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## **RESEARCH ARTICLE - ANTS**

# Ant Fauna on *Cecropia pachystachya* Trécul (Urticaceae) Trees in an Atlantic Forest Area, Southeastern Brazil

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# Abstract

*Cecropia* are pioneer successional trees frequently associated with ants. Generally a single dominant colony of *Azteca* ant inhabits each mature *Cecropia* tree, but other ant species may be found living or foraging on the same tree. In this study, we assessed the diversity of ant species on *Cecropia pachystachya* trees in two sites in the Brazilian Atlantic Forest: a dust-free roadside and a dusty roadside. We also investigated the influence of tree architecture on ant species richness. We found a total of 24 ant species distributed in 11 genera and five subfamilies on *C. pachystachya* trees; 18 in the dust-free roadside and 14 in the dusty roadside. We found up to five ant species on a single tree, but only *Azteca alfari* was frequently encountered. Ant species richness per tree did not differ significantly between sites and was related to tree architectural traits. On the other hand, ant species composition on trees differed significantly between sites. Our study indicates that heavy dust deposition on *Cecropia* trees may affect associated ant communities, not by changing ant species richness, but by causing different species to live and forage on trees under different dust exposure.

#### Introduction

Cecropia genus comprises pioneer trees which are characteristic elements of forest borders and gaps in the Neotropics (Vasconcelos & Casimiro, 1997; Sposito & Santos, 2001). Most species are inhabited by mutualistic ants that nest inside hollow internodes where they store eggs, larvae and pupae (Harada & Benson, 1988), and feed on glycogenrich Müllerian bodies called trichilium located at the base of leaf petioles (Yu & Davidson, 1997). Although Cecropia can host a variety of resident ant genera, Azteca spp are the most common ant inhabitants (Longino, 1991a). Usually only one mature Azteca colony inhabits a Cecropia tree, however up to five Azteca species may be found in a Cecropia population (Longino, 1991a). In addition to a resident Azteca colony, other ant species can be found living or foraging on the same Cecropia tree (Vieira et al., 2010). However, the diversity of non-Azteca ants living on Cecropia trees or using them as foraging substrates is still poorly known, as well as the factors that might affect ant richness and abundance on these trees.

Complex tree architecture is an indicative of resource diversity and availability, such as nesting sites, sites for oviposition, and food (Lawton, 1983). Therefore, tree architecture might be an important factor affecting the diversity of ant fauna associated to trees as reported for other insects (Haysom & Coulson, 1998; Espírito-Santo et al., 2007; Neves et al., 2013). *Cecropia* trees are known for their great variation in size and architectural traits (Sposito & Santos, 2001), but the way this variation affects the associated ants remains unexplored.

Distribution of ants on *Cecropia* trees is also likely influenced by external factors such as changes of environmental conditions caused by human activities, which can directly or indirectly affect both plants and ants. The Brazilian Atlantic Forest range largely coincides with the most populated areas of Brazil (Tabareli et al., 2005), thus this forest ecosystem suffers with impacts of a dense road network. Roads are known to have many ecological effects on surrounding biota (Spellerberg, 1998), mainly by altering landscape spatial patterns and processes and modifying abiotic and biotic conditions (Forman &



Alexander, 1998; Trombulak & Frissell, 2000; Barbosa et al., 2010). Besides, production of dust either by cars exhausts or road surfaces may have important potential impacts on vegetation in roadsides including plant mortality (Spellerberg, 1998), especially on unpaved roads where dust production is higher than on paved roads (Farmer, 1993). Dust deposition on leaves may increase water loss, cause leaf injuries, also leading to changes in associated invertebrate communities (Farmer, 1993). Nevertheless, the effects of roadside dust on ant fauna associated to *Cecropia* trees have never been studied so far.

In this context, our aim was to assess the diversity of ant fauna on *Cecropia pachystachya* Trécul (Urticaceae) trees in a Brazilian Atlantic Forest area in Southeastern Brazil. We evaluated ant species richness and composition on trees located in two roadside forest sites: one where plants were heavily covered by road dust and another where dust on vegetation was negligible. We also evaluated the relationship between ant species richness and the architectural traits of *C. pachystachya* trees. We hypothesized that ant species richness on trees should be positively related to higher complexity in plant architecture and should be lower in plants disturbed by heavy dust deposition. We also hypothesized that contrasting dust deposition should affect the composition of ant fauna associated to *C. pachystachya* trees.

### **Material and Methods**

#### Study area

This study was conducted in the Rio Doce State Park (Parque Estadual do Rio Doce-PERD hereafter) (19°40'S; 42°33'W) and its surroundings in Minas Gerais State, Southeastern Brazil (Fig 1). PERD is the largest continuous remnant of Atlantic Forest in the state (*c*. 36,000 ha) (Instituto Estadual de Florestas de Minas Gerais [IEF-MG], 2012) and thus has a major importance to the regional conservation of this biome. It is surrounded by agriculture, pasturelands, large areas of *Eucalyptus* plantations, metallurgical plants, and a substantial road network (Brito et al., 1997). *Cecropia* trees are abundant in early successional stages of forest both inside and outside the park, and at the roadsides of unpaved roads *Cecropia* trees are often heavily covered by dust (Fig 2).



**Fig 1**. Rio Doce State Park in Southeastern Brazil. The circle shows the dust-free sampling area inside the park. Adapted from IBGE/ Brasil topographical map by Philippe Maillard - Instituto de Geociências-IGC, Universidade Federal de Minas Gerais.



Fig 2. Cecropia pachystachya leaves. A - Inside Rio Doce State Park, not covered by road dust. B - Covered by unpaved road dust in the surroundings of the park.

# Sampling

Data were collected in July 2012 in two Atlantic Forest early successional sites: a dust-free roadside nearby Lake Carioca inside PERD and a dusty roadside in the surroundings of the park. Although both sites are roadsides, the one inside PERD is located in a rarely used airplane landing field while the road outside the park connects small municipalities and is frequently used by cars. To sample the associated ant fauna, we selected 25 mature Cecropia pachystachya trees (Circumference at Breast Height (CBH)  $\ge$  20 cm) at least 30 m away from each other along a transect in each site. Ants found on leaves and trunks were manually captured with the assistance of a ladder and a pole pruner, and additional arboreal pitfall traps with sardine as bait were used to catch ants that we did not capture manually (Bestelmeyer et al., 2000; Ribas et al., 2003). Pitfalls were set up at approximately 1.50 m height in C. pachystachya trunks and were removed 48 hours later. In order to avoid that ground-living ants were caught, we used grease in the trunks below pitfall traps. All ants caught were taken to the Myrmecology Laboratory of CEPEC/CEPLAC in Ilhéus, Bahia. In this laboratory, ant species were identified after preparation and deposited in the collection referred by CPDC acronym (#5687). For each C. pachystachya tree we measured the following architectural parameters: CBH, total height, number of branches and leaves, and first branch height (FBH) following Sposito & Santos (2001). We used ground-based methods of access (Von Matter et al., 2010) because of the difficulty of sampling very tall and fragile trees. Total height of trees was measured with a 12 m pole, and branches and leaves were counted by eye.

#### Statistical Analyses

In order to evaluate the effectiveness of our sampling effort, we constructed rarefaction curves for each site using Mao Tao observed richness based on 100 randomizations. A generalized linear model (GLM) was constructed to investigate the effect of area (dusty and dust-free roadsides; explanatory variable) on ant richness (response variable). Difference in ant species composition between sites was tested using Non-metric multidimensional scaling (NMDS) with Raup-Crick index for incidence data and Analysis of similarities (ANOSIM). ANOSIM provides a way to check whether there is a significant difference between groups or not (Clarke, 1993). Similarity percentage (SIMPER) analysis was used to determine which ant species mostly contributed to differentiation between groups (dust-free and dusty sites). In order to determine which architectural variables (CBH, total height, number of branches and leaves, or FBH) affected ant species richness, we used Hierarchical Partitioning. Then, generalized linear models (GLMs) were built to estimate the effect of these pre-selected architectural variables (explanatory variables) on ant species richness (response variable).

Residual analyses were conducted to check data adequacy to the probability distribution used as well as error distribution (Crawley, 2007). Species rarefaction curves were constructed on Estimates 8.0 software (Colwell, 2006); NMDS, ANOSIM and SIMPER were carried out on PAST (Paleontological Statistics) version 2.15 (Hammer et al., 2001); Hierarchical partitioning and GLMs were performed using software R 2.6.2 (R Development Core Team, 2008).

#### **Results and Discussion**

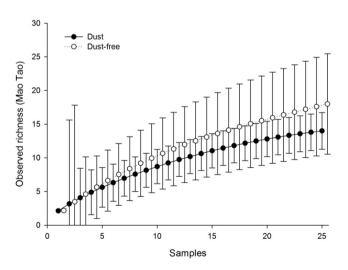
A total of 24 ant species distributed in 11 genera and five subfamilies were found on *Cecropia pachystachya* trees (Table 1). The absence of stabilization of the rarefaction curves (Fig 3) indicated that ant species richness associated to *C. pachystachya* trees is probably underestimated in both study sites. Therefore, additional studies are needed to determine more accurately the richness of ant species associated to these trees.

Formicinae and Myrmicinae were the most representative subfamilies with eight and nine species respectively (71% of all ant species found); and Camponotus Mayr, 1861 was the most representative genus with seven species. Although more than one Azteca species can inhabit a single population of Cecropia trees (Longino, 1991a), we only found Azteca alfari Emery, 1893 on the trees we analyzed. Azteca alfari is known to inhabit Cecropia trees located in open and disturbed habitats such as the early successional sites we studied while other Azteca species such as Azteca constructor and Azteca xanthochroa are more common in older forest stages (Longino, 1991b). The presence of various non-Azteca ant genera such as Camponotus Mayr, 1861, Crematogaster Lund, 1831 and Pseudomyrmex Lund, 1831 (Table 1) on trees of Cecropia was also reported by Longino (1991a) and their presence indicates low aggressiveness of the Azteca resident colony. As we observed on the field, some C. pachystachya trees inhabited by A. alfari also housed other ant genera nests (e.g. Cephalotes species). Indeed, A. alfari along with Azteca ovaticeps Forel, 1904 are the least aggressive species among all Cecropia-inhabiting Azteca ants (Longino, 1991a).

Eighteen ant species were found on *C. pachystachya* trees in the dust-free site while 14 were found on the road dust covered trees. Only eight ant species were common to both sites, six species were found exclusively in the dusty roadside and 10 were exclusive of the dust-free roadside inside the park (Table 1). *Azteca alfari* Emery, 1893 and *Camponotus senex* (Smith, F., 1858) were the most frequent species on *C. pachystachya* trees in the dust-free area - e.g. *Camponotus balzani* Emery, 1894. Unlike the expected, each *C. pachystachya* tree under heavy dust deposition held an average number of ant species similar to each tree in the dust-free roadside:  $2.12\pm0.24$ ; dust-free roadside:  $2.25\pm0.21$ ; Deviance<sub>147</sub> = 0.094; p = 0.687).

Table 1. Ant species captured on *Cecropia pachystachya* trees in Rio Doce State Park and surroundings, southeastern Brazil. Sampled sites were a dusty roadside (in the surroundings of the park) and a dust-free roadside (inside the park).

Subfamily/Species Du		C. pachystachya venant species Dust-free roadside (n=25)
Du	sty roadside	Dust-free roadside
	•	
Dolichoderinae	(11-25)	(11-23)
Azteca alfari Emery, 1893	24	22
Dolichoderus diversus Emery, 1894	1	
Dolichoderus lutosus (Smith, F., 1858)	-	1
Ectatomminae		-
Ectatomma tuberculatum (Olivier, 1791)		3
Formicinae		5
Brachymyrmex sp.01	1	
Camponotus balzani Emery, 1894	2	4
Camponotus cingulatus Mayr, 1862	1	
Camponotus gp. Fastigatus sp.01	2	
Camponotus renggeri Emery, 1894	3	3
Camponotus rufipes (Fabricius, 1775)	2	-
Camponotus senex (Smith, F., 1858)	9	3
Camponotus vittatus Forel, 1904	1	-
Myrmicinae	_	
Atta rubripilosa Forel, 1908		1
Cephalotes goeldii (Forel, 1912)		1
Cephalotes minutus (Fabricius, 1804)		1
Cephalotes pusillus (Klug, 1824)	1	4
Crematogaster brasiliensis Mayr, 1878		1
Crematogaster curvispinosa Mayr, 1862	2	4
Nesomyrmex costatus (Emery, 1896)		1
Solenopsis sp.01	2	2
Solenopsis sp.02		1
Pseudomyrmecinae		
Pseudomyrmex gracilis (Fabricius, 1804)	3	1
Pseudomyrmex oculatus (Smith, F., 1855)		1
Pseudomyrmex schuppi (Forel, 1901)		1
Total species	14	18



**Fig 3**. Rarefaction curves of ant species captured on *Cecropia pachystachya* trees in the dusty roadside and in the dust-free roadside in Rio Doce State Park and surroundings, southeastern Brazil. Bars denote 95% CI.

Among the architectural variables evaluated, only first branch height (FBH) and circumference at breast height (CBH) were significantly related to ant species richness. Ant species richness on trees ranged from zero to five, being negatively related to FBH (Fig 4; p = 0.02) and positively related to CBH (Fig 5; p = 0.001). Low FBH and large CBH are indicatives of structural complexity, since low FBH trees usually have long and dense crowns, and high CBH trees are generally large. Larger crowns and trees probably support higher resource availability and higher diversity of conditions than smaller trees (Campos et al., 2006) allowing more ant species to live or forage on the same tree. In fact, other studies found similar results of increase in ant and/or other insects' richness with increase in tree or crown size (e.g. Campos et al., 2006; Costa et al., 2011; Neves et al., 2013).

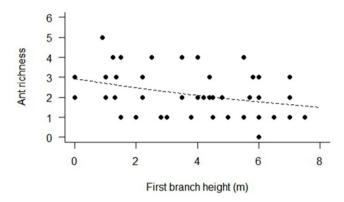


Fig 4. Effect of first branch height (FBH) of *Cecropia pachystachya* trees on ant species richness in Rio Doce State Park and surroundings, southeastern Brazil.

Composition of ant species found on C. pachystachya trees was significantly different between sampling sites ( $R_{ANOSIM} = 0.039$ ; p = 0.04; Fig. 6). SIMPER analysis revealed that *Camponotus* senex (Smith, F., 1858), Crematogaster curvispinosa Mayr, 1862, Camponotus balzani Emery, 1894 and Azteca alfari Emery, 1893 were the species that mostly contributed to differentiation of groups and together explained 40.21% of the dissimilarity between sites (Table 2). These ant genera (Table 2) are commonly associated with myrmecophytes and are classified as dominant arboreal ants (Brandão et al., 2011). Therefore, ant fauna on C. pachystachya trees differed between sampling sites mainly due to dominant species that live on the trees, while opportunistic ants had a smaller contribution to differentiation between dusty and dust-free roadsides. Camponotus senex is conspicuous and can occur both in mature and disturbed forests, nesting in high regions of canopy including dead branches of Cecropia trees (Longino, 2002). The greater frequency of C. senex in the dusty roadside (Table 1) might be related to the abundance of dead branches on C. pachystachya trees in this site, as observed by us in the field. In turn, the abundance of dead branches in the dusty roadside could possibly be caused by road dust effects on C. pachystachya trees such as increased water loss. The second species that most contributed to differentiation between sites - Crematogaster curvispinosa (Myrmicinae) is a very common but inconspicuous ant that feeds on extrafloral nectaries and inhabits initial secondary forests as well as highly disturbed areas such as roadsides (Longino, 2003).

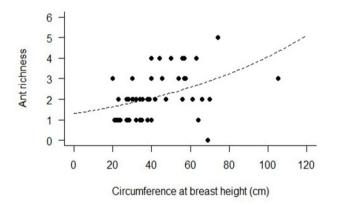
It can tolerate other ant species nests nearby it (e.g. *Camponotus* and *Cephalotes*) and its colonies can occupy *Cecropia* internodes that are located between resident *Azteca* nests (Longino, 2003).

Table 2. Similarity Percentage (SIMPER) analysis outcome. Av. Dissim.: average dissimilarity of taxon between sites; Contrib. %: relative contribution of taxon to dissimilarity between sites; Cumulative %: cumulative contribution of taxon to dissimilarity between sites; Mean adund. 1: mean abundance of taxon in the dusty roadside; Mean abund. 2: mean abundance of taxon in the dust-free roadside.

Taxon	Av. dissim.	Contrib. %	Cumulative %	Mean abund. 1	Mean abund. 2
Camponotus senex	8.202	15.51	15.51	0.36	0.125
Crematogaster curvispinosa	4.816	9.105	24.61	0.08	0.167
Camponotus balzani	4.205	7.949	32.56	0.08	0.167
Azteca alfari	4.047	7.651	40.21	0.96	0.917

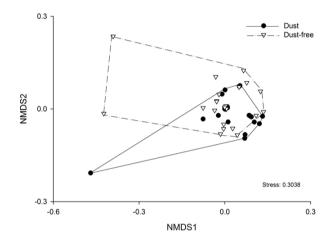
Despite its high tolerance in disturbed areas, *C. curvispinosa* was more abundant inside the park possibly because of the abundance of shrubs in the dust-free roadside in contrast to the scarcity of these in dusty site. Abundant shrubs are indicatives of higher availability of extrafloral nectaries which are important food sources to *C. curvispinosa*. Dust deposition might limit the abundance of shrubs outside the park through negative effects on plants as well as block access to the remaining extrafloral nectaries in the dusty roadside.

As shown by our results, composition of ant communities on *Cecropia pachystachya* trees differed between sites with distinct exposure to dust. The substantial difference in



**Fig 5**. Effect of circumference at breast height (CBH) of *Cecropia pachystachya* trees on ant species richness in Rio Doce State Park and surroundings, southeastern Brazil.

ant species composition between sampling sites, in spite of their similarity (both roadsides of early successional Atlantic Forest) and geographical proximity, suggests a significant effect of road dust on arboreal ant fauna. Dust could directly affect ants hampering some species settlement and survival for instance, or it could indirectly affect ants through effects on vegetation that generate avoidance of dusty trees by some ant species and preference by others (e.g. dead branches for *Camponotus senex*). *Cecropia pachystachya* trees under heavy dust deposition may also have impaired development and growth which may reduce the quantity and quality of essential resources for resident and opportunistic ants. In addition, dust may have effects on associate invertebrate communities and these changes likely affect ant fauna especially through competition for resources. Finally, our results indicated that effects of road dust might transcend impacts on vegetation and might have important impacts on animal associate communities especially ants.



**Fig 6**. Ordination based on incidence of ant species on 50 *Cecropia pachystachya* trees using the Raup-Crick index through analysis of non-metric multidimensional scaling (NMDS).

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