

Ant–plant interaction in the Neotropical savanna: direct beneficial effects of extrafloral nectar on ant colony fitness

Jonas Byk · Kleber Del-Claro

Received: 19 May 2010 / Accepted: 16 August 2010 / Published online: 8 September 2010
© The Society of Population Ecology and Springer 2010

Abstract Current evidence suggests that ant–plant relationships may influence species composition, abundance, and interactions at the community scale. The main resource that plants offer to ants is extrafloral nectar (EFN) and the major part of published studies shown benefits from ants to plants possessing EFNs. However, the complementary question of whether and how ants benefit from EFNs is rarely addressed. Here, we present the results of a long-term study to demonstrate whether EFN has a positive effect on ant colony fitness. We quantified colony growth rate, survival and the final weight of individuals as measures of benefit derived from EFN. Our results provide clear evidence that EFN can have a significant positive impact on the survivorship, growth and reproduction of the Myrmicinae *Cephalotes pusillus*. In fact, a diet rich in EFN (providing at least 30 cal per day) resulted in five times more individuals per colony, greater body weights, and more eggs. These results have shed new light on the relationships between ants and EFN-bearing plants such as in tropical and temperate systems. The ant *C. pusillus* is the first case in which we have firm evidence that EFN improves colony growth and development, corroborating more than 100 years of experimental evidence of benefits to plants in these widespread relationships.

Keywords *Cephalotes* · *Chamaecrista* · Colony growth rate · Mutualism · Tropical

Introduction

One of the biggest questions in ecology is what determines the distribution and assembly of species. In communities, the assembly of species is linked directly or indirectly through resources and consumption (Borer et al. 2005). Thus, studies on the direct trophic relationships that influence species interactions and biodiversity are basic to our comprehension of how communities are established and organized (Thompson 2005). Because of this, studies of interactions such as parasitism and mutualism can provide particular insight into community dynamics (Simberloff 2006; Clement et al. 2008), and allow us to predict community stability through the study of outcomes of interactions to related species (Sachs and Simms 2006). Ant–plant relationships have made enormous contributions to our understanding of communities (Bronstein 1998; Rico-Gray and Oliveira 2007 and references therein).

The commonest resource plants offer to ants is extrafloral nectar (EFN), a liquid substance rich in carbohydrates with dilute concentrates of amino acids, lipids, phenols, alkaloids and volatile organic compounds (González-Teuber and Heil 2009). Carbohydrates have been suggested to be key resources for arboreal ants (Davidson et al. 2003). Extrafloral nectaries (EFNs) are present in at least 93 plant families and 332 genera round the world (Koptur 1992). Although few studies have directly investigated the effects of associations between ants and EFN-bearing plants on communities, current evidence suggests that EFN may influence species composition, abundance, and interactions

J. Byk
Programa de Pós-Graduação em Ecologia e Conservação de Recursos Naturais, Universidade Federal de Uberlândia, Uberlândia, MG 38400-902, Brazil

K. Del-Claro (✉)
Instituto de Biologia LECl, Universidade Federal de Uberlândia, Uberlândia, MG 38400-902, Brazil
e-mail: delclaro@ufu.br

at the community scale (Rico-Gray et al. 1998; Blüthgen et al. 2000; Heil et al. 2001).

To qualify an interaction as mutualistic, benefits to both participants must be demonstrated (Cushman and Beattie 1991). However, experiments to prove benefits to both participants are difficult to conduct in field or laboratory conditions (Fiedler and Saam 1995; Lach et al. 2009). Furthermore, some studies have revealed that ant–plant mutualisms can be exploited by several species, mainly ants (e.g., Janzen 1975; Letorneau 1990), that obtain resources from the plants but provide no services. These species are termed cheaters, exploiters, or parasites of the mutualism (Clement et al. 2008). Nevertheless, studies that show benefits from ants to plants possessing EFNs are predominant in the literature (Rico-Gray and Oliveira 2007; Rosumek et al. 2009), but the complementary question of whether and how ants benefit from EFNs, such as in parasitic and mutualistic relationships, is rarely addressed (Bronstein 1998; Rosumek et al. 2009). We extensively searched in the literature for evidence of benefit to ants that visit EFN-bearing plants, tend exudate-secreting insects such as hemipterans and butterfly larvae, or collect seeds, and we found only five published papers that demonstrated clear benefit to the ants. Three studies investigated the benefits of elaiosome-bearing seeds: the first showed positive effect of elaiosome on gyne abundance (Morales and Heithaus 1998), the second in larval weight and abundance (Gammans et al. 2005), and the third in pupal abundance (Fokuhl et al. 2007). In a short time experiment, Cushman et al. (1994) showed that ants with access to lycaenid butterfly larva had 40% greater survival than workers with access only to plants. More recently, Lach et al. (2009) in two short laboratory experiments showed that in EFN-bearing plants, herbivory can induce a higher production of EFN that can improve worker survivorship 7–11 times greater than unfed ants. Thus, despite the hundreds of studies on ant–plant interactions, we still lack clear evidence from long-term experimental studies of the effect of EFN on ant fitness.

Here, we present the results of a long-term study, in a controlled laboratory environment, to demonstrate whether EFN has a positive effect on ant colony fitness. For this study, we used one of the most studied arboreal ant species in the Brazilian tropical savanna, *Cephalotes pusillus* (e.g., Korndörfer and Del-Claro 2006; Sendoya et al. 2009) and the EFN-bearing shrub *Chamaecrista desvauxii* (Caesalpinioidea). We hypothesized that ant colonies with access to EFN-bearing plants would have higher benefit than colonies without access, because they would have an additional source of energy and amino acids from EFN. We quantified colony growth rate, survival and the final weight of individuals as measures of benefit derived from EFN.

Materials and methods

Species descriptions

Cephalotes pusillus Klug (Myrmicinae) is a black and polymorphic species of ant, with size differences between workers (3–4 mm), soldiers (5–6 mm) and gyne (9–11 mm). They nest in natural cavities of trees, or in cavities produced by beetles, but can also be found in dead stems or trunks on the ground (Powell 2008). Mature colonies have only one gyne (queen) and 170 workers (mean 173 ± 19 SD; $n = 16$) and 25 soldiers (mean 24 ± 7 SD; $n = 16$). These ants have small mandibles and feed primarily plant and insect exudates (Del-Claro and Oliveira 2000; Davidson et al. 2003). In nature, their diet is composed by EFN (74%, Fig. 1) and hemipteran honeydew (17%), fallen fruits (2%), feces of birds (2%), dead animal matter (5%), and rarely by slow-moving prey (Del-Claro et al. 2002). Previous field observations showed that, in the tropical savanna cerrado, the reproductive period begins in late September and October, and in December initial nests can be easily found in stems of *Stryphnodendron polyphyllum* (Mimosaceae), *Ouratea spectabilis* and *O. hexasperma* (Ochnaceae).

The genus *Chamaecrista* (Moench) (Caesalpinioideae) is widely distributed in the tropical savanna, where there are more than 130 species, several of them bearing EFNs (Irwin and Barneby 1982), and only recently studied in terms of the outcomes of ant–plant associations (Nascimento and Del-Claro 2010). The shrub *C. desvauxii* presents yellow flowers and pinnately compound leaves with EFNs in the base and tip of each leaf (Fig. 1). These EFNs are active during the whole year. This species is common in the cerrados of southeast Brazil.

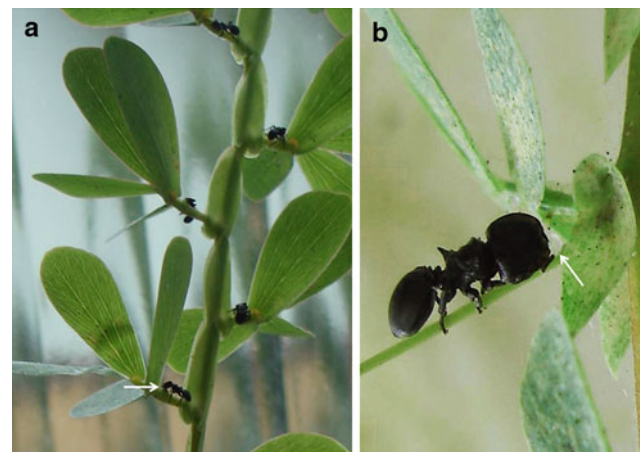


Fig. 1 **a** General view of a *Chamaecrista desvauxii* stem. The arrow is pointing an EFN being visited by *C. pusillus*. **b** *Cephalotes pusillus* worker collecting one droplet of extrafloral nectar (in the tip of arrow)

Experiments

Extrafloral nectar production and quality can vary among plant species, plants of the same species and leaves of the same plant, depending also on season, soil quality and herbivory (Koptur 1992; Heil and McKey 2003; González-Teuber and Heil 2009). To reduce individual variation in nectar quality and production, we collected in the field (tropical savanna reserve, Clube de Caça e Pesca Itororó, CCPIU, Uberlândia, MG, Brazil) seeds of one shrub and cultivated it in laboratory conditions (8 h of light, 23–27°C, 50–60% humidity) in pots (500 ml) previously prepared with the same quantity and quality of soil substrate. Each flowerpot was placed in the right side of a plastic rectangular basin (20 × 30 cm length and 10 cm height), entirely covered by a thin and transparent tulle. These cages ($n = 30$) were prepared between October and November 2007. Seeds germinated in 2 weeks and, after 45 days (December), we had seedlings with one main branch, 8–10 cm tall. The *C. desvauxii* EFNs produced nectar during the whole day. However, it was more intense in dawn (0630 hours) and dusk (1730 hours), when we could sample it. Thus, to know the contribution of each EFN to the ants in terms of calories, with a microcapillary tube (10 μ l), we measured the sugar concentration in a refractometer (Eclipse®). The values recorded are in Brix scale and represent the percentage of sugar (sucrose) per volume, and we converted it to mass, according to Kearns and Inouye (1993). The quantity of nectar found in each EFN was finally converted to calories according to Dafni (1992; 1 mg of sugar = 4 cal). One experiment that had used seeds of many plants perhaps presented a more general result considering the great variability of tropical plants. Thus, we compared the nectar (volume production and sugar concentration) between experimental control plants with different shrubs ($n = 15$) in natural conditions. We did not observe significant differences.

In December 2007, also in the tropical savanna (CCPIU), we collected nests ($n = 30$) of *C. pusillus* in the initial phase of establishment. They were transported to laboratory and were conditioned in glass tubes (five per colony; tubes of 10 cm long and 1 cm diameter, filled up to 2 cm with water concealed by hydrophilic cotton). Colonies were fed with water and 5 g of protein diet (Bhatkar and Whitcomb 1970) for 2 weeks. In the last week of December, we manipulated colonies that had all castes present ($n = 22$) to obtain in each one the final composition: 1 queen, 3 workers, 2 soldiers and 5 eggs. These colonies were divided, by the flip of a coin, in control or treatment group. In the first week of January 2008, the colonies were introduced in the cages with seedlings of *C. desvauxii*. In control groups, plants were maintained with 14 active EFNs, but in the treatment ones, the EFNs

were damaged with a needle, to stop nectar production. Excessive (in control) or news EFNs (in treatment plants) were damaged, causing sclerotization. Ants continued to visit plants after EFN manipulation. In both groups, the plants received 30 ml of water and ants were fed with water and 5 g of protein diet every 2 days. This quantity of protein diet was enough to feed the colony, because it was never finished in 2 days. We took care of the colonies for 1 year, and in December, we counted the number of individuals and eggs in each colony. All queens, eggs, workers, and soldiers in each colony were weighed.

Data normality was tested using Kolmogorov–Smirnov test. If normal, mean values were compared using Student's *t* test, if not normal, we used Mann–Whitney test, both one-tailed.

Results

Extrafloral nectar had a significant and positive impact on ant fitness in terms of colony growth rate, survival and weight of eggs and individuals. Colonies fed with EFN developed better than colonies lacking EFNs, producing a higher number of individuals (Fig. 2a) and eggs (Fig. 2b). The analysis of EFN sugar concentration revealed that each gland produced at least 2.5 μ l of nectar per day (mean 2.5 ± 0.1 SD; $n = 264$ samples; two glands of each control plant, sampled monthly), what corresponded to 2.16 cal. Because each plant in the control group had 14 active EFNs, we expected that each plant offered to ants at least 7.56 mg of nectar per day or 30.26 cal. This

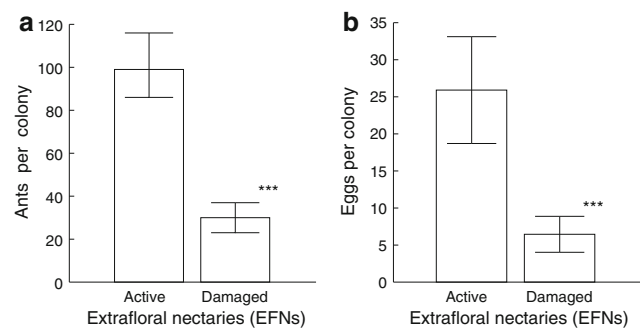


Fig. 2 **a** Number of individuals (mean \pm SD) in colonies of the ant *Cephalotes pusillus* (Myrmicinae) that had access to plants of *Chamaecrista desvauxii* (Caesalpinaceae) with active EFNs ($n = 11$ control colonies) or plants with damaged and non productive EFNs ($n = 11$ treatment colonies) during 1 year. ***Statistical difference between groups ($P < 0.0001$, $t = 8.1164$, $df = 20$). **b** Number of eggs (mean \pm SD) in colonies of the ant *Cephalotes pusillus* (Myrmicinae) that had access to plants of *Chamaecrista desvauxii* (Caesalpinaceae) with active EFNs ($n = 11$ control colonies) or plants with damaged and non productive EFNs ($n = 11$ treatment colonies) after 1 year. ***Statistical difference between groups ($P < 0.0001$, $t = 8.4896$, $df = 20$)

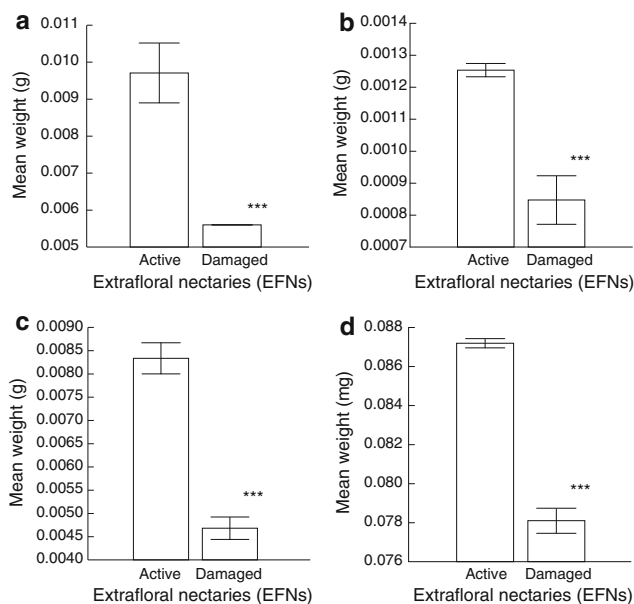


Fig. 3 Comparative weight (mean \pm SE, **a** queens, **b** workers, **c** soldiers, **d** eggs) of *Cephalotes pusillus* ants bred with access to *Chamaecrista desvauxii* plants with active (control group, $n = 11$) and damaged and non-active extrafloral nectaries (treatment group, $n = 11$). ***Statistical difference ($P < 0.0001$; Mann–Whitney test)

additional energetic and protein supplement had a significant positive impact on the final weight of individuals, queens ($U = 0.00$; $n = 11$; $P < 0.0001$), workers ($U = 0.00$; $n = 11$; $P < 0.0001$), soldiers ($U = 0.00$; $n = 11$; $P < 0.0001$) and eggs ($U = 1.00$; $n = 11$; $P < 0.0001$) of control ant colonies (Fig. 3).

Discussion

EFN effect on ant colonies

Despite the large number of studies that have focused on ant–plant interactions in both temperate and tropical systems (Rosumek et al. 2009), our results and that of Lach et al. (2009) are the first to provide firm evidence that EFN can have a significant positive impact on the survivorship, growth and reproduction of ants. In the case of *C. pusillus*, a diet rich in EFN (providing at least 30 cal per day) resulted in five times more individuals per colony, greater body weights, and more eggs. Despite being conducted in the laboratory, our results can be interpreted as holding true in nature. Data from the same field site showed that approximately 74% of *C. pusillus* diet is composed by EFN (Del-Claro et al. 2002). This is considered a typical EFN-gathering ant in the tropical savanna (e.g., Byk and Del-Claro 2010 and citations therein). Although EFN may vary in production, composition and concentration, both

within and among plant species (Heil et al. 2000; Blüthgen et al. 2004a), carbohydrates, mainly sucrose, fructose, and glucose usually comprise the major part ($\geq 90\%$) of nectar (Blüthgen et al. 2004a; but see also González-Teuber and Heil 2009). In ant colonies, some authors (Davidson 1998) have pointed out carbohydrates as the primarily fuel for workers in foraging and defense activities, indirectly benefiting the other castes. The results presented here also show that this benefit can be direct, because worker body weights were significantly higher. Carbohydrates have been hypothesized to be the key resources for maintaining worker activity levels of ecologically dominant ant species (Davidson 1998). EFN possesses a variety of qualities that place it as a key resource for ants. For example, EFN is predictable in time and space, and is a reliable source of sugar, water and amino acids (Koptur 1992; Rudgers and Gardner 2004; González-Teuber and Heil 2009) that increase many measures of colony fitness, including body size, colony growth rate, and brood growth. By preventing nectar accumulation in EFNs, ants may induce continue production of nectar by host plants (Heil et al. 2000). On the other hand, plants attacked by herbivores may increase nectar production to become more attractive to protecting ants (Pulice and Packer 2008; Lach et al. 2009; but see also Korndörfer and Del-Claro 2006). Plant nectar and hemipteran honeydew have a key role structuring ant communities in the rainforest, with dominant species avoiding the use of same plants, but co-occurring with non-dominant ant species (Blüthgen et al. 2004b). *Cephalotes pusillus* is not an aggressive ant, and commonly co-occurs with other species in the same source of resource in the savanna (Del-Claro and Oliveira 2000; Sendoya et al. 2009; Nascimento and Del-Claro 2010). As EFN is the main source of energy, water and protein to this ant species, competition with other EFN visitors, mainly ants, can obligate individuals of a *C. pusillus* colony to forage in more than one plant or resource type (Del-Claro et al. 2002). However, Cephalotini ants normally inhabit pre-existing cavities, like abandoned tunnels of wood-boring insects (Powell 2008). It is a common strategy of *C. pusillus* in cerrado vegetation, nesting in cavities of EFN-bearing plants like Vochysiaceae (*Qualea grandiflora*, *Q. multiflora* and *Q. parviflora*), Ochnaceae (*Oratea spectabilis* and *O. hexasperma*), Mimosaceae (*Stryphnodendron polyphyllum* and *S. adstringens*) and Caryocaraceae (*Caryocar brasiliensis*). This behavior means that this non-aggressive ant could dominate an important resource by proximity and numeric superiority (Del-Claro et al. 2002). *Cephalotes pusillus* can also possess satellite nests in distinct parts of same plant or in fallen trunks close to EFN sources, like shrubs of *C. desvauxii*, improving the colony's competitive capabilities (K. Del-Claro, unpublished data).

Implications to the study of communities

Several authors have demonstrated that associations between ants and EFN-bearing plants can decrease foliar herbivory (Rutter and Rausher 2004) and/or increase fruit set (Nascimento and Del-Claro 2010). However, some studies have shown that the positive effects of ants on ant–plant associations are not universal (O’Dowd and Catchpole 1983; Rashbrook et al. 1992; Rosumek et al. 2009). Although *C. pusillus* is one of many species that have been cited as providing benefits to plants (Korndörfer and Del-Claro 2006; Nascimento and Del-Claro 2010), several recent studies have demonstrated that, in some cases, this ant species may provide no plant protection (Sendoya et al. 2009), or may even act as a parasite of mutualism (Byk and Del-Claro 2010). Studies that investigated the strategies of parasite ants in ant–plant interactions showed that the cheater relies less on host-derived food resources than do the mutualists (Clement et al. 2008). In the present case, if *C. pusillus* may be considered a parasite as predicted by Byk and Del-Claro (2010), our data contradict the observations made by Clement et al. (2008) in myrmecophytic systems. Because *C. pusillus* strongly depends on EFN (e.g., Del-Claro et al. 2002), we provide the first evidence that EFN is an important resource for an ant parasite of mutualisms.

Organisms interact not only directly but also indirectly, and indirect effects are important forces driving ecological communities (Ohgushi 2008) that can cause variation in the outcomes of interspecific relationships (Del-Claro and Oliveira 2000). Although few studies have directly investigated the effects of EFN on communities (Rosumek et al. 2009), current evidence suggests that EFN may influence species composition, abundance, and interactions at the community scale (Keeler 1978; Rico-Gray et al. 1998). Thus, the prevalence of EFN-bearing plants, reaching up to 30% in some regions (Blüthgen et al. 2000; Rico-Gray and Oliveira 2007), shows the potential of EFN to mediate species interactions beyond the simple tri-trophic food chain of plant–herbivores–enemies (Rudgers and Gardner 2004). We know that the outcomes of interactions vary among populations in space and time (Bronstein 1998). Species in local populations are likely to experience different patterns of selection, with reciprocal selection between ants and plants, for example, occurring only in some locations (“Geographic mosaic theory of coevolution”; Thompson 2005). In this context, the study of reciprocal benefits in ant–EFN-bearing plants systems in multiple regions may be an important tool for testing differences among populations in traits shaped by coevolutionary processes, and as a predictor of geographic structures coevolution (Thompson 2005). This knowledge will contribute to our understanding of how variation in the outcomes of multispecies interactions

affects evolution of EFN traits. Knowing that EFN is an important resource for ants with measurable benefits is a basic step in this context.

In conclusion, our results have shed new light on the relationships between ants and EFN-bearing plants. The ant *C. pusillus* is the first case in which we have firm evidence that EFN improves colony growth and development, corroborating more than 100 years of experimental evidence of benefits to plants in these widespread relationships.

Acknowledgments We thank to Judith Bronstein, Heraldo L. Vasconcelos, Sérgio P. Ribeiro, Gustavo Q. Romero and three anonymous referees for valuable comments on the early version of the manuscript. We specially thank Michele Lanan for suggestions and a strong English review. Authors thank financial support from Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (J. Byk/graduate fellowship) and Conselho Nacional de Ciência e Tecnologia (K. Del-Claro, PQ/research grant).

References

- Bhatkar A, Whitcomb WH (1970) Artificial diet for rearing various species of ants. Fla Entomol 53:229–232
- Blüthgen N, Verhaagh M, Goitia W, Jaffe K, Morawetz W, Barthlott W (2000) How plants shape the ant community in the Amazonian rainforest canopy: the key role of extrafloral nectaries and homopteran honeydew. Oecologia 125:229–240
- Blüthgen N, Gottsberger G, Fiedler K (2004a) Sugar and amino acid composition of ant-attended nectar and honeydew sources from an Australian rainforest. Austral Ecol 29:418–429
- Blüthgen N, Stork NE, Fiedler K (2004b) Bottom-up control and co-occurrence in complex communities: honeydew and nectar determine a rainforest ant mosaic. Oikos 27:344–358
- Borer ET, Seabloom EW, Shurin JB, Anderson KE, Blanchette CA (2005) What determines the strength of a trophic cascade? Ecology 86:528–537
- Bronstein JL (1998) The contribution of ant–plant protection studies to our understanding of mutualism. Biotropica 30:150–161
- Byk J, Del-Claro K (2010) Nectar- and pollen-gathering *Cephalotes* ants provide no protection against herbivory: a new manipulative experiment to test ant protective capabilities. Acta Ethol 13:33–38
- Clement LW, Köppen SCW, Brand WA, Heil M (2008) Strategies of a parasite of the ant–*Acacia* mutualism. Behav Ecol Sociobiol 62:953–962
- Cushman JH, Beattie A (1991) Mutualisms: assessing the benefits to hosts and visitors. Trends Ecol Evol 6:193–195
- Cushman JH, Rashbrook VK, Beattie AJ (1994) Assessing benefits to both participants in a Lycaenid–ant association. Ecology 75:1031–1041
- Dafni A (1992) Pollination ecology: a practical approach (the practical approach series). Oxford University Press, New York
- Davidson DW (1998) Resource discovery versus resource domination in ants: a functional mechanism for breaking the trade-off. Ecol Entomol 23:484–490
- Davidson DW, Cook SC, Snelling RR (2003) Explaining the abundance of ants in lowland tropical rainforest canopies. Science 300:969–972
- Del-Claro K, Oliveira PS (2000) Conditional outcomes in a neotropical treehopper–ant association: temporal and species-specific variation in ant protection and homopteran fecundity. Oecologia 124:156–165

- Del-Claro K, Santos JC, Júnior ADS (2002) Etograma da formiga arborícola *Cephalotes pusillus* (Klug, 1824) (Formicidae: Myrmicinae). Rev Etol 4:31–40 (in Portuguese with English abstract)
- Fiedler KL, Saam C (1995) Ants benefit from attending facultatively myrmecophilous Lycaenidae caterpillars: evidence from a survival study. Oecologia 104:316–322
- Fokuhl G, Heinze J, Poschlod P (2007) Colony growth in *Myrmica rubra* with supplementation of myrmecochorous seeds. Ecol Res 22:845–847
- Gammans N, Bullock JM, Schonhogge K (2005) Ants benefit in a seed dispersal mutualism. Oecologia 146:43–49
- González-Teuber M, Heil M (2009) Nectar chemistry is tailored for both attraction of mutualists and protection from exploiters. Plant Sign Behav 4:809–813
- Heil M, McKey D (2003) Protective ant–plant interactions as model systems in ecological and evolutionary research. Annu Rev Ecol Evol Syst 34:425–453
- Heil M, Fiala B, Baumann B, Linsenmair KE (2000) Temporal, spatial and biotic variations in extrafloral nectar secretion by *Macaranga tanarius*. Funct Ecol 14:749–757
- Heil M, Koch T, Hilpert A, Fiala B, Boland W, Linsenmair KE (2001) Extrafloral nectar production of the ant-associated plant, *Macaranga tanarius*, is an induced, indirect, defensive response elicited by jasmonic acid. Proc Natl Acad Sci USA 98:1083–1088
- Irwin HS, Barneby R (1982) The American Cassinae A synoptical revision of Leguminosae tribe Cassieae subtribe Cassinae in The New World. Mem New York Bot Gard 35:1–918
- Janzen DH (1975) *Pseudomyrmex nigropilosa*: a parasite of a mutualism. Science 188:936–937
- Kearns CA, Inouye D (1993) Techniques for pollinations biologists. Colorado University Press, Niwot
- Keeler KH (1978) Insects feeding at extrafloral nectaries of *Ipomoea carnea* (Convolvulaceae). Entomol News 89:163–168
- Koptur S (1992) Extrafloral nectary mediated interactions between insects and plants. In: Bernays E (ed) Insect–plant interactions. CRC Press, Boca Raton, pp 81–129
- Korndörfer AP, Del-Claro K (2006) Ant defense versus induced defense in *Lafoensia pacari* (Lythraceae), a myrmecophilous tree of the Brazilian cerrado. Biotropica 38:786–788
- Lach L, Hobbs ER, Majer EJD (2009) Herbivory-induced extrafloral nectar increases native and invasive ant worker survival. Popul Ecol 51:237–243
- Letorneau DK (1990) Code of ant–plant mutualism broken by parasite. Science 248:215–217
- Morales MA, Heithaus ER (1998) Food from seed-dispersal mutualism shifts sex ratios in colonies of the ant *Aphaenogaster rudis*. Ecology 79:734–739
- Nascimento EA, Del-Claro K (2010) Ant visitation to extrafloral nectaries decreases herbivory and increases fruit set in *Chamaecrista debilis* (Fabaceae) in a Neotropical savanna. Flora. doi: 10.1016/j.flora.2009.12.040
- O’Dowd DJ, Catchpole EA (1983) Ants and extrafloral nectaries: no evidence for plant protection in *Helichrysum* spp.–ant interactions. Oecologia 59:191–200
- Ohgushi T (2008) Herbivore-induced indirect interaction webs on terrestrial plants: the importance of non-trophic, indirect, and facilitative interactions. Entomol Exp App 128:217–229
- Powell S (2008) Ecological specialization and the evolution of a specialized caste in *Cephalotes* ants. Funct Ecol 22:902–911
- Pulice CE, Packer AA (2008) Simulated herbivory induces extrafloral nectar production in *Prunus avium*. Funct Ecol 22:801–807
- Rashbrook VK, Compton SG, Lawton JH (1992) Ant–herbivore interactions: reasons for the absence of benefits to a fern with foliar nectaries. Ecology 73:2167–2174
- Rico-Gray V, Oliveira PS (2007) The ecology and evolution of ant–plant interactions. The University of Chicago Press, Chicago
- Rico-Gray V, Palacios-Rios M, Garcia-Franco JG, Mackay WP (1998) Richness and seasonal variation of ant–plant associations mediated by plant-derived food resources in the semiarid Zapotitlan Valley, Mexico. Am Mid Nat 140:21–26
- Rosumek FB, Silveira FAO, Neves FS, Barbosa NP, Diniz L, Oki Y, Pezzini F, Fernandes GW, Cornelissen T (2009) Ants on plants: a meta-analysis of the role of ants as plant biotic defenses. Oecologia 160:537–549
- Rudgers JA, Gardner MC (2004) Extrafloral nectar as a resource mediating multispecies interactions. Ecology 85:1495–1502
- Rutter MT, Rausher MD (2004) Natural selection on extrafloral nectar production in *Chamaecrista fasciculata*: the costs and benefits of a mutualism trait. Evolution 58:2657–2668
- Sachs JL, Simms EL (2006) Pathways to mutualism breakdown. Trends Ecol Evol 21:585–592
- Sendoya SF, Freitas AVL, Oliveira PS (2009) Egg-laying butterflies distinguish predaceous ants by sight. Am Nat 174:134–140
- Simberloff D (2006) Invasional meltdown 6 years later: important phenomenon, unfortunate metaphor, or both? Ecol Lett 9:912–919
- Thompson JN (2005) The geographic mosaic of coevolution. University of Chicago Press, Chicago