SHORT COMMUNICATION

Influence of its associated ant species on the life history of the myrmecophyte *Cordia nodosa* in French Guiana

Alain Dejean*[1], Angélique Quilichini*, Jacques H. C. Delabie†, Jérôme Orivel*, Bruno Corbara‡ and Marc Gibernau*

* Laboratoire d'Évolution et Diversité Biologique, Université Toulouse III, Bâtiment 4R3, 118 route de Narbonne, 31062 Toulouse cedex 4, France
† U.P.A. Laboratório de Mirmecologia, Convênio UESC/CEPLAC, C.P.7, 45600-000 Itabuna, Bahia, Brazil
‡ Laboratoire de Psychologie Sociale de la Cognition (UMR CNRS 6024), Université Blaise Pascal, 34 avenue Carnot, 63037 Clermont-Ferrand Cedex, France
(*Accepted 12 January 2004*)

Variation in the ant species associated with myrmecophytes (plants that provide their associated ants with nesting space, and sometimes with a complete diet) has been noted at both the regional and local levels, with plant distribution generally wider than that of the ants (Fonseca & Ganade 1996). This is the case for *Cordia nodosa* Lamark (Boraginaceae, subfamily Ehretioideae) whose most frequent associate ant species in Peru are *Allomerus demararae* (Wheeler) and *Azteca* spp. (Yu & Pierce 1998), while *Azteca* sp.1 and *Allomerus octoarticulatus* are the most frequent in Amazonian Brazil and French Guiana, respectively (Fowler 1993, Solano *et al.* 2003). *Cordia nodosa* plants are understorey treelets mostly less than 2 m tall, but taller individuals can be found. Their domatia are swollen, hollow stem nodes that form as the growing shoot tip invaginates through a subapical pore that then closes over when the domatium is mature (Yu 2001). So, plant-ants must reopen the chamber where the pore used to be, thus forming a kind of prostoma.

We hypothesised that differences in the ant species associated with *C. nodosa* individuals from two geographical areas subsequently affect plant growth and reproduction. We therefore sought to verify if all the ant species associated with *C. nodosa* are able to open the domatia, if they protect their plants against herbivores, and if the plants produce fruit.

This study was conducted between 1999 and 2003 during seven 1-mo-long surveys on five hills of pristine rain forest situated around the Petit Saut dam in Sinnamary, French Guiana. In the study area, consisting of hills separated by small valleys with running streams, *C. nodosa* individuals are distributed haphazardly (without any aggregations) at the mid-slopes of the hills (see also Solano *et al.* 2003). We conducted extensive research by selecting all treelets taller than 1.5 m (n = 212) on the five hills (like numerous other myrmecophyte species, small *C. nodosa* do not produce domatia; Heil & McKey 2003). We cut one branch per plant, the domatia of which were decorticated later in the laboratory in order to harvest the ants. For plants apparently devoid of associate ants, all domatia were opened. Due to morphological and ecological similarities in the genus *Allomerus* (see Izzo & Vasconcelos 2002), we thoroughly examined workers from each colony. Voucher specimens of ants were deposited in the *Laboratório de Mirmecologia*, Itabuna, Brazil.

In the study areas *Cordia* individuals were mostly associated with *A. octoarticulatus* or sheltered the colonies of six other ant species, principally *Azteca* sp. 1 and *Crematogaster laevis* (Table 1). The remaining plants were devoid of associate ants or sheltered the incipient colonies of several species.

As the presence of a stoma can permit opportunistic ants to install themselves in the domatia, we checked if the youngest domatia (those in the process of formation excepted) had a stoma. All 20 *Cordia* sheltering adult colonies of *C. laevis*, *Pheidole minutula*, *Pachycondyla* sp. and *Brachymyrmex* sp. had numerous unopened domatia at the ends of their branches. This never occurred on *Cordia* when associated with *Crematogaster limata*, *Azteca* sp. 1 or *A. octoarticulatus*. Also, incipient colonies of both latter species recorded on *Cordia* nested in the young domatia, while the incipient *C. laevis* and *P. minutula*

[1] Corresponding author. Email: dejean@cict.fr

Table 1. Ant species associated with *Cordia nodosa* (n = 212).

Associated ant species	Number of plants	%
Allomerus octoarticulatus Mayr	138	65.1
Azteca sp.1	20	9.4
Crematogaster laevis Mayr*	13	6.1
Crematogaster limata F. Smith	4	1.9
Pheidole minutula Mayr*	5	2.4
Pachycondyla sp.*	1	0.5
Brachymyrmex sp.*	1	0.5
Incipient colonies	8	3.8
No ants	22	10.4

* The colonies sheltered only in domatia situated in the lower parts of the plants, numerous younger domatia being devoid of stoma. One or two incipient colonies per plant were noted for *A. octoarticulatus* (six plants), *Azteca* sp.1 (three plants), *C. laevis* (seven plants), and *P. minutula* (two plants).

Figure 1. Comparisons between the level of defoliation of the six youngest leaves of *Cordia nodosa* individuals and non-myrmecophytic control plants situated within a radius of 3 m (* = P < 0.05; *** = P < 0.001).

colonies occupied lower, older domatia. So, it is likely that a kind of sorting occurs, with only *C. limata*, *Azteca* sp. 1 and *A. octoarticulatus* able to gnaw a stoma in domatia. Colonies of other species, mature or incipient, shelter in the lower, older and already open domatia. Also, all 22 *Cordia* devoid of associate ants had their old domatia pierced with a stoma, while younger domatia did not. These plants probably sheltered an ant colony that moved or died before we began our observations. These results are consistent with those of Solano *et al.* (2003) who concluded that numerous plants were abandoned by their associated ants, mostly *Azteca* sp. 1. Indeed, *C. laevis* (*Crematogaster* sp. 1 in Solano *et al.* 2003), known to move during its life cycle in order to nest in hollow twigs of the leaf litter (Vasconcelos & Davidson 2000), cannot be at the heart of this phenomenon as workers and founding queens of this species seem unable to gnaw a stoma in the *Cordia* domatia.

In our study areas, leaf-cutting ants (Attini of the genera *Atta* and *Acromyrmex*) are the major defoliators. When they attack a plant, the rate of defoliation is extremely variable from one leaf to another, independently of the leaf's age, so that a qualitative analysis (attacked, or not), is informative enough to illustrate the defensive role of the associated plant-ants. In order to focus our study on this defensive role, we chose a threshold of 10% of the leaf surface defoliated under which the leaf was considered as protected. Indeed, defoliation under 10% was rare and mostly concerned attacks by winged insects (Orthoptera and adult chrysomelid beetles) that had begun to gnaw a leaf before being discovered and attacked by plant-ants. They are able to escape by jumping, flying away or dropping while chrysomelid and Lepidoptera larvae are easily eliminated by plant-ants at the egg or first larval stages. As a result, in order to examine ant protection against herbivory, we noted for each selected plant the number of leaves attacked to more than 10% of their surface out of the six youngest. The same procedure was conducted each time on a control
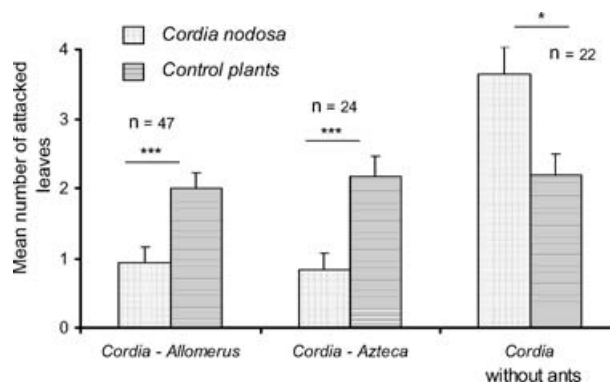
non-myrmecophytic plant nearby of about the same size. For statistical comparisons, the attack rates of the plants were normalized by square-root transformation before an analysis of variance using SYSTAT 8.0. For multiple comparisons we performed a *post-hoc* test for pairwise mean differences with a Bonferroni adjustment.

Allomerus octoarticulatus- or *Azteca* sp. 1-associated *Cordia* were similarly protected as the rate of defoliation was similar (P > 0.9; Figure 1). They were significantly less attacked than individuals devoid of associate ants (P < 0.001 in both cases), and the neighbouring non-myrmecophytic control plants (mean ± SE; control vs. *C. nodosa* sheltering (1) *A. octoarticulatus*: 0.94 ± 0.23 vs. 2.00 ± 0.22 attacked leaves out of the six youngest of each plant; df = 46, F = 11.7, P = 0.001; (2) *Azteca* sp. 1: 0.83 ± 0.25 vs. 2.17 ± 0.29; df = 23, F = 15.0, P < 0.001; and (3) devoid of associate ants: 3.63 ± 0.41 vs. 2.18 ± 0.31; df = 21, F = 4.8, P < 0.05).

We noted that *A. octoarticulatus* and *Azteca* sp. 1 workers protect their host plant by attacking the Attine scouts, preventing them from recruiting nestmates to defoliate their host plants (see also Michelangeli 2003). Also, these workers are very attracted to young *C. nodosa* leaves that they patrolled day and night. We noted 19.2 ± 1.3 (n = 120) *A. octoarticulatus* and 1.5 ± 0.2 (n = 80) larger *Azteca* sp.1 workers per leaf, while this was not the case for *C. laevis* (0.7 ± 0.2 of these small workers per leaf; n = 40; four diurnal and four nocturnal series of controls on 15, 10 and 5 plants hosting *A. octoarticulatus*, *Azteca* sp.1 and *C. laevis*, respectively) (see also Yu 2001).

These results confirm those of numerous previous studies reporting that myrmecophytes are protected by their plant-ants (Heil & McKey 2003, Michelangeli 2003, Moog *et al.* 2002, Nery & Vasconcelos 2003, but see Fowler 1993). Nevertheless, *Cordia* devoid of associate ants were unexpectedly more defoliated than the control plants (Figure 1), suggesting that the difference is because control plants produce chemical defences that

were completely or partially lacking in *Cordia* (see Izzo & Vasconcelos 2002).

We verified fruit presence-absence on 122 *Cordia* in April 2002 and March 2003 (i.e. short dry season or major fruit production period for *Cordia*; when present, the fruits are abundant). For statistical comparisons we used Fisher's exact-tests. Appropriate probabilities were adjusted for the number of simultaneous tests, using the sequential Bonferroni procedure (Rice 1989).

We indeed observed that, like *A. demararae* in Peru (Yu & Pierce 1998), *A. octoarticulatus* workers destroyed certain flowers at the budding stage, but we focused our study on fruit production. We noted that fruit presence was rare in 1.5–2-m-tall plants sheltering *A. octoarticulatus* (2 plants with fruit vs. 69 without) while all other plants sheltering another ant species produced fruit (12, 8 and 2 *C. nodosa* hosting *Azteca* sp.1, *C. laevis* and *C. limata*, respectively). Most *C. nodosa* devoid of associated ants or hosting incipient colonies produced fruit (9 with and 3 without fruit). The comparison *Allomerus*-inhabited plants vs. four other cases pooled resulted in a significant difference (Fisher's exact-test and the sequential Bonferroni procedure: P < 0.001). Nevertheless, taller individuals (> 2.5 m tall) sheltering *A. octoarticulatus* can produce fruit (8 plants with vs. 6 without; comparison with 1.5–2-m-high plants: P < 0.001).

As a result, in well-protected *Allomerus*-inhabited plants, workers do not destroy all the flowers in tall individuals. This could be due to a decrease in colony activity (non-apparent) or to a change in worker behaviour when the size of the colonies reaches a certain threshold and produces numerous winged sexuals. By destroying flowers early and preventing fruit production, the colonies favour plant growth and subsequent domatia production (Yu & Pierce 1998, see also Fonseca 1999).

The *Azteca* sp. 1-inhabited plants, well protected, began to produce fruit when relatively small, but precociously lost this associate. This could be because the colonies were limited by host-plant size as, in contrast to *Azteca* spp. associated with *C. nodosa* in Peru (Yu & Pierce 1998), they do not build satellite carton nests. After the loss of their associated ants, these plants then sheltered *A. octoarticulatus*, *Azteca* sp. 1 again, or an opportunistic ant species. In both latter cases they produced fruit, but were abandoned and defoliated again. In Peru, both *A. demararae* and *Azteca* spp. colonies protect the foliage of their associate *C. nodosa* and do not abandon them, but the former ant species castrates its associated plants (Yu & Pierce 1998).

Crematogaster limata needs to be considered separately as this is a dominant species able to build carton nests. Although we did not note incipient *C. limata* colonies, we gathered evidence that workers can open *C. nodosa* domatia, but colonies move then probably due to limited nesting space in the host plant domatia as they do not build carton nests (see Fonseca 1999).

Compared with leaf pouches that can be destroyed by defoliators in most other myrmecophytes of the Guianan understorey, the perennial domatia of *C. nodosa* permit this plant to be recolonized by plant-ants after being abandoned and defoliated. *Azteca* sp. 1-associated plants, likely to be abandoned once, if recolonized by *A. octoarticulatus*, or several times, if recolonized by *Azteca* sp. 1 or an opportunistic ant species, will suffer one or several major defoliation(s), with a lethal risk if defoliated during a drought. They begin to produce fruit early in their life, while *A. octoarticulatus*-associated plants, well protected but castrated for the greater part of their life, produce fruit later. In both cases offspring can be produced. Therefore, *C. nodosa* in the Guianan environment has developed two kinds of associations with ants. One relationship is safe, but plants produce offspring late; the other is risky, but permits offspring production early.

ACKNOWLEDGEMENTS

LITERATURE CITED

FONSECA, C. R. 1999. Amazonian ant-plant interactions and the nesting space limitation hypothesis. *Journal of Tropical Ecology* 15:807–825.

FONSECA, C. R. & GANADE, G. 1996. Asymmetries, compartments and null interactions in an Amazonian ant-plant community. *Journal of Animal Ecology* 65:339–347.

FOWLER, H. G. 1993. Herbivory and assemblage structure of myrmecophytous understory plants and their associated ants in the central Amazon. *Insectes Sociaux* 40:137–145.

HEIL, M. & MCKEY, D. 2003. Protective ant-plant interactions as model systems in ecological and evolutionary research. *Annual Review of Ecology, Systematics and Evolution* 34:425–553.

IZZO, T. J. & VASCONCELOS, H. L. 2002. Cheating the cheater: domatia loss minimizes the effects of ant castration in an Amazonian ant-plant. *Oecologia* 133:200–205.

MICHELANGELI, F. A. 2003. Ant protection against herbivory in three species of *Tococa* (Melastomataceae) occupying different environments. *Biotropica* 35:181–188.

MOOG, J., FELDHAAR, H. & MASCHWITZ, U. 2002. On the caulinary domatia of the SE-Asian ant-plant *Zanthoxylum myriacanthum* Wall.

ex Hook. f. (Rutaceae) their influence on branch statics, and the protection against herbivory. *Sociobiology* 40:547–574.

NERY, A. S. & VASCONCELOS, H. L. 2003. Growth and survival of incipient ant colonies in two Amazonian ant-plants: effects of habitat, host-plant, and mode of colony founding (Hymenoptera: Formicidae). *Sociobiology* 42:151–162.

RICE, W. R. 1989. Analyzing tables of statistical tests. *Evolution* 43:223–225.

SOLANO, P. J., DUROU, S., CORBARA, B., QUILICHINI, A., CERDAN, P., BELIN-DEPOUX, M., DELABIE, J. H. C. & DEJEAN, A. 2003.

Myrmecophytes of the understory of French Guianian rainforests: their distribution and their associated ants. *Sociobiology* 41:605–614.

VASCONCELOS, H. L. & DAVIDSON, D. W. 2000. Relationship between plant size and ant associate in two Amazonian ant-plants. *Biotropica* 32:100–111.

YU, D. W. 2001. Parasites of mutualisms. *Biological Journal of the Linnean Society* 72:529–546.

YU, D. W. & PIERCE, N. E. 1998. A castration parasite of an ant-plant mutualism. *Proceedings of the Royal Society of London, Biological Sciences* 265:375–382.