

The Curious Case of the Camelthorn: Competition, Coexistence, and Nest-Site Limitation in a Multispecies Mutualism

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ABSTRACT: Myrmecophyte plants house ants within domatia in exchange for protection against herbivores. Ant-myrmecophyte mutualisms exhibit two general patterns due to competition between ants for plant occupancy: (i) domatia nest sites are a limiting resource and (ii) each individual plant hosts one ant species at a time. However, individual camelthorn trees (*Vachellia erioloba*) typically host two to four ant species simultaneously, often coexisting in adjacent domatia on the same branch. Such fine-grain spatial coexistence brings into question the conventional wisdom on ant-myrmecophyte mutualisms. Camelthorn ants appear not to be nest-site limited, despite low abundance of suitable domatia, and have random distributions of nest sites within and across trees. These patterns suggest a lack of competition between ants for domatia and contrast strongly with other ant-myrmecophyte systems. Comparison of this unusual case with others suggests that spatial scale is crucial to coexistence or competitive exclusion involving multiple ant species. Furthermore, coexistence may be facilitated when co-occurring ant species diverge strongly on at least one niche axis. Our conclusions provide recommendations for future ant-myrmecophyte research, particularly in utilizing multispecies systems to further our understanding of mutualism biology.

Keywords: ant-plant interactions, coexistence, domatia, myrmecophytes, mutualism, nest-site limitation.

Introduction

Ant-plant interactions are used as model systems to test hypotheses about ecological networks (Lange and Del-Claro 2014), mutualism dynamics (Heil and McKey 2003), and the effects of global change on interacting species (Mayer et al. 2014). Interactions vary from facultative to obligate

and range from mutualism to parasitism. Plants benefit from ants via services including seed dispersal (Gallegos et al. 2014), pollination (de Vega et al. 2009), nutrient enrichment (Wagner and Fleur Nicklen 2010), and protection against herbivory (Trager et al. 2010). In exchange, myrmecophilic plants provide food to ants, a process that is taxonomically widespread (Weber and Keeler 2013). Food may take the form of extrafloral nectar, honeydew via sap-feeding insects, and food bodies rich in lipids and proteins (Heil and McKey 2003). In addition, myrmecophytic plants also provide housing for ants within domatia. These are modified plant structures such as thorns, stems, and leaves that form hollow chambers specifically for ant nest sites (Davidson and McKey 1993). Myrmecophytes are restricted to the tropics (Heil and McKey 2003) with well-studied taxa including *Cecropia* (Dejean et al. 2012), *Macaranga* (Nomura et al. 2011), *Tococa* (Michelangeli 2003), and, perhaps most famously, *Vachellia* (formerly *Acacia*; Janzen 1974; Palmer et al. 2008).

Most studies support the common view that there is intense inter- and intraspecific competition between ant colonies for sole occupancy of myrmecophytes (Davidson et al. 1989; Kautz et al. 2012; Palmer 2004; Webber et al. 2007) and that plant-ants are extremely aggressive and territorial (e.g., Palmer et al. 2000). Ant colonies may compete not only for food resources provided by the plant but also for domatia in which to nest (Fonseca 1999). We define three occupancy categories for ants on a given myrmecophyte plant at a given time: (i) single-colony occupancy (SCO), (ii) single-species (multiple-colony) occupancy (SSO), and (iii) multiple-species co-occupancy (MSC). An individual plant usually shows SSO (which may be SCO, but this is often not tested), although the plant species may associate with more than one ant species (Davidson et al. 1989; Gaume and McKey 1999; Palmer et al. 2003). An individual plant may show MSC as a sapling

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(Longino 1989; Djéto-Lordon et al. 2005) or sequentially throughout its lifetime, because ant colonization is moderated by species dominance hierarchies (Palmer et al. 2000). Outside of myrmecophyte systems—for example, in cavity-nesting ants—nest-site limitation influences ant species richness, and therefore possibly coexistence, although the effect is variable across arboreal (Philpott and Foster 2005) and leaf-litter assemblages (Byrne 1994). In myrmecophytes, ant colony size is limited by the size and availability of domatia for nesting sites (Fonseca 1993, 1999; Campbell et al. 2013a), but the effect on species coexistence is unknown.

Competition is a key element in structuring ant communities (see review by Cerdá et al. 2013). Interspecific competition leads to nonrandom structure in communities, because species with similar niches co-occur less often than expected by chance (Diamond 1975), a pattern observed across multiple taxa (Gotelli and McCabe 2002). In arboreal ant assemblages, dominant species defend “absolute territories”—discrete spatial units extending beyond the location of individual food or nest resources. This leads to a patchy distribution of ant species across the forest canopy, known as an ant mosaic (Room 1971, 1975; Jackson 1984; Dejean et al. 2007). Outside forest ecosystems, the arboreal patterns of ant species coexistence and competition are not well documented. In savannas, where trees are widely spaced, there is no continuous canopy layer in which ant mosaics can form (Dejean et al. 2007). The dominant trees of African savannas are acacias, many of which are myrmecophytes that possess swollen-thorn domatia inhabited by ants (Dharani 2006). Coexistence of ant species on neighboring myrmecophyte acacias is thought to result from dominance hierarchies and competitive trade-offs (Palmer et al. 2000, 2010; Stanton et al. 2005). However, coexistence at a finer spatial scale, across or within branches of the same tree, has seldom been studied. This is largely because the few intensively studied systems (e.g., Gaume and McKey 1999; Palmer et al. 2010) do not show long-term MSC. MSC on myrmecophytes or semi-myrmecophytes has been documented occasionally (Rico-Gray and Thien 1989a, 1989b; Moog et al. 2002; Raine et al. 2004; Gaume et al. 2005b), but these unusual examples contrast with the many myrmecophytes that exhibit competitive exclusion and SSO.

In this study, we investigated the camelthorn tree, *Vachellia erioloba* (fig. 1A), a southern African myrmecophyte with swollen-thorn domatia (fig. 1B, 1C) inhabited by four ant species: an unidentified *Crematogaster* species, *Catantopius intrudens*, an unidentified *Tapinoma* species, and *Tetraponera ambigua*. A previous study revealed 41% MSC of *V. erioloba* trees, with some evidence of species sorting through nest-site selection on the basis of domatia characteristics (Campbell et al. 2013a). Given this surprising finding, we undertook comprehensive sampling specifically

to quantify MSC in *V. erioloba* and to test for evidence of nest-site limitation and competition. We also test whether nest-site selection differs between ant species on the basis of microhabitat characteristics related to the location of domatia on the tree.

Our findings bring into question two widely held assumptions: (i) myrmecophyte ants utilizing domatia are nest-site limited and (ii) ant species compete for sole occupancy of individual plants. To establish whether the unusual patterns in the camelthorn system are unique, we then review published patterns of plant and domatia occupancy to test for nest-site limitation and MSC in other myrmecophyte systems.

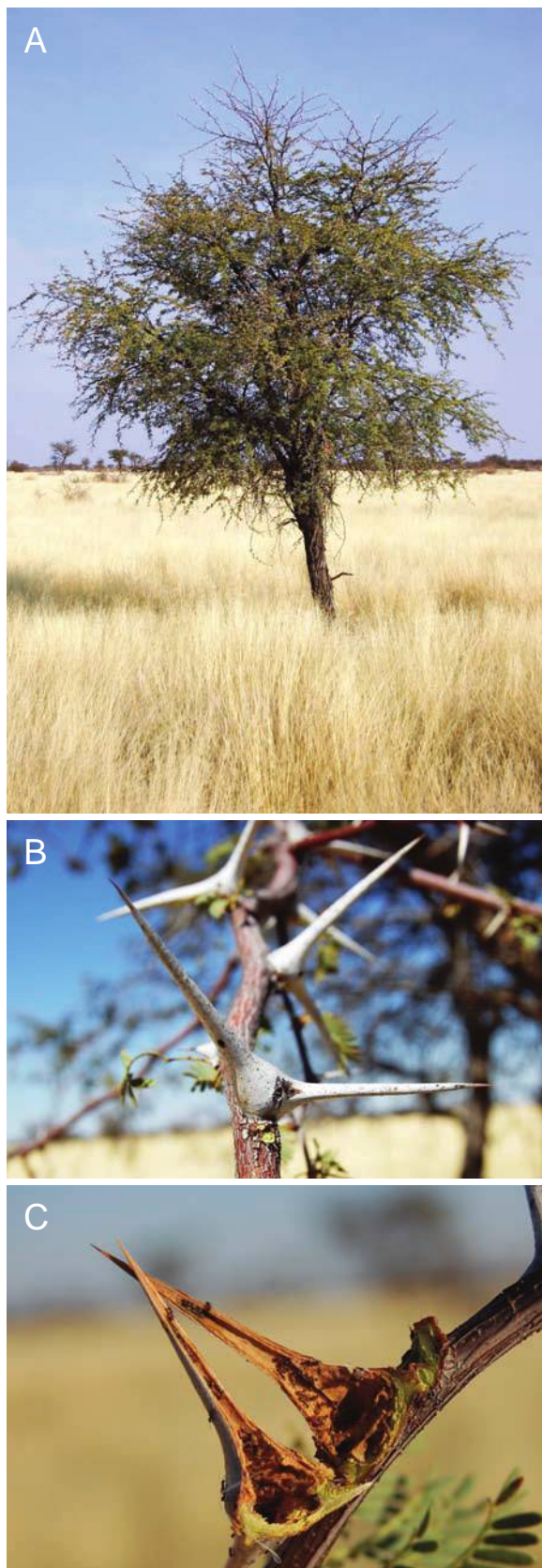
Methods

Study Site and Sampling

Fieldwork was conducted in savanna at Kuzikus Wildlife Reserve (23°13'S, 18°24'E; elevation, 1,340 m) in Namibia (for full site description, see Campbell et al. 2013b). Surveys were completed in April, September, and October 2011 for ant occupants within swollen thorns on camelthorn trees, *Vachellia erioloba* (E. Mey) P.J.H. Hurter. Ants defend *V. erioloba* trees from insect herbivores (Campbell et al. 2013b), and low-intensity sampling in an earlier study suggested MSC of individual trees (Campbell et al. 2013a). For each of 20 haphazardly selected trees, we surveyed 8–13 branches, except for one tree with only four accessible branches. Using secateurs, we removed branches and opened all thorns to examine the contents. We also recorded microhabitat data on branch height from ground (to the nearest 0.1 m), branch cardinal direction (bearing in degrees), thorn status (swollen or nonswollen), thorn age (new, young, old, or dead), and thorn position on branch (surveying from the tip inward toward the trunk, with thorn 1 being the most distal thorn sampled). For each thorn, we recorded ant species identity and an estimate of the number of ants at different life stages (workers, brood, alates, and queens).

Analysis

To test for associations between ant species at the branch level, we performed Spearman's rank correlations on the presence of species on the same branch. To assess the role of competition in structuring within-tree patterns of ant species co-occurrence, we assembled a presence-absence matrix for each of the 20 trees and then performed Monte Carlo randomization tests on each matrix. We used three co-occurrence indexes (CHECKER, COMBO, and C-score) under two null models (fixed-fixed and fixed equiprobable; for details, see appendix, available online). We calculated the standardized effect size (SES) to allow comparison between



matrices (Gotelli and McCabe 2002). To test the null hypothesis that the mean SES measured did not differ from zero, we used a one-sample Wilcoxon test to compare across matrices. We performed six tests (three indexes \times two null models) and therefore applied a Bonferroni correction.

To assess whether the distribution of nest sites across a tree was influenced by microhabitat, we tested for correlations of ant species with thorn and branch variables. In tests of branch variables, we analyzed only species presence or absence on a branch. We tested ant species identity against branch height and cardinal direction using ANOVA and made multiple post hoc comparisons using Tukey honest significant differences tests (Crawley 2005). Thorn variables and ant species identity were tested only for occupied thorns (i.e., domatia that were currently in use as nest sites). Although ants may defend empty neighboring thorns before colony expansion, it was impossible to assign species identity to an unoccupied thorn, so this is not accounted for in our analyses. Due to differential branch growth across trees, thorn position along a branch does not directly predict thorn age, so, for all occupied thorns, we tested whether species identity was correlated with thorn position or thorn age separately using Fisher's exact tests (Crawley 2005). Thorn position was converted to a categorical variable, assigned as follows: distal (thorns 1–8), medial (thorns 9–15), or proximal (thorns 16 and higher).

We assessed species co-occurrence with respect to microhabitat characteristics of branch height and cardinal direction (following the procedure of Belinchón et al. 2012). We constructed 16 presence-absence matrices representing branch cardinal direction (north: 316° – 45° ; east: 46° – 135° ; south: 136° – 225° ; and west: 226° – 315°) combined with one of four branch-height categories (low: 0–1.50 m; medium low: 1.51–2.0 m; medium high: 2.01–2.5 m; and high: ≥ 2.51 m). These analyses followed the same procedure described for within-tree matrices (see appendix). To test the effects of branch height and direction on species interactions, we performed ANOVA on all co-occurrence indexes generated from these matrices.

Co-occurrence randomization analyses (for additional details, see the appendix) were conducted in EcoSim (Gotelli and Entsminger 2009). All other statistical analyses were performed in R (R Core Development Team 2012).

Literature Review

To establish whether the unusual patterns in the camelthorn system are unique, we reviewed published studies

Figure 1: A, *Vachellia erioloba*, camelthorn tree in acacia-dominated savanna at Kuzikus Wildlife Reserve, Namibia. B, Swollen-thorn domatia of *V. erioloba* with ant nest entrance hole visible on left thorn. C, Open swollen-thorn domatia containing a *Tapinoma* nest.

of plant and domatia occupancy to test for nest-site limitation and MSC in other systems. We examined 34 primary studies relating to 49 plant species (table A6; tables A1–A6 available online) for examples of MSC. We included studies featured in three meta-analyses of ant-plant mutualisms (Chamberlain and Holland 2009; Rosumek et al. 2009; Trager et al. 2010) as a representative subset of studies likely to have suitable data. We also added data from several studies published after these meta-analyses and for cases in which inadequate data were provided in the original cited references.

Results

Multiple-Species Co-occupancy (MSC) on Camelthorn Trees

We collected data on 3,448 thorns from 197 branches across 20 *Vachellia erioloba* trees; the full data set is deposited in the Dryad Digital Repository (<http://dx.doi.org/10.5061/dryad.s9f7c>; Campbell et al. 2015). Our data confirmed that *V. erioloba* domatia were occupied by four ant species: an

unidentified *Crematogaster* species, *Cataulacus intrudens*, an unidentified *Tapinoma* species, and *Tetraponera ambigua* (hereafter referred to by genus). The smallest, *Tapinoma*, was the most abundant, accounting for 77% of all individual ants surveyed (fig. 2), followed by *Crematogaster* (13%), *Cataulacus* (8%), and then *Tetraponera* (2%). *Tapinoma* was also the most prevalent, found on 19 of 20 trees. Despite the numerical dominance of *Tapinoma*, it occupied a similar proportion (41%) of nest sites to *Crematogaster* (42%; see fig. 2), whereas the other two species occupied far fewer (*Cataulacus*, 14%; *Tetraponera*, 3%).

Most trees (16 of 20) were co-occupied by two or three ant species, and 3 of 20 trees were occupied by all four. The only tree with a single ant species was occupied by *Crematogaster*. Although MSC was common within trees, the figure decreased to 27% at the branch level, whereas 50% of branches were occupied by a single species (fig. A1, available online). On branches, *Cataulacus* and *Tapinoma* were positively associated ($r_s = 0.17$, $S = 1,051,634$, $P < .05$, by Spearman correlation test), but there were no other significant associations between species pairs (table A5).

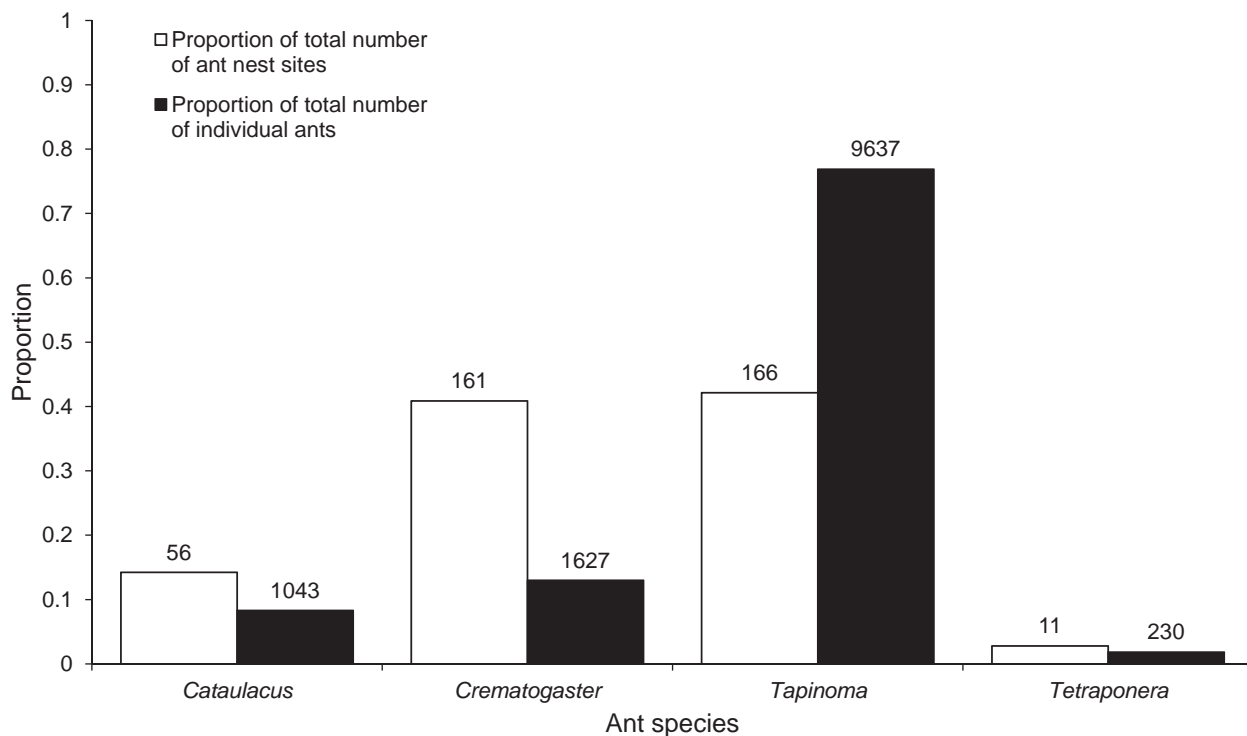


Figure 2: Occupation of nest sites and numerical dominance of four ant species on *Vachellia erioloba* at Kuzikus Wildlife Reserve. Open bars represent each species as a proportion of the total number of ant nests found on trees. Filled bars represent each species as a proportion of the total number of individuals (including workers, alates, queens, and brood) recorded during surveying. Sample size is indicated by numbers above each bar. Data underlying figure 2 are deposited in the Dryad Digital Repository, <http://dx.doi.org/10.5061/dryad.s9f7c> (Campbell et al. 2015).

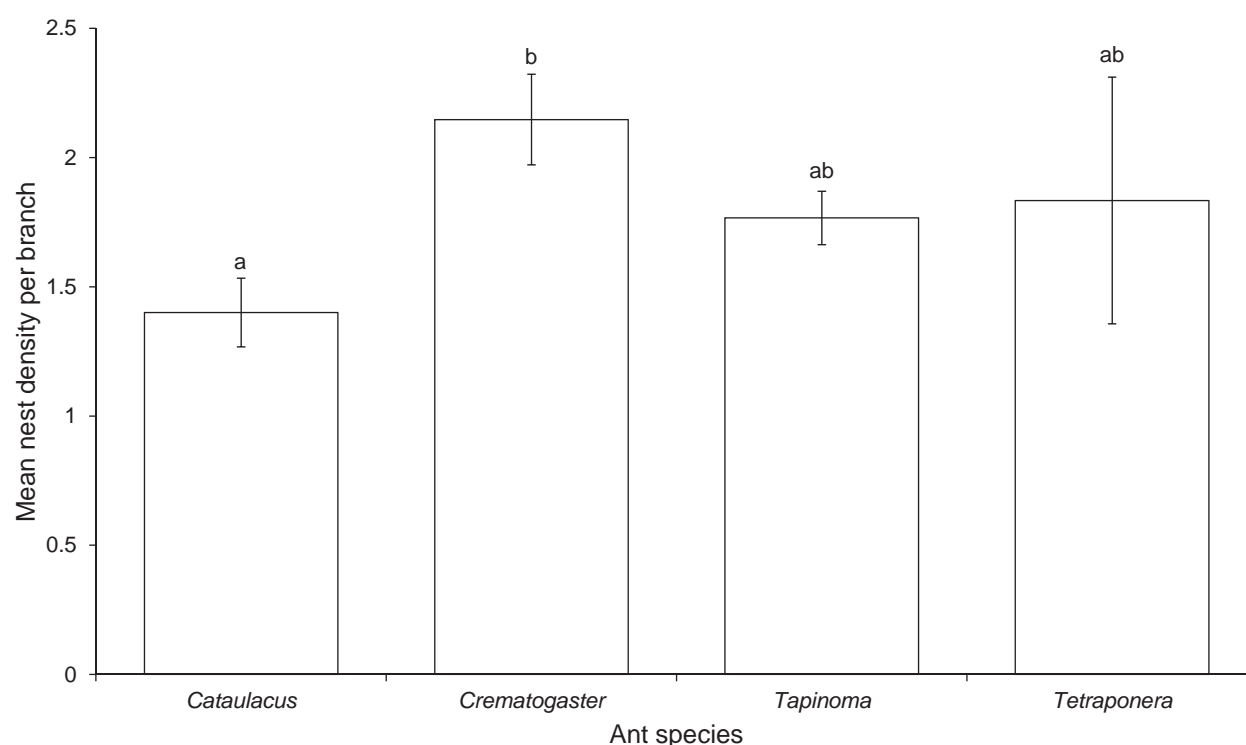


Figure 3: Mean (\pm SE) number of nest sites per branch for the four ant species on *Vachellia erioloba* at Kuzikus Wildlife Reserve. Analysis of variance showed that nest-site density varies significantly ($F_{3,211} = 3.603$, $P < .05$) between ant species (*Cataulacus intrudens*, $n = 40$; *Crematogaster* species, $n = 75$; *Tapinoma* species, $n = 94$; and *Tetraponera ambigua*, $n = 6$). Different letters indicate significant differences ($P < .001$) using Tukey honest significant difference test for post hoc comparisons.

Nest-Site Limitation and Interspecific Competition

Only 31% ($n = 1,052$) of thorns were suitable as nest sites, because ants were not found inside soft, new-growth thorns or nonswollen thorns. Hereafter the term “domatia” refers only to swollen and hardened thorns suitable as nest sites. Only 37% of domatia were occupied by nesting ants. Nest-site density varied significantly between species, with *Crematogaster* occupying the most domatia per branch and *Cataulacus* the least (fig. 3).

Within trees, we found little evidence of positive or negative associations between ant species. Only one tree showed a nonrandom distribution of species with an observed C -score of 17.33 that was significantly larger ($P = .001$) than the mean simulated C -score of 7.95. The SES was 3.104, indicating segregation of ant species on that tree (table A1). Our meta-analysis across matrices for all trees showed that the mean SES did not differ significantly from zero for any of the indexes under either null model (table A3).

Microhabitat Preferences

We found no evidence that different ant species used domatia in different locations on trees with regard to branch

height ($F_{3,211} = 1.16$, $P = .33$, by ANOVA), branch cardinal direction ($F_{3,211} = 2.048$, $P = 0.11$, by ANOVA), or thorn position along a branch (Fisher’s exact test, $P = .14$). Nor did we detect evidence of nonrandom patterns for nest sites grouped by microhabitat characteristics (table A2). This was confirmed by meta-analyses of co-occurrence indexes that showed species distributions are not influenced by branch height or direction (table A4). However, *Cataulacus* and *Tapinoma* were associated with old thorns, and *Crematogaster* and *Tetraponera* were associated with young thorns (Fisher’s exact test, $P < .01$).

Discussion

Ant community ecology has focused heavily on the role of competition in structuring ant assemblages (Parr et al. 2005; Parr and Gibb 2010; Cerdá et al. 2013), but this has been less frequently applied to ants on myrmecophilic or myrmecophytic plants (however, see the review by Palmer et al. [2003]). Given the abundance of studies on ant-plant mutualisms, the data are most likely already available for studies of competition within these systems and would significantly advance our understanding of the dynamics of

ant-ant interactions as well as mutualism biology. Although it is thought that ants compete for host plants (Stanton et al. 2005) or that domatia nest sites are limiting (Longino 1989), there has been little experimental proof to find evidence of competition in these systems or to identify the mechanisms for this (Palmer et al. 2003).

There is no evidence that ants on *Vachellia erioloba* are nest-site limited, because only 38% of domatia are occupied. This rate of domatia occupancy does not seem atypical of ant-plant systems in general (see table A6). In the studies that we reviewed, domatia occupancy ranged from 42% to 64% (Maschwitz et al. 1994; Moog et al. 2002), although variation between individual plants was high (0%–100%) for one species (Dyer and Letourneau 1999). *Zanthoxylum myriacanthum* was the only MSC myrmecophyte with rate of domatia occupancy reported (tables 1, A6), which at 42% is very similar to domatia occupancy on camelthorn trees. However, it is striking that domatia occupancy was not reported in 93% of primary studies, even though this information was very likely collected. Overall, the few existing studies do not support the notion that individual domatia are a limiting resource, but this does not rule out the availability of whole plants as limiting for ant colonies.

At the whole-plant level, overall occupancy of myrmecophytes was between 41% and 100% in the reviewed studies (table A6) and 100% on *V. erioloba*. If we focus on MSC myrmecophytes, the rate of plant occupancy was slightly higher, ranging from 62% to 100%. The high rate of plant occupancy relative to domatia occupancy implies that the availability of individual plants may often be the key to ant colony success. This also explains why competition between foundresses and young colonies for possession of a host plant is so intense (Stanton et al. 2005), whereas once a colony has secured a plant, individual domatia availability may exceed colony requirements.

The apparently random distribution of species within trees provides further evidence that *V. erioloba* ants are not competing for domatia. This pattern is surprising and contrasts with spatial patterns on a Neotropical acacia exhibiting MSC (Raine et al. 2004). We propose that, on *V. erioloba*, fine-scale within-tree coexistence and lack of strong spatial structure reflect an absence of competition for individual domatia. Moreover, each ant species tends to choose different subsets of nests (e.g., species sorting according to thorn age and size; see Campbell et al. 2013a) and exhibits different patterns of nest-site distribution and density. Species coexistence on *V. erioloba* may also be facilitated by the differing life-history strategies of each ant species (Campbell et al. 2013a), as has been demonstrated in acacia-*Pseudomyrmex* interactions (Kautz et al. 2012). Establishing colony boundaries between conspecific ants co-occupying a tree is the next step in understanding spatial patterns of domatia-inhabiting

ants on *V. erioloba*. However, in a pilot study to test colony boundaries, we did not observe any intra- or interspecific aggression between *V. erioloba* ants. Future work could compensate for the lack of behavioral assay data by utilizing genetic microsatellite and cuticular hydrocarbons data instead (Kautz et al. 2012).

At the species level, most plants (30 of 49 species; table A6) associate with multiple ant species, and this is very likely an underestimate. Most myrmecophytes associate with a guild of ants, via either (i) an individual plant associating with multiple ant species over its lifetime or (ii) different individual plants in a population associating with different ant species. Ant partners may also vary across geographic scales (Longino 1989), and some myrmecophytes lose their mutualist ants altogether (Moraes and Vasconcelos 2009). This highlights the important issues of scale and specificity in studying species interactions (Thompson 2005). A 1 : 1 level of partner species matching may often be recorded on individual plants, but additional ant partners may be added by looking across plants in a population or across populations through the plants' geographic range.

It was typically difficult to establish species occupancy states (unknown for 10 of 49 species; table A6) and, in particular, MSC from the literature, because authors were not always explicit about this. Ultimately, we found only four examples of MSC (table 1; Rico-Gray and Thien 1989a; Moog et al. 2002; Gaume et al. 2005a), and only one of these described the pattern of ant species coexistence within individual host plants (Raine et al. 2004). These four cases appear extremely divergent and occur on different continents, in contrasting habitats, and incorporate a diversity of plant types and domatia structures (table 1). In our study, 95% of individual camelthorn trees showed MSC. Ant species were consistent with a previous study (Campbell et al. 2013a), suggesting temporal stability of species assemblages, at least over short periods of time. This level of simultaneous coexistence is highly unusual and, to our knowledge, has not previously been recorded for an African ant-acacia. It contrasts directly with other African acacias, notably the intensely studied *Vachellia drepanolobium* system, in which trees are usually occupied by not only a single species (SSO) but also a single colony of ants (SCO; Palmer et al. 2000; Stanton et al. 2005). This raises the following questions: Do mutualism dynamics differ when a plant has one or more ant partners? And what drives transitions between SCO, SSO, and MSC states?

In systems where guilds of ants inhabit different individual host plants within a population, it is frequently discovered that not all ants are mutualists (Gaume and McKey 1999; Itioka et al. 2000; Edwards et al. 2010) and that mutualists differ in their effectiveness (Young et al. 1997; Frederickson 2005), although increased competition can actually encourage cooperation of multiple mutualist partners (Adam 2010). Nonetheless, it is critical to understand

Table 1: Summary of myrmecophytic plants that exhibit multiple-species co-occupancy (MSC) by their resident ant species

Plant species	Plant and domatia type	Habitat	Country	Common domatia-inhabiting ant species	No. ant species	Plant/domatia occupancy (%)	MSC occupancy (%)
<i>Humboldtia brunonis</i> (Fabaceae, Caesalpinioideae)	Tree with hollow internodes	Wet evergreen forest	India	Unknown	7	100/...	...
<i>Myrmecophila christinae</i> (Orchidaceae, Epidendroideae)	Epiphyte with hollow pseudobulbs	Coastal sand dune mattoral	Mexico	<i>Crematogaster brevispinosa</i> , <i>Camponotus planatus</i> , <i>Camponotus abdominalis</i> , <i>Camponotus rectangularis</i> and <i>Ectatomma tuberculatum</i>	13	.../...	...
		Flooded mangrove forest	Mexico	<i>Dolichoderus bispinosus</i> and <i>Pachycondyla villosa</i>	18	77.2/...	...
		Low inundated forest	Mexico	<i>C. planatus</i> , <i>Cyphomyrmex minutus</i> and <i>Monomorium ebeninum</i>	20	85/...	...
<i>Vachellia erioloba</i> (Fabaceae, Mimosoideae)	Tree with swollen thorns	Savanna	Namibia	<i>Crematogaster</i> sp., <i>Cataulacus intrudens</i> , <i>Tapinoma</i> sp., and <i>Tetraponera ambigua</i>	4	100/37.45	95
<i>Vachellia mayana</i> (Fabaceae, Mimosoideae)	Shrub/small tree with swollen thorns	Lowland wet forest	Mexico	<i>Pseudomyrmex ferrugineus</i> ; <i>C. planatus</i>	2	100/...	30.7
<i>Zanthoxylum myriacanthum</i> (Rutaceae, Toddalioideae)	Tree with hollow stems	Lowland forest	Malaysia	Many species, including <i>Cataulacus</i> , <i>Crematogaster</i> , <i>Tapinoma</i> , <i>Technomyrmex</i> , <i>Camponotus</i> , and <i>Tetraponera</i>	28	62 ^a /42	83

Note: For the full list of species included in our review of myrmecophyte occupancy states, see table A6, available online. Occupancy rates for plants and domatia are given as percentages. MSC occupancy is the percentage of plants that are occupied by colonies of multiple different ant species nesting within domatia.

^a On *Zanthoxylum myriacanthum*, 62% of all trees sampled (including those with only closed-stem domatia) were occupied by ants; however, 100% of all trees sampled with open hollow-stem domatia were occupied by ants.

the nature of the relationship between ants and their host plant, because parasites may be more likely to co-occupy hosts (Kautz et al. 2012). A great deal of literature is directed toward analyzing the costs and benefits of interactions and how systems may allow for the existence of “cheaters” (e.g., Edwards et al. 2006; Clement et al. 2008; Kautz et al. 2012). The key difference in myrmecophytes exhibiting MSC is the scale at which coexistence occurs (i.e., tree or branch level rather than population level). However, both empirical and theoretical frameworks designed to tackle population-level questions of ant-species coexistence (as reviewed by Palmer et al. 2003) are equally applicable to this finer spatial scale. The main difficulty in performing experimental work in MSC cases is logistical. Manipulations on mature plants would be complex, and the best approach would be to perform exclusions and additions of different combinations of ant species using seedlings until plants reach maturity. This would be problematic on a large, slow-growing tree, such as *V. erioloba*, but better suited to a pioneer species, such as the MSC myrmecophyte, *Z. myriacanthum* (Moog et al. 2002). This would also allow for experimentation over a longer timescale and facilitate cost-benefit analysis over the lifetime of a plant or ant colony that may potentially reveal insights that differ from those of a short-term study (e.g., a short-term study such as Palmer et al. 2000 vs. a long-term study such as Stanton and Palmer 2011).

Multispecies mutualisms may arise as evolutionary by-products (Fayle et al. 2011) when ants defend trees as part of their normal foraging activity. Multiple simultaneous ant partners may provide more benefits (greater defense) or wider benefits (defense against diverse foes) to an individual host plant. In this context, tree size or density may influence the costs and benefits of MSC for myrmecophytes. Large, isolated desert or savanna trees might require a suite of ant occupants to be effectively defended. Smaller plants or those found in dense forests with many potential opportunistic ant mutualists may fare well with one resident ant colony. MSC might alternatively arise from a lack of host sanctions, resulting in the presence of multiple opportunistic species. For example, an inability to limit access to domatia leads to the presence of parasites on the rattan ant-palm *Korthalsia furtadoana* (Edwards et al. 2010).

Environmental stress can determine levels of species diversity in ant assemblages. Habitat productivity may also play a role in diversity within mutualisms via species coexistence mechanisms; for example, competitively dominant ants occupy faster-growing host plants and more productive habitats (Palmer 2003). Subsequently, should we expect MSC systems to be found in more or less stressful environments? Multiple limiting resources can increase niche dimensionality, leading to higher levels of diversity. Conversely, a decrease in the number and heterogeneity of limiting resources causes fewer trade-off opportunities, decreased niche dimensionality, and

fewer coexisting species (Harpole and Tilman 2007). If niche dimensionality does influence myrmecophyte-inhabiting ants, we predict that more stressful environments with multiple limiting resources will contain a greater number of MSC plants and a higher diversity of ant partners. Although data are limited, the anecdotal evidence somewhat supports this prediction; *V. erioloba* is a savanna and desert species in a high temperature and aridity region, and mangrove orchids with MSC are found in very high-salinity environments (Rico-Gray and Thien 1989b). To test stress-diversity relationships in ant-myrmecophyte systems, MSC mutualisms could be compared across environmental (stress/productivity) gradients or subjected to local resource availability manipulations. Not only could a factorial experiment of this kind tease out the relative importance of different resources on species, but also changes observed in number of ant occupants following restriction or addition of resources would indicate a role for niche dimensionality in ant-myrmecophyte interactions.

Two widely held assumptions about ant-myrmecophyte mutualisms are challenged by our findings on *V. erioloba*: (1) ant-plants are inhabited by a single species at a time, and therefore, ant coexistence occurs only across a population of plants or a single plant’s lifetime; and (2) plant-ants are nest-site limited on their host myrmecophyte (Fonseca 1993; Yu et al. 2004). We found other examples of MSC in the literature (Rico-Gray and Thien 1989b; Moog et al. 2002; Raine et al. 2004), but domatia occupancy rates are sorely missing from most published studies. Despite this, the few studies reporting patterns suggest no saturation of domatia (Maschwitz et al. 1994; Dyer and Letourneau 1999; Moog et al. 2002). Competition-colonization trade-offs (Stanton et al. 2002) and dispersal-fecundity trade-offs (Yu et al. 2004) have both been highlighted as likely mechanisms for species coexistence on myrmecophytes, but most research has focused on a few popular study species. The approaches taken with these species could easily be expanded to MSC myrmecophytes to aid our understanding of competition and coexistence in ant/plant systems. The study of multispecies systems has been highlighted as vital to progress our understanding of mutualisms (Palmer et al. 2003; Fayle et al. 2011). We hope that the opportunity presented to study ant communities on *V. erioloba*, as well as the other MSC systems highlighted here, provides a starting point for a better understanding of multispecies mutualism dynamics.

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