Chapter 9. Diversity of ants in burned and unburned grassland, and dry deciduous forest in the Beanka Reserve, Melaky Region, western Madagascar

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Abstract

Our study of ant diversity in the dry deciduous forest and anthropogenic grassland mosaic of Beanka Forest, Région Melaky, central western Madagascar, documents the difference in species richness between these habitats. In grassland sites, ants were surveyed using standardized methods along a transect (mini-Winkler associated with leaf litter sifting, pitfall traps, soil monoliths, and baiting), as well as with non-standardized general collecting. In the forest, ants were surveyed using only standardized maxi-Winkler transects and non-standardized general collecting. We found that ant diversity was far greater in forest (94 spp.) than in grassland (33 spp.), including those grassland sites that have recently experienced the passage of fire. Only 15 species were shared between the forest and grassland sites. A number of restricted range species were found in the dry forest, while all grassland species had wide distributions and included 14 (42%) introduced species. The efficiency of the collecting techniques in grassland areas differed between recently burned and not recently burned sites. The shape of the species accumulation curves indicate that a combination of pitfall and soil monoliths (19 spp.) was more efficient in burned grassland, while the "Ants of Leaf Litter" (ALL) protocol (pitfall and leaf litter sifting, with 23 spp.) was more efficient in grassland not recently burned. Species composition also differed between recently and not recently burned grasslands. About 43% of species (13 spp.) found in not recently burned grassland were absent from the recently burned grassland sites. With respect to grasslands, our findings indicate that not recently burned sites have greater species richness. Efforts to prevent grassland encroachment in Beanka will help to protect a considerable number of ant species.

Key words: grasslands, dry deciduous forest, *tsingy*, ants, diversity, methods

Résumé détaillé

Le niveau de dissimilarité entre la biodiversité présente dans la forêt sèche et dans les prairies secondaires est très apparent en milieu tropical. Une différence est aussi observable selon l'impact du feu sur la faune des prairies secondaires. L'étude de la biodiversité des fourmis dans la forêt sèche de Beanka et sur les prairies secondaires environnantes a permis de confirmer ces affirmations. En prairie secondaire, un site récemment brulé et un site non brulé ont été choisis. Les fourmis ont été échantillonnées par quatre méthodes quantitatives standardisées (le tamisage des litières mini-Winkler, le pitfall, le monolithe et les pièges à appât) et une méthode non standardisée : la collecte systématique. L'efficacité de ces méthodes a été également évaluée. En forêt, deux sites ont été choisis et les fourmis ont été échantillonnées avec une méthode standardisée du tamisage des litières : le maxi-Winkler et la méthode non standardisée : la collecte systématique.

Pour l'ensemble des sites étudiés, il en résulte d'une part que c'est dans la forêt que la diversité de la myrmécofaune a été la plus élevée avec 94 espèces contre 33 espèces dans les prairies secondaires. Le nombre d'espèces partagées a été de 15. Une moyenne de 60 espèces endémiques a été inventoriée dans la forêt de Beanka. Une estimation a été proposée puisqu'à l'heure actuelle, une partie de la systématique des principaux genres et espèces de fourmis malgaches est encore sous révision. Dans les prairies secondaires par contre aucune espèce endémique n'a été recensée. La majorité des ces espèces de prairies secondaires avait une large distribution dans d'autres prairies et forêts de Madagascar. Treize espèces de fourmis exotiques ont été répertoriées sur l'ensemble des sites. Dix d'entre elles ont été collectées dans la forêt contre huit dans les prairies secondaires.

Dans les prairies secondaires, d'après l'allure de la courbe d'accumulation des espèces, l'efficacité des méthodes utilisées varie selon l'état de la végétation, ayant subi ou non de l'influence du feu. En prairie

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récemment brûlée, la combinaison du pitfall et du monolithe était la plus efficace (19 spp.). Par contre, en prairie non brûlée pendant au moins trois ans, le protocole « Ants of Leaf Litter » (ALL) (pitfall + mini-Winkler) a été l'assemblage de méthodes le plus efficace avec 23 espèces. Par ailleurs, le niveau de similarité des espèces trouvées dans les deux types de prairies était différent. Près de la moitié des espèces répertoriées en prairie non brûlée (13 spp.) ont été trouvées uniquement dans ce type de prairie.

Cette étude permet d'affirmer que seules une combinaison de méthodes d'échantillonnage est adéquate pour faire un inventaire des fourmis dans les prairies brulées et non brulées de Beanka. L'historique des prairies aux environs de Beanka a montré que celle-ci sont potentiellement brulées chaque année. Cependant, la fréquence du feu est différente en fonction de la proximité d'un village. Cette comparaison a montré que les fourmis sont plus diversifiées dans les sites non brulés que dans les sites brulés. Enfin, comme aucune espèce endémique n'a été recensée, cette étude conforte l'hypothèse d'une origine anthropique des prairies de Beanka. Une origine très ancienne de ces prairies, du type de celle avancée pour les Hautes Terres centrales du côté d'Ankazomivady à Ambalavao ou consécutives à la distribution des graminées de type C4 à Madagascar est à exclure. Cette étude a permis les recommandations suivantes : la mise en place d'une bonne gestion du feu dans les prairies pour préserver la diversité, pour épargner le risque d'extension du feu dans la forêt et pour la protéger contre les espèces exotiques.

Mots clés : Prairies secondaires, forêt sèches, *tsingy*, fourmis, diversité, méthodes

Introduction

The high endemicity of the fauna and flora of Madagascar is often cited in the biodiversity literature (Myers *et al.*, 2000; Johanna & Niemel, 2006). However, the natural habitat of this exceptional biota is threatened by human activities. In 1950, 27% of Madagascar was estimated to be covered by forest and this rate declined to 16% by 2000 (Harper *et al.*, 2007). Major forest habitat losses have occurred because of inadequate management of natural resources. For example, the recent and massive deforestation of eastern Madagascar is often ascribed to traditional agricultural practices such as field rotation or slash-and-burn agriculture (Humbert, 1927; Green & Sussman, 1990; Randriamalala, 2009). Providing data on the patterns of species richness in human modified ecosystems, particularly with respect to native versus introduced species, will help better manage the remaining natural areas in Madagascar. Several studies have examined this question through the lens of entomology, specifically concerning ants (Fisher & Robertson, 2002; Dejean *et al.*, 2010; Rakotonirina, 2010) and beetles (Johanna & Niemel, 2006). Further, important insights on these aspects have been provided by research of agricultural practices on plant succession at the forest-grassland ecotone (Randriamalala *et al.*, 2008; Randriambanona, 2008). These studies demonstrate that in most anthropogenic landscapes, biodiversity decreases for both native plants (Lowry *et al.*, 1997; Binggeli, 2003) and mammals (Goodman & Rakotondravony, 2000; Irwin *et al.*, 2010). In general, most species react negatively to anthropogenic disturbance, although specific details on how they respond remains poorly known.

Ants have the potential to play an important role in conservation monitoring programs (Underwood & Fisher, 2006). A number of research projects studying ant biodiversity have been conducted across different habitat types on Madagascar (e.g., Fisher, 1997, 1999; Rakotonirina, 2010). In the Central Highlands, where the landscape is composed of a mosaic of grassland and relict forest, Fisher & Robertson (2002) found that ants have similar diversity in adjacent grassland and montane forest sites; both site types hold endemic species. This was one of a few studies to date indicating the presence of microendemic ant species in the island's upland grasslands and the occurrence of endemic ants in these formations is contrary to the prevailing hypothesis that such habitats on Madagascar were anthropogenically derived (Willis *et al.*, 2008; cf. Bond *et al.*, 2008). Population genetic studies on isolated lemur populations also suggested that perhaps some grassland formations are older than the arrival of the first humans (Quéméré *et al.*, 2012). Thus, the question remains if grasslands could be part of the natural landscapes on Madagascar.

To investigate further the biodiversity of grassland habitats, we initiated a program to replicate the Fisher & Robertson (2002) study in other grassland systems on the island, specifically lowland areas in central western Madagascar, which have important levels of endemic biota. In this paper, we present research conducted in central western Madagascar dry deciduous forest, recognized for its high rates of plant and animal endemism, and surrounded by grasslands that are regularly burned and of presumed anthropogenic origin (Lowry *et al.*, 1997). We have chosen the Beanka Forest and its surrounding grasslands to address the following questions: How diverse are ants in dry forests and in the adjacent anthropogenic grasslands surrounding these forests? How do fires affect grassland ant diversity? Which sampling methods are best for collecting ants in grasslands? Does the period since the last passage of fire in a grassland zone influence its ant diversity? To address these questions, we used several different methods to sample ants: pitfall traps, leaf litter sifting, baiting, soil monoliths, and hand collecting.

Methods

Study sites

Beanka Forest is located in Belitsaka Commune, Melaky Region. The forest includes a north-south band of dry forest with *tsingy* outcrops (Chatelain *et al.*, 2013); *tsingy* are water-eroded limestone formations that form sharp, needle-like pinnacles. This dry deciduous forest formation is surrounded by extensive grasslands, portions of which are burned annually (Figure 9-1). The climate alternates between a hot, wet season from late November to April and a warm, dry season that predominates during the rest of the year. The mean annual temperature is between

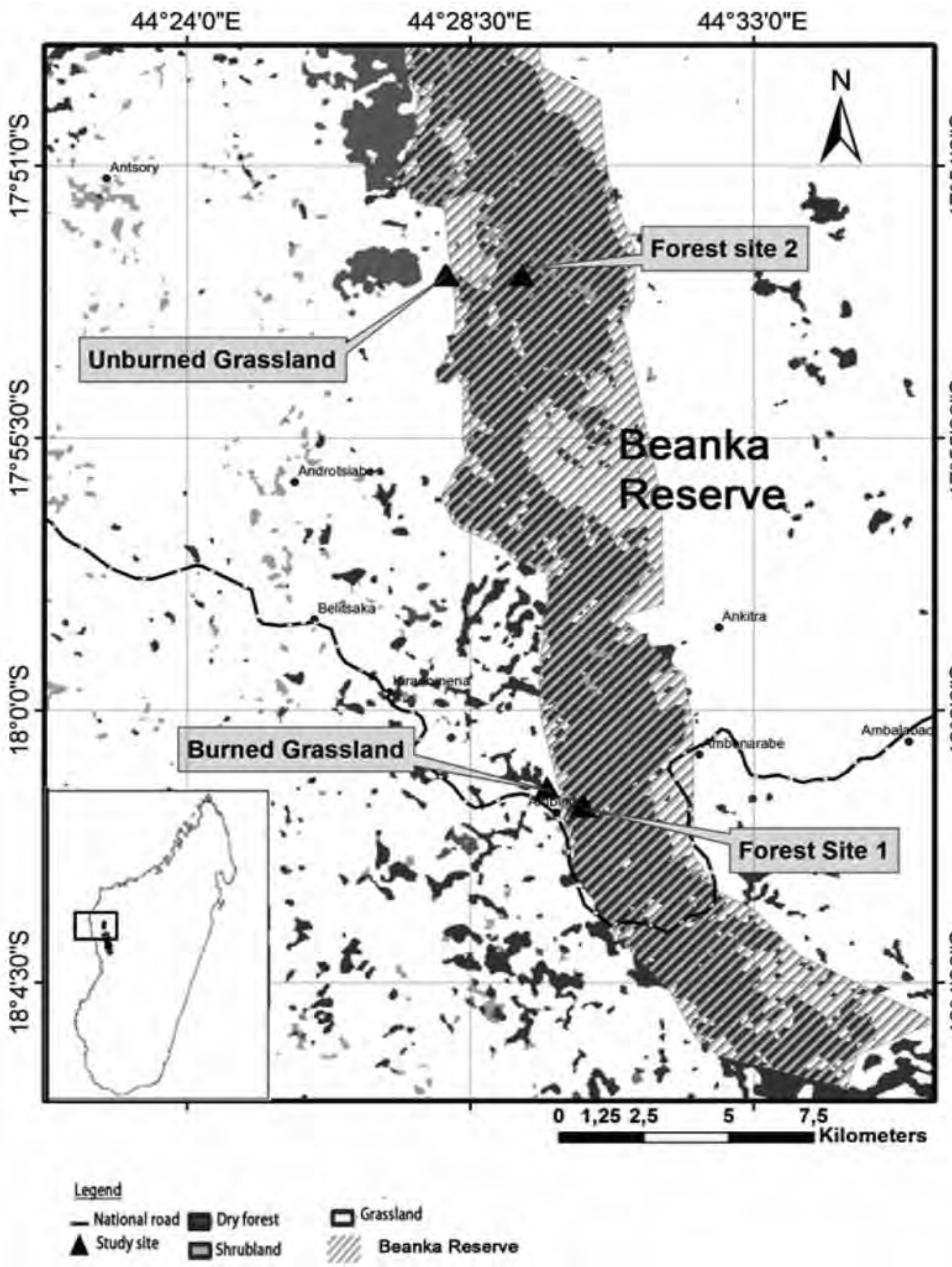

Figure 9-1. Map of sampling sites in the Beanka Reserve.

26°C and 28°C, and mean monthly temperatures remain above 20°C. Extremes of 38°C and 9°C have been registered in December and July, respectively (UNESCO, 2011). To the south, the Beanka Forest is adjacent to the Tsingy du Bemaraha reserve complex, an area classified by UNESCO as a Natural World Heritage Site for its exceptional ecosystems.

Sampling was conducted 19 to 31 October 2009, at the end of the dry season. Four sites were inventoried, two in grasslands and two in dry deciduous forest:

1) Recently burned grassland occurring at 250 m, last burned in December 2008 (18.0213°S, 44.4947°E),
2) Not recently burned (unburned hereafter) grassland situated at 160 m, not burned for at least three years before the study (17.8873°S, 44.4711°E),
3) Forest site 1 was situated at 250 m, dry deciduous forest on *tsingy* limestone (18.0264°S, 44.5051°E), and
4) Forest site 2 was situated at 160 m altitude, dry deciduous forest on *tsingy* limestone (17.8802°S, 44.4887°E).

Each grassland site was separated from the forest edge by a distance of at least 250 m straight ground distance. According to the elders in the local community, the grasslands sites included in this study have been this type of formation for at least 50 years, perhaps notably older. Topopgraphic maps produced by the Institut Cartographique de Madagascar in 1964 based on aerial photographs of the region also indicate the grassland sites surveyed in 2009 were already present.

Sampling

Collecting techniques

In the recently burned and unburned grassland sites, we used the following standardized sampling protocol: 25 mini-Winkler leaf litter samples, 25 pitfall traps, 25 soil monoliths, and 25 baiting stations (details presented below). In addition, opportunistic general collecting was also conducted. For all methods, with the exception of general collecting, we used a transect of 250 m to assure the collection of a maximum variety of ants in different micro-habitats. Within each site, transects were separated from one another by a lateral distance of 50 m to avoid auto-correlation effects. Each transect consisted of 25 stations at intervals of 10 m.

At the two forest sites, we used a different size Winkler sack, referred to as a maxi-Winkler. The maxi-Winkler sacks take four times the amount of leaf litter than the mini-Winkler sacks. At each of the forest sites, nine maxi-Winkler leaf litter samples were taken, in addition to general collecting.

Mini-Winkler – This method is particularly useful for collecting litter and soil fauna that are not readily captured in pitfall traps (Olson, 1991; Fisher, 1999). The leaf litter samples involved establishing 25 plots, each 1 m^2, at 10 m intervals along the transect line, for 25 bags of litter. The litter samples were collected using 1 cm-grid sieves. We collected a volume of 1 l on average at each station. Each litter sample was placed in a single mesh bag and left in the mini-Winkler sacks for 48 hours.

Maxi-Winkler – The maxi-Winkler trap holds four mesh bags, compared to the single bag for the mini-Winkler. We used nine plots, each 2 m^2, at 10 m intervals along the transect line, for a total of 36 bags of litter. These litter samples were left in maxi-Winkler sacks for 48 hours.

Pitfall trap – The pitfall trap consists of an open container, or pitfall tube, placed in a hole dug 20 cm deep and 11 cm in diameter. Tubes were placed at 10 m intervals along the transect. To avoid disturbance, each hole was dug with a shovel matching the pitfall tube size. Each tube was filled with 20 ml water, 40 ml of 90% ethanol, and three drops of liquid soap. This mixture minimizes capillary action and facilitates the immediate submersion of arthropods to the bottom of the traps. The pitfall traps were left in place for four days.

Soil monolith – This technique consists of digging a small hole (30 x 30 x 30 cm) and pouring the collected soil onto a white cloth, where the ants are separated from soil particles and other invertebrates, and placed in vials containing 95% ETOH. Each monolith was dug at 10 m intervals along the transect. This technique allowed us to find living underground colonies of ants that would have been missed by the pitfall trapping and Winkler sampling methods.

Baiting – This method consists of attracting the ants with a piece of dried and salted fish. Baits were placed at 10 m intervals. We employed this technique twice a day, at 8:00 and 20:00 h and collections were made for approximately 1 hour thereafter. Night baiting was conducted to collect ant species, which had lower daytime activity levels (mainly due to the dry season).

General hand collecting – General collecting was limited to ants living in particular microhabitats, such as rotten and broken pieces of fallen wood, dead and living branches, and under rocks. Around 1 h total was spent hand collecting at each site.

Data analysis

All specimens were identified to species using published keys (Blaimer, 2012; Garcia & Fisher, 2012; Rakotonirina & Fisher, 2013) or to a morphospecies code established for the ant fauna of Madagascar (AntWeb, 2013). Species accumulation curves were used to estimate the efficiency of the various sampling methods by comparing the shape of each curve (Longino & Colwell, 1997). Species accumulation curves were analyzed for each method (except general hand collecting) and at each site by plotting the observed number of species as a function of the number of samples, using the freeware application EstimateS (Colwell, 2006). Only records of ant workers were used in these analyses, since the presence of queens or males in samples does not necessarily signify the establishment of a colony of that species within the transect zone. Since the shape of the species accumulation curve can depend on the ordering of samples (Colwell & Coddington, 1994; Fisher & Robertson, 2002), all curves were smoothed through randomization.

Table 9-1. Ant species collected in burned (20 species) and unburned (30 species) grasslands at Beanka using mini-Winkler (W), pitfall (P), soil monoliths (SD), day baiting (BD), night baiting (BN), and general hand collecting (GC). Species introduced to Madagascar are indicated in bold.

Species	Burned grassland					Unburned grassland					
	W	P	BD	BN	SD	W	P	BD	BN	SD	GC
Aphaenogaster swammerdami		1					1		1		
Camponotus hova					1	1	1		1	1	
Camponotus imitator resinicola					1						
Camponotus MG 079						1			1		
Camponotus niveosetosus madagascarensis						1	1	1		1	1
Camponotus quadrimaculatus opocatus											1
Cardiocondyla emeryi		1				1					
Crematogaster dentata											1
Crematogaster kelleri	1	1									
Crematogaster ramamy					1	1	1				
Crematogaster sewellii		1		1	1				1		
Melissotarsus insularis											1
Meranoplus mayri							1	1		1	1
Monomorium clarinode		1									
Monomorium destructor						1	1	1	1	1	1
Monomorium madecassum		1	1				1		1		
Monomorium nigricans		1	1		1		1				
Monomorium robustior		1						1	1		
Monomorium shuckardi		1				1	1		1		
Nylanderia madagascarensis rufescens		1		1	1	1	1		1		
Pachycondyla ambigua					1						
Paratrechina longicornis	1					1	1	1	1	1	1
Pheidole megacephala		1	1	1	1	1	1	1	1	1	
Tanipone scelesta		1									1
Technomyrmex pallipes						1	1				
Tetramorium bessonii						1	1				
Tetramorium delagoense		1				1	1				
Tetramorium lanuginosum						1					
Tetramorium malagasy						1					
Tetramorium scytalum						1					
Tetramorium sericeiventre		1	1		1		1				
Tetramorium zenatum						1					
Tetraponera diana					1			1			
Number of species per method	**2**	**14**	**4**	**3**	**10**	**18**	**16**	**7**	**11**	**6**	**8**

Results

Efficacy of inventory methods

The total number of species collected in burned and unburned grassland sites without hand collecting was 20 and 26, respectively. The total number of species collected using all methods in grassland sites was 33 spp. (Table 9-1). At each of the two grassland sites, most accumulation curves for each method approached an asymptote (Figure 9-2). Soil monoliths in burned grassland are the exception; the curve continues to increase sharply after 25 samples. Overall, for the grassland sites, sampling was sufficient to compare efficiency of methods between burned and unburned sites. Species accumulations curves (Figures 9-2 & 9-3) indicate that efficiency of methods differed between burned and unburned sites.

In burned grasslands, pitfall traps (14 spp.) had a higher rate of species accumulation than any other single method (Figure 9-2a), followed by soil monoliths (10 spp.), and then baiting. Soil monoliths were the only method for which the shape of the curve was still rising sharply at the end of the transect.

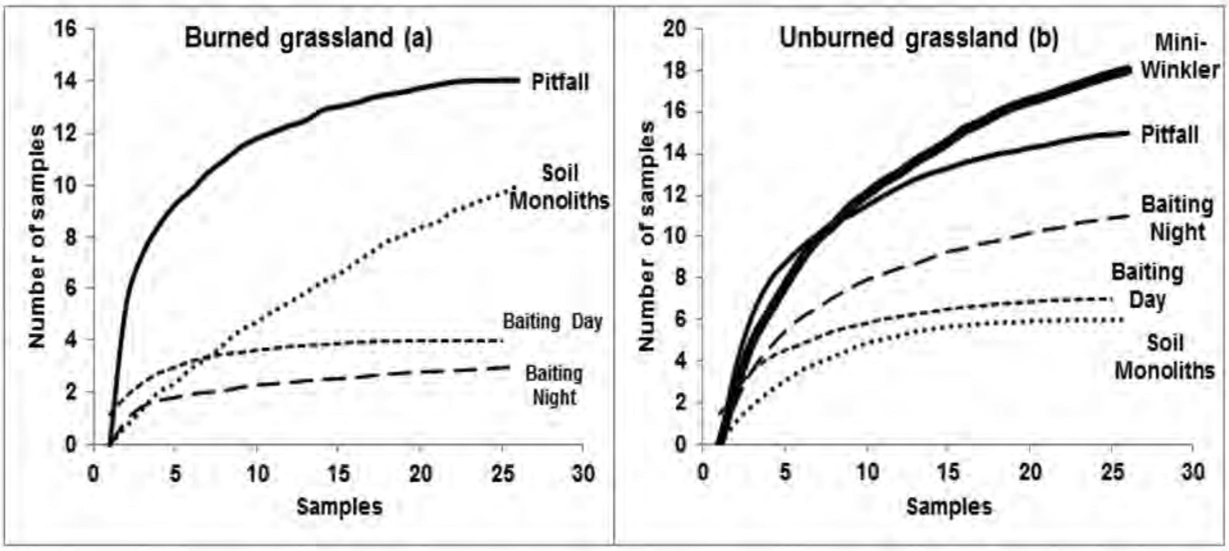

Figure 9-2. Species accumulation curves for ants at Beanka collected in recently burned (a) versus unburned grassland (b) sites.

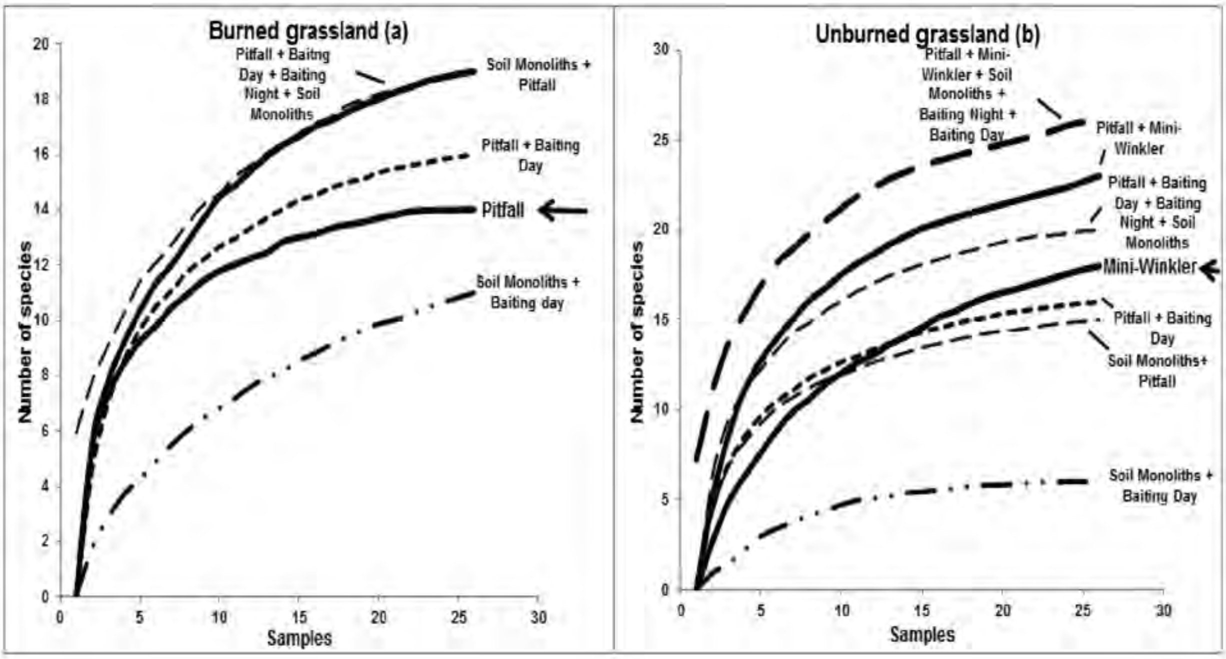

Figure 9-3. Comparison of different combinations of methods used at Beanka in burned (a) and unburned (b) grassland sites, indicating with arrows the most efficient method in burned (pitfall traps) and unburned (mini-Winkler) sites.

Baiting methods approached an asymptote at the end of the transect but its efficiency to inventory species was very poor and captured fewer than four species (Figure 9-2a).

At the burned sites, leaf litter was scarce and correspondingly mini-Winkler sampling only yielded two individual ants from the entire transect and the accumulation curve was omitted from Figure 9-2a and Figure 9-3. The most efficient combination of two methods was pitfall trapping and soil monoliths, which resulted in the capture of 19 spp. (Figure 9-3a).

At the unburned grassland sites, however, the single most efficient ant collecting method was the mini-Winkler, which yielded 18 spp. (Figure 9-2b). Pitfall trapping was the next most efficient method (15 spp.). Baiting at night was the next most efficient single technique (11 spp.). Both baiting during the day and soil monolith techniques resulted in species asymptotes at the end of transect, but collected relatively few species (Figure 9-2b).

Six different combinations of methods were evaluated (Figure 3b). The most efficient and productive combined approach included all methods (mini-Winkler + pitfall + soil monolith + baiting) with 26 spp. followed closely by the Ants of the Leaf Litter (ALL) protocol (mini-Winkler + pitfall) with 23 spp.

At the forest sites, the shape of the species accumulation curve continued to rise steeply at the end of the survey (Figure 9-4). The number of species captured using the maxi-Winkler at forest site 1 was 47 spp. and at forest site 2 was 30 spp. Overall, 94 taxa were found at the forest sites, including specimens obtained from hand collecting (Table 9-2). The results suggest that the diversity of ants occurring in the Beanka Forest is far greater than the 94 species collected in this study.

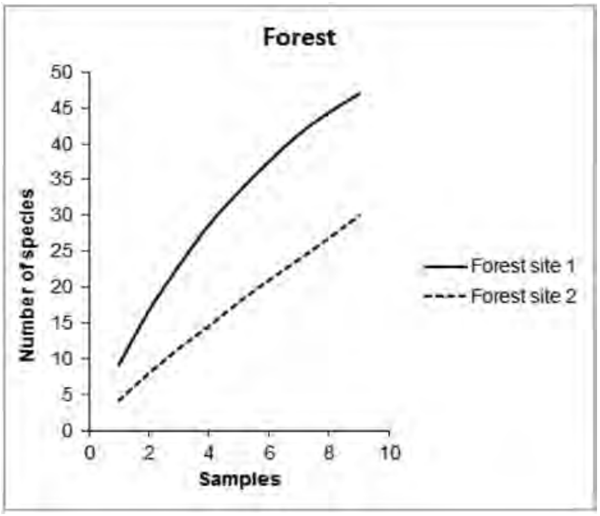

Figure 9-4. Species accumulation curves for ants in the dry deciduous forest of Beanka collected with maxi-Winkler method.

Comparison of grassland and dry forest ant richness and composition

Grassland sites harbored far lower ant diversity than dry forest sites. A total of 33 spp. was found in the combined grassland sites using the four quantitative methods (mini-Winkler, pitfall, soil monolith, and baiting methods) and general collecting, while 94 species were found in dry forest sites using two methods (maxi-Winkler and hand collecting).

Within grassland sites, the level of species complementarity (uniqueness) from each method was high, on average 40% in burned site and 65% in unburned sites – each method (pitfall, soil monolith, and mini-Winkler) collected a unique suite of species.

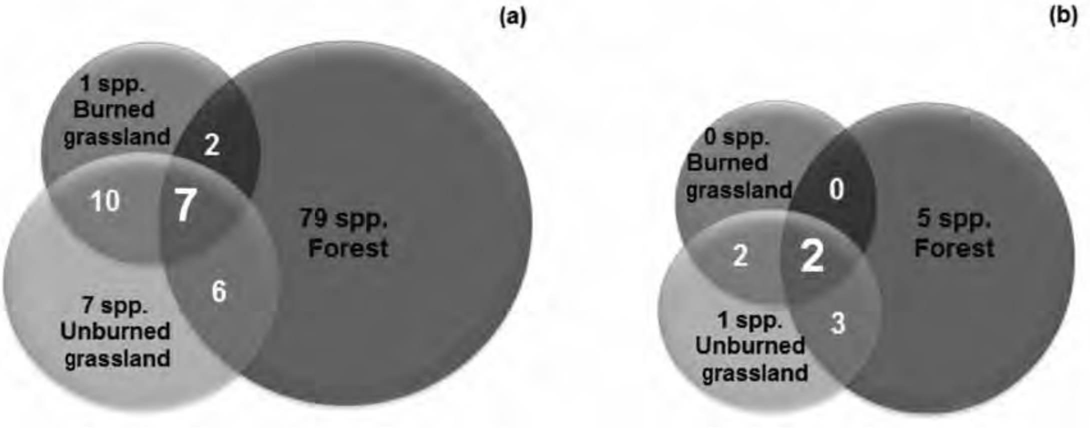

Figure 9-5. Comparison of shared species at Beanka in grasslands and dry forests for (a) all species collected and (b) introduced ant species. In total, 30 species were collected in unburned grassland, 20 in burned grassland, 94 in forest for 127 species in total, including 13 introduced species.

Table 9-2. Ant species collected in dry deciduous forest sites 1 and 2 at Beanka based on maxi-Winkler (W) and general hand collecting (GC). Species introduced to Madagascar are indicated in bold.

Species	Forest site 1		Forest site 2	
	GC	W	GC	W
Anochetus grandidieri		1		
Anochetus madagascarensis			1	
Aphaenogaster swammerdami	1		1	1
Camponotus darwinii			1	
Camponotus hova	1			
Camponotus imitator resinicola	1			
Camponotus legionarium	1	1	1	
Camponotus MG 02	1		1	
Camponotus MG 05	1			
Camponotus MG 46	1	1	1	
Camponotus MG 66			1	
Camponotus MG 129	1			
Camponotus quadrimaculatus opacatus	1			1
Camponotus repens			1	
Carebara Beanka 01				1
Carebara FAS 01		1		
Carebara FAS 02		1		
Cataulacus ebrardi	1		1	
Cerapachys biroi				1
Crematogaster grevei	1			
Crematogaster kelleri	1		1	
Crematogaster lobata			1	
Crematogaster maina	1		1	
Crematogaster ramamy	1			1
Crematogaster rasoherinae	1	1		
Crematogaster tsisitsilo	1			
Discothyrea MG 03		1		
Hypoponera Beanka 01		1		1
Hypoponera indigens	1	1		1
Hypoponera johannae		1		
Hypoponera MG 066		1		
Leptogenys oswaldi	1	1	1	
Melissotarsus insularis	1			
Monomorium cryptobium		1		1
Monomorium destructor	1		1	1
Monomorium exiguum		1		1
Monomorium fisheri		1		1
Monomorium hanneli		1		1
Monomorium hildebrandti		1		
Monomorium lepidum	1	1		
Monomorium sakalavum	1	1		
Monomorium termitobium	1	1		1
Monomorium versicolor	1			
Myrmicine_genus16 MG 04		1		
Mystrium MG 03		1		
*Mystrium voeltzkowi*_complex		1		
Nesomyrmex Beanka 01	1		1	
Nesomyrmex Beanka 02	1			
Nesomyrmex Beanka 04			1	
Nesomyrmex Beanka 05	1			
Nylanderia Beanka 02		1		
Nylanderia bourbonica	1	1		
Nylanderia humbloti		1	1	1
Pachycondyla ambigua	1	1	1	1
Pachycondyla wasmannii	1	1	1	1
Paratrechina longicornis	1		1	1
Pheidole Beanka 01		1		
Pheidole Beanka 03	1			
Pheidole Beanka 04	1	1	1	1
Pheidole megacephala	1	1	1	1
Plagiolepis alluaudi	1	1	1	
Plagiolepis madecassa		1		
Plagiolepis MG 04				1
Platythyrea mocquerysi			1	
Prionopelta MG 03		1		1
Strumigenys alapa		1		
Strumigenys fanano		1		
Strumigenys fronto		1		
Strumigenys luca		1		
Strumigenys lucomo		1		1
Strumigenys mandibularis		1		1
Strumigenys seti		1		1
Tapinoma MG 03		1		
Tapinoma subtile	1			
Technomyrmex difficilis	1			
Technomyrmex madecassus	1			1
Technomyrmex pallipes			1	
Terataner Beanka 01			1	
Terataner MG 11	1		1	
Tetramorium arya				1
Tetramorium delagoense	1	1	1	1
Tetramorium malagasy	1	1	1	1
Tetramorium plesiarum		1		1
Tetramorium popell			1	
Tetramorium sada	1			
Tetramorium simillimum		1		
Tetramorium zenatum		1		
Tetraponera Beanka 01	1			
Tetraponera Beanka 02	1		1	
Tetraponera Beanka 03	1		1	
Tetraponera Beanka 05			1	
Tetraponera Beanka 06	1			
Tetraponera Beanka 07			1	
Tetraponera Beanka 08	1		1	
Number of species per method	**47**	**47**	**35**	**29**

Seventeen species were shared between burned and unburned (Figure 9-5a). At least eight subfamilies of ants were represented in the Beanka Forest, while only six of them occurred in grassland sites. In both habitats, the dominant species belong to the Myrmicinae and Formicinae (Figure 9-6).

The ant species found in dry forest versus grassland were very different; only 15 species were in common to both habitats (Figure 9-5a). Among the shared species were *Aphaenogaster swammerdami*, *Camponotus hova*, *Monomorium destructor*, and *Pachycondyla ambigua;* most with geographically broad distributions and common in anthropogenic habitats.

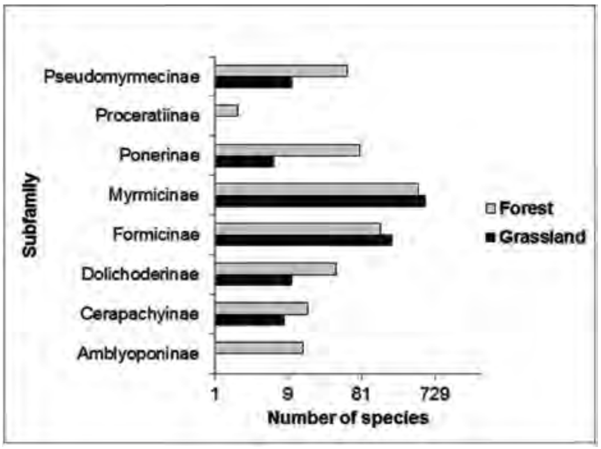

Figure 9-6. Comparison of ant subfamily diversity in the Beanka grassland and dry deciduous forest sites.

Our sampling efforts recorded 13 introduced species, 10 spp. in the dry deciduous forest and eight spp. in the grassland sites (Tables 9-1 & 9-2, Figure 9-5b). Among the introduced species were *Cardiocondyla emeryi*, *Monomorium destructor*, *Paratrechina longicornis*, *Pheidole megacephala*, and *Technomyrmex pallipes*.

Discussion

Efficiency assessment of the collecting methods

The species accumulation curves for all methods employed at the grassland sites approached an asymptote (Figure 9-2), indicating the number of subsamples for each method was sufficient to assess efficiency and species richness in burned and unburned sites. Thus, in the grassland sites, a nearly complete inventory was conducted, at least for the period of this study. The shape of the species accumulation curves differed between methods, however, indicating that the associated efficiency was different in burned versus unburned grassland.

In burned grassland, the curve showed that pitfall + soil monolith were the most efficient combination of methods (19 spp.). For individual collection methods, the highest number of species was captured by pitfall trapping followed by soil monolith (Figure 9-2). These two methods also complemented one another in terms of species captured; around 57% of species (7 spp.) collected were unique to one of the two methods. The productivity of the two methods can be explained by the fact that pitfall traps tend to capture ants in open areas, while soil monoliths allows direct sampling of ant nests in the ground. The lack of vegetation did not affect the efficiency of these two methods. Other methods used, such as baiting and mini-Winkler, did not enhance the diversity of inventoried taxa. Baiting sampled only the common dominant species (Cherry & Nuessly, 1992). In this study, the number of species captured was not influenced by the period (day or night) the sampling was conducted, or the period the baits were left out. The absence of leaf litter after a recent passage of fire clearly impacted the efficiency of mini-Winkler.

For unburned grassland, the accumulation curves showed that a combination of all methods (mini-Winkler, pitfall trap, baiting, and soil monolith) was the most effective technique, collecting 26 spp. The most efficient two-method combination was mini-Winkler + pitfall traps, which collected 23 spp. The pitfall + mini-Winkler combination corresponds to the methods recommended as part of the standard ALL protocol. For unburned grasslands, only minor additions were made by adding soil monolith and baiting to used survey methods, making pitfall and mini-Winkler the most efficient approaches. The most useful single collecting technique in unburned grassland was the mini-Winkler followed by pitfall traps, methods differing by only two species (Figure 9-2b). In terms of species diversity, the 35% of taxa collected by pitfall traps and mini-Winkler were different, which is why the combination of these two methods led to the capture of twice the number of species, as compared to a single method. In general, the results obtained by pitfall trapping were approximately the same in both types of grassland: 14 and 15 spp., respectively.

The utility of mini-Winkler and pitfall traps in grasslands has been documented in previous studies. In the Central Highland grasslands at Ankazomivady (Fisher & Robertson, 2002), ranked mini-Winkler, followed by pitfall trapping, as the most efficient methods. Pitfall traps have also often been highlighted in grassland studies, such as in the Vohibasia Forest, in the central southwest of

Madagascar (Fisher & Razafimandimby, 1997) and in South Africa (Parr & Chown, 2001). Baiting captured very few ants in our study and soil monolith captured fewer species in unburned grassland than in burned grassland. This result is unanticipated, suggesting that fewer species are nesting in the soil in unburned grasslands, even though overall species diversity may be higher in unburned areas. Our data indicated that the best methods to inventory ants in burned habitats include pitfall and soil monolith, while in unburned grassland, min-Winkler extraction and pitfall traps.

For dry forest sites, the species accumulation curve for maxi-Winkler continued to rise and did not approach an asymptote. This tendency is typical for tropical forests (Fisher, 1999; Gotelli & Colwell, 2010). Though more sampling is needed to ascertain the species richness of ants in dry forest, it is clear from this current study that dry forest diversity is greater than in grassland habitats and is characterized by species with more restricted geographical ranges.

Species diversity

The ant diversity in grasslands is unequally distributed. It depends on the condition of the vegetation, which is related to the last period a given zone was burned. Based on all methods, including hand collecting, more species were found in unburned (30 spp. total) than in burned grassland (20 spp. total) (Table 9-2). In addition, 50% of species occurring in unburned grassland were unique to that habitat. No endemic species was found at any of the Beanka grassland sites. The differences in ant assemblages in the two types of grasslands, with some species unique to unburned grassland sites, provides insights into the utility of ants as bio-indicators for monitoring grassland sites (Underwood & Fisher, 2006). While our results also support the importance of preserving grasslands from fire, this survey should be replicated at different sites with more extensive sampling to see whether this pattern holds. Most critical, fire prevention is needed to protect the Beanka Forest, as abusive or uncontrolled fires have played an important role in the loss of biodiversity and forest cover in other areas of the island (Bloesch, 1999; Ehrensperger *et al.*, 2013).

The diversity of ant species found in grasslands (33 spp.) is notably less than in the forest (94 spp.). This result is in stark contrast to that obtained at Ankazomivady in Madagascar's Central Highlands, where grassland habitats appear similar, but have a slightly higher diversity of ant species (31 spp.) compared to slightly degraded forest (26 spp.) (Fisher & Robertson, 2002). The differences between Beanka and Ankazomivady may be related to elevation, soil types, distance from forest, and the origin of these different grasslands. At Ankazomivady, the sites were in close proximity to the forest, which might provide access to greater resources to support a more diverse ant assemblage. The grasslands around Beanka appear to be anthropogenic in nature and do not represent a natural habitat type. In contrast, at Ankazomivady, grasslands systems may include some natural elements but have been enlarged by human activities and include ants that adapted to grassland habitats (Fisher & Robertson, 2002).

The seasonality of the survey might have influenced the relative diversity estimates obtained in grassland versus forest. The survey was conducted near the end of the dry season. The extreme dry condition may impact ant activity greater in the grassland than in the forest where the canopy protects the soil from direct sunlight. A great diversity of ants may have been captured if the survey was conducted during the rainy season, which was the case in the study by Fisher & Robertson (2002). Follow up studies will be needed to better understand the phenology of ants in western grassland habitats.

Our analyses suggest that the collection of 94 ant species in the two Beanka Forest sites does not represent the full local diversity in this habitat. The absolute and relative diversity of the ants collected at the two forest sites was greater than the difference between the burned and unburned grassland sites. As a point of comparison, 166 species were recorded in the *tsingy* dry deciduous forest of the Parc National Tsingy du Bemaraha, located 50 km south of Beanka (B. L. Fisher, unpublished data). The Bemaraha study includes a greater number of methods, which may in part explain the higher diversity. Interestingly, only 34 species were shared between Bemaraha and Beanka, indicating high turnover between these two *tsingy* habitats.

Resilience to fire

On Madagascar, fire is regarded as source of considerable disturbance for ants, as well as other biotic groups (Irwin *et al.*, 2010). Despite this factor, 17 species are apparently resilient to fire, including *Aphaenogaster swammerdami*, *Paratrechina longicornis*, *Pheidole megacephala*, *Tetramorium sericeiventre*, and *Meranoplus mayri* (Table 9-2) – all found in unburned and recently burned grassland.

These ants are also broadly distributed in Madagascar and in a wide range of habitats (AntWeb, 2013). The ecology of these 17 fire resistant ant species should be investigated to determine their role in grassland recovery after fire.

Capture rates for each method also differed for the species shared between burned and unburned grassland sites. For example, four taxa were only captured with soil monoliths at burned sites, while these same species were captured by pitfall and mini-Winkler, but not soil monoliths, at unburned sites. Thus, the same species may have different foraging behavior in the two habitats. This result also supports the conclusion that habit impacts the efficiency of different collection methods (Fisher, 1999).

Fire may also influence certain ecological processes, such as seed dispersal. *Aphaenogaster swammerdami* was one of the most abundant ants in the dry deciduous forest and grassland sites. This species is important for seed dispersal of *Commiphora guillaumini,* a dominant tree in western dry forests of Madagascar (Böhning-Gaese *et al.*, 1999). This process may be of considerable importance in open areas, where ants are seed dispersers, for example in Australian *Rhytidoponera* (Gove *et al.*, 2007). The impact of fire on the seed dispersal process in Madagascar has yet to be investigated. The Beanka Reserve provides an ideal system to study this aspect.

Introduced species

Eight introduced ant species were recorded in grasslands and 10 species in dry deciduous forest (Tables 9-1 & 9-2). Five species, *Monomorium descructor*, *Paratrechina longicornis, Pheidole megacephala*, *Technomyrmex alpibes*, and *Tetramorium delagoense*, were found in both habitats. Grassland habitats, with their cycle of disturbance, may provide a gateway for invasive ants to enter native dry forests (Figure 9-5). Of particular concern is *P. megacephala*, which is considered a threat to native ants (Hoffmann & Parr, 2008) and is listed as one of the hundred "world's worst" invaders (Global Invasive Species Database, 2013). However, other species, such as *Technomyrmex pallipes,* should also be monitored. Two studies on a related introduced species, *T. albipes,* in eastern Madagascar, specified the need to establish a monitoring program for this taxon (Ratsirarison *et al.*, 2005; Dejean *et al.*, 2010). The Beanka Forest provides an interesting setting to study the ecology and dynamics of invasive species.

Management of grasslands

Our survey shows that after more than 50 years of deforestation and habitat fragmentation, ant diversity in grasslands in western Madagascar is a fraction of that found in adjacent forest. The presence of only introduced or wide spread generalist species in grasslands suggests that this habitat is not a natural part of the lowland Beanka ecosystem. This is in contrast with the Central Highlands, where ant diversity appears similar in both grassland and adjacent forest habitats and microendemics were found. The primary threat to dry deciduous forests in western Madagascar appears to be human-set fire, which is the source of more than 94% of all fires in western Madagascar (Bloesch, 1999). Though fire is intended to renew pasturelands, its passage reduces ant diversity, which in turn is presumed to have some adverse effects associated with ecosystem functioning. For example, ants play a key role as soil engineers (Dauber *et al.*, 2008) and have positive interactions with some plant roots (Gove *et al.*, 2007).

Conclusion

Our survey determined the best methods for sampling ants in two types of Beanka grasslands (burned and unburned) and found that higher ant diversity occurred in the unburned habitat. Species found in both grassland types were considered adapted to regular fire regimes, and those found uniquely in burned or unburned grasslands should be considered as indicator taxa to characterize and monitor the state of grasslands in western Madagascar. Overall, our results provide preliminary data useful for establishing monitoring programs and fire management policies. Grasslands at additional localities across Madagascar should be inventoried to test the wider application of these indicator taxa.

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