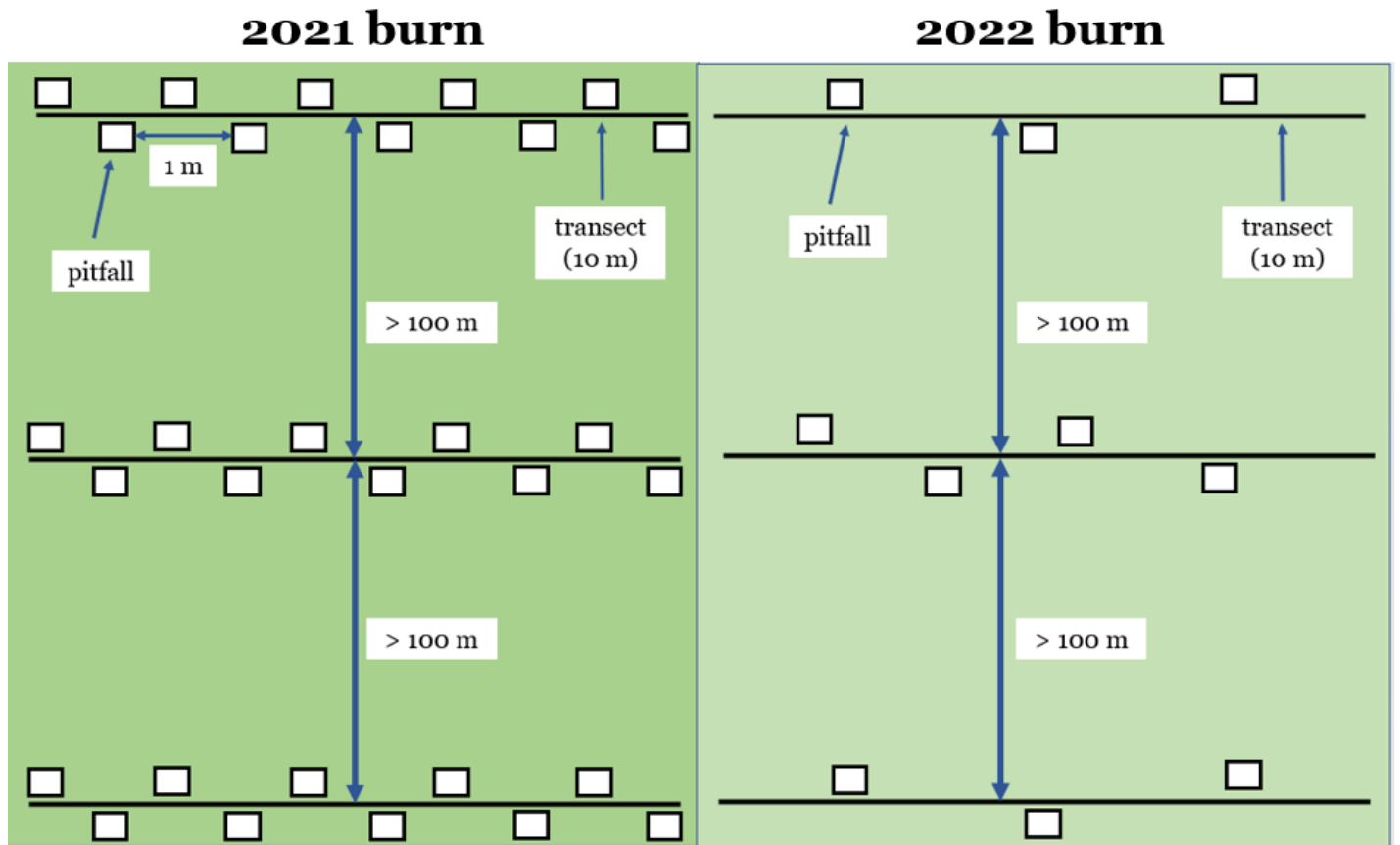
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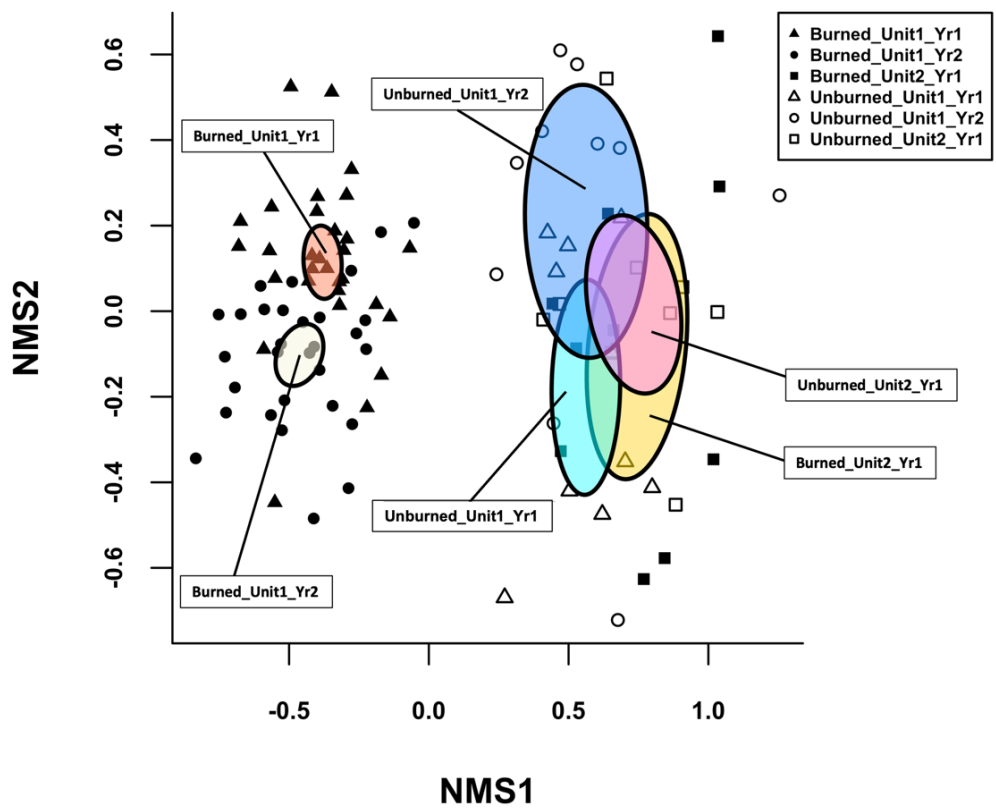
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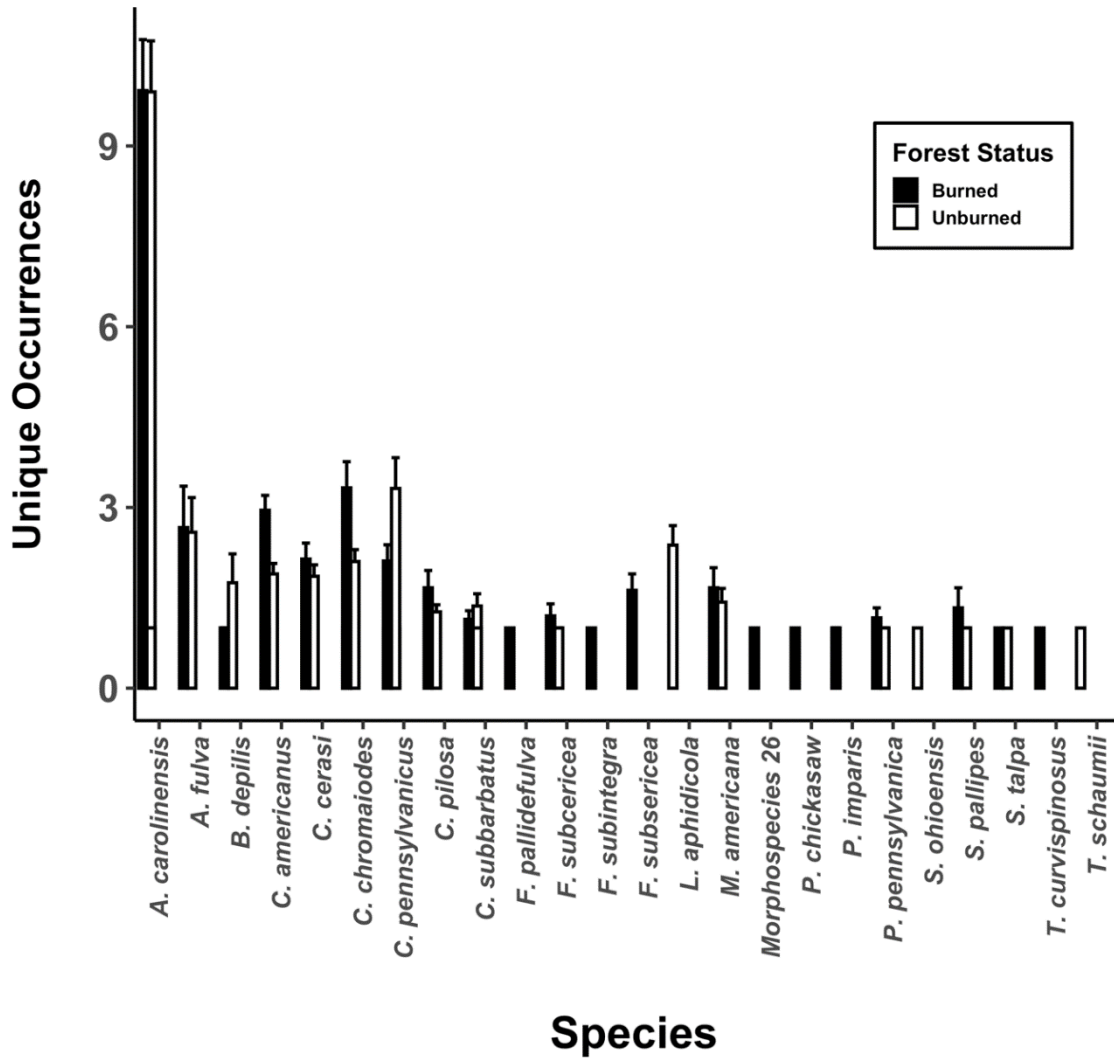
463 **Fig. 2.** Pitfall setup for both forest treatments (burned vs. unburned). The first panel  
464 symbolizes the layout of pitfall traps during sampling of the 2021 burn among burned  
465 and unburned plots. The second panel symbolizes the layout during 2022 sampling of  
466 the burned and unburned 2022 burn plots, and continued sampling of burned and  
467 unburned 2021 burn plots. White rectangles represent individual pitfall traps and black  
468 lines represent 10 m transects.

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**Fig. 3.** Non-metric multidimensional scaling (NMDS) ordination plot illustrating differences in ant species composition between forest treatment (burned vs. unburned) and collection year (2021 vs. 2022).

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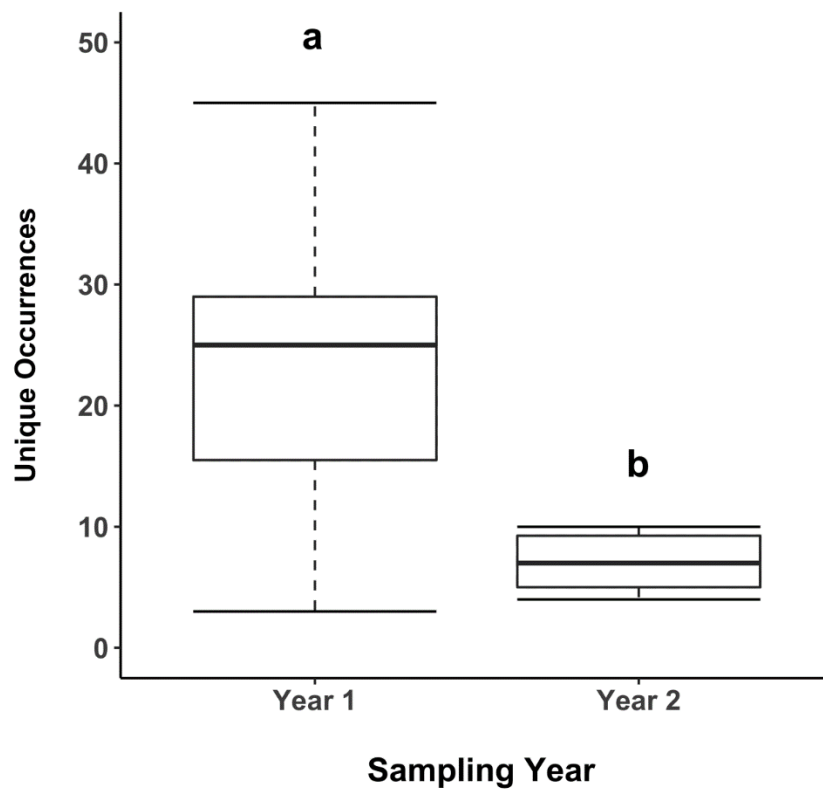


496 **Fig. 4.** The number of unique species occurrences ( $\pm 1$  SE error bars) by each ant  
497 species collected in this study.

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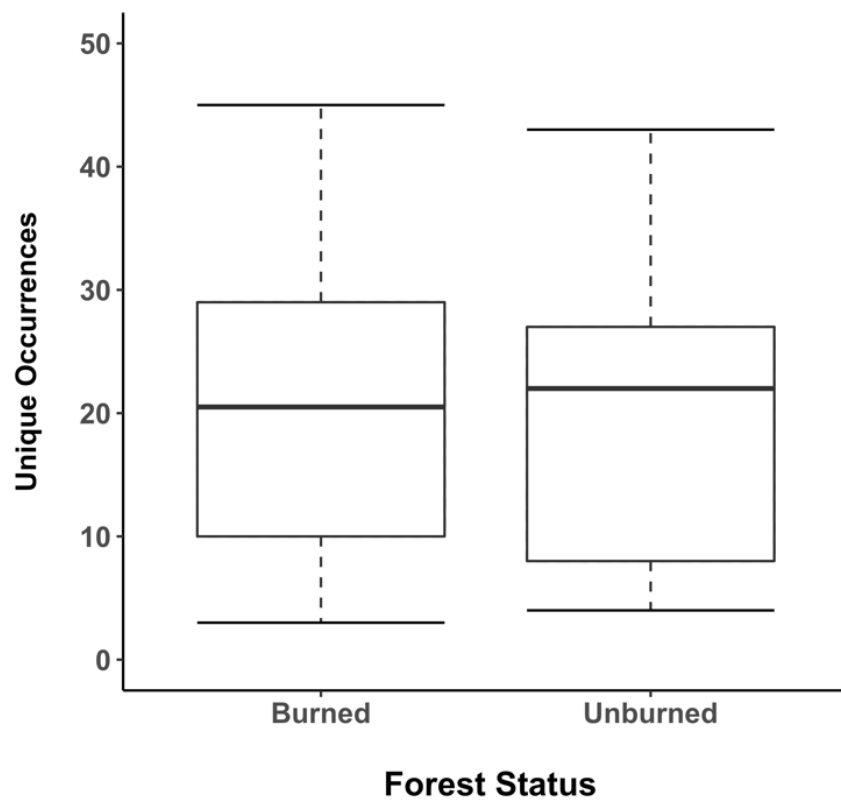


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**Fig. 5.** Box and whisker plot illustrating the pooled data for each pitfall trap and the unique occurrences across sampling years.

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548 **Fig. 6.** Box and whisker plot illustrating the pooled data for each pitfall trap and the  
549 unique occurrences in burned and unburned forest traps.

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