

# Interaction between ants, extrafloral nectaries and insect herbivores in Neotropical coastal sand dunes: herbivore deterrence by visiting ants increases fruit set in *Opuntia stricta* (Cactaceae)

P. S. OLIVEIRA,\* V. RICO-GRAY,† C. DÍAZ-CASTELAZO† and C. CASTILLO-GUEVARA†

\*Departamento de Zoologia, Universidade Estadual de Campinas, C.P. 6109, 13083–970 Campinas SP, Brasil and †Departamento de Ecología Vegetal, Instituto de Ecología, A.C., Apdo. 63, Xalapa, Veracruz 91000, México

## Summary

1. This study examines the anti-herbivore effect of ants visiting the extrafloral nectaries (EFNs) of *Opuntia stricta* (Cactaceae) and its possible influence on the plant's reproductive output in Mexican coastal sand dunes. *Opuntia*'s EFNs are located in the areoles of the developing tissue of emerging cladodes and flower buds.
2. Ants visited the EFNs of *O. stricta* on a round-the-clock basis. The associated ant assemblage was formed by nine species distributed in four subfamilies, and the species composition of the principal ant visitors changed markedly from day to night period.
3. Cladodes of control (ants present) and treatment (ants excluded) plants of *Opuntia* were equally infested by sucking bugs and mining dipterans. Damage to buds by a pyralid moth, however, was significantly higher on treatment than on control plants. Ant visitation to *Opuntia*'s EFNs translated into a 50% increase in the plant's reproductive output, as expressed by the number of fruits produced by experimental control and treatment branches. Moreover, fruit production by ant-visited branches was positively and significantly associated with the mean monthly rate of ant visitation to EFNs.
4. This is the first demonstration of ant protection leading to increased fruit set in the Cactaceae under natural conditions. Although the consequences of damage by sucking and mining insects remain unclear for *Opuntia*, the results show how the association of EFNs with vulnerable reproductive plant organs can result in a direct ant-derived benefit to plant fitness.

*Key-words:* Ant foraging schedule, ant–plant mutualism, ant protection, cactus, herbivory, reproductive output

*Functional Ecology* (1999) **13**, 623–631

## Introduction

Insect herbivores may eat virtually all types of plant tissue and herbivore damage may occur at any stage of a plant's life cycle (Crawley 1983). However, because herbivores consume both vegetative and reproductive tissue, the impact of herbivory on plant fitness may depend largely on the type of tissue being consumed (Marquis 1992). A number of plant characteristics are hypothesized to have evolved as responses to selective pressure exerted by herbivores, including structural, chemical, physiological and life-history traits (reviewed by Marquis 1992). Mutualistic associations with ants constitute one such defence strategy, and hundreds of plant species produce domatia (structures that house ant colonies) and/or food rewards (food bodies, extrafloral nectar) to attract ants which in turn

provide the plant with some protection against herbivores (see Beattie 1985; Davidson & McKey 1993; Whitman 1994).

Extrafloral nectaries (EFNs) are nectar-secreting organs not directly involved in pollination which are found on virtually all above-ground plant parts (Bentley 1977a; Elias 1983; Koptur 1992; Oliveira & Pie 1998). Plants bearing EFNs are widely distributed around the world, and available evidence supports the general contention that they are more common in tropical than in temperate environments (Bentley 1977a; Oliveira & Leitão-Filho 1987; Pemberton 1988; Oliveira & Oliveira-Filho 1991; Schupp & Feener 1991; Coley & Aide 1991). Although EFNs attract a variety of nectar-feeding insects (Koptur 1992), ants are by far the most frequent visitors to

EFN-bearing plants both in temperate and tropical habitats (Oliveira & Brandão 1991; and included references). In the past two decades, a number of experimental field studies have demonstrated that ant visitation to EFNs may increase plant fitness by deterring leaf herbivores (Koptur 1979; Stephenson 1982; Smiley 1985), bud or flower herbivores (Schemske 1980; Rico-Gray & Thien 1989; Oliveira 1997) and seed predators (Inouye & Taylor 1979; Pickett & Clark 1979; Keeler 1981). Some studies, however, have found no apparent benefit to the plant from ant visitation (e.g. O'Dowd & Catchpole 1983; Whalen & MacKay 1988). As stressed by Schemske (1983), ant-plant mutualisms mediated by EFNs are facultative and non-specialized, as indicated by the wide variety of associated ant visitors (see also Thompson 1988; Bronstein 1998). In fact, ant-derived benefits to EFN-bearing plants can be conditioned by factors such as time (Tilman 1978), habitat type (Barton 1986), aggressiveness of ant visitors (Horvitz & Schemske 1984; Oliveira, Silva & Martins 1987; Rico-Gray & Thien 1989), as well as the capacity of herbivores to overcome ant predation (Koptur 1984; Heads & Lawton 1985; Freitas & Oliveira 1996).

The present study examines the anti-herbivore effect of ants visiting the EFNs of *Opuntia stricta* Haw. (Cactaceae) and its possible influence on the plant's reproductive output in Mexican coastal sand dunes. Although Pickett & Clark (1979) showed that herbivore deterrence by visiting ants can increase fruit production by plants of *Opuntia acanthocarpa* in greenhouse conditions, the authors were not able to show such an effect in the species' natural habitat (Sonoran Desert, AZ). In this study we use field observations and controlled ant-exclusion experiments to test the hypothesis of ant protection leading to reproductive benefits in *O. stricta* under natural conditions. Three questions were addressed: (1) which are the ant visitors to the EFNs of *O. stricta*; (2) does ant visitation reduce the infestation levels or damage by the plant's principal herbivores; (3) does ant presence, and ant density on plants, affect the reproductive output (i.e. fruit set) of *O. stricta*?

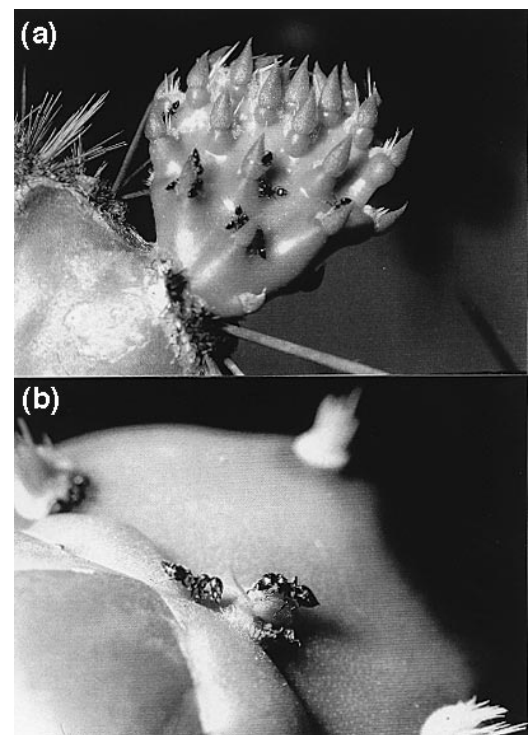
#### THE PLANT, AND THE STUDY SYSTEM

*Opuntia stricta* is a succulent cactus (up to c. 2 m tall) that commonly occurs along the coastal sand dunes of Veracruz, México (Novelo 1978). *Opuntia* flowers can be pollinated by bees and birds (see Grant & Grant 1981), and in México the fruits are consumed by several birds, rodents and other mammals (González-Espinoza & Quintana-Ascencio 1986; Janzen 1986). Ants actively visit the plant both day and night, and gather around the EFNs located in the areoles of the developing tissue of emerging cladodes, as well as in the areoles of developing flower buds (Fig. 1; see also Rico-Gray 1993). *Opuntia*'s extrafloral nectar contains sugars and amino acids at average

concentrations of 11 M and 279 mM, respectively (Pickett & Clark 1979; for *O. acanthocarpa*). *Opuntia stricta* is attacked by four principal herbivores in the sand dunes of Veracruz: (1) *Narnia* sp. (Hemiptera: Coreidae) adults mate on the plant and egg batches (eight to 14) are laid on the spines; nymphs and adults suck plant juice from cladodes and produce typical white rings around punctures (Mann 1969); (2) *Hesperolabops* sp. (Hemiptera: Miridae) egg batches not seen but nymphs and adults suck plant juice from cladodes and punctures are detectable by white dots; (3) mining insects' (Diptera) mining/feeding activity by developing larvae leave easily detectable tunnels within infested cladodes; (4) bud-destroying moth (Lepidoptera: Pyralidae, Phycitinae) eggs are laid on floral buds and developing cladodes, and larval burrowing/feeding activity within the plant organ leaves characteristic external marks (see also Mann 1969).

#### Study site and methods

Field work was carried out from April 1997 to March 1998, at the Centro de Investigaciones Costeras La Mancha located in the state of Veracruz on the coast of the Gulf of México (19° 36' N, 96° 22' W; elevation < 100 m). The climate is warm and sub-humid. Mean annual temperature is 24–26 °C, with total annual precipitation ranging from 1100 to 1500 mm, and a rainy season occurring between June and September. The



**Fig. 1.** (a) Several workers of *Crematogaster brevispinosa* gathering at the areoles of a developing cladode of *Opuntia stricta*, where they actively collect extrafloral nectar. (b) A close-up view of the areoles of developing floral buds with associated *Cr. brevispinosa* workers at active extrafloral nectaries.

dune system at La Mancha extends over 2 km along the coast, and the structure and composition of the vegetation depend on sand movement, protection from wind and salt spray (see Novelo 1978; Moreno-Casasola *et al.* 1982). These factors create a mosaic of vegetation physiognomies within the dune system. Mobile and stabilized dunes in higher areas are inhabited mostly by herbaceous plants and scattered shrubs, whereas the neighbouring thickets located in the lower parts host a scrub of shrubs and trees (further details in Moreno-Casasola *et al.* 1982). All individuals of *O. stricta* used in the present study were located on stabilized dunes covered by a dense herbaceous layer.

#### THE ANT ASSEMBLAGE AT EFNS

The species composition of the ant visitors at the EFNs of *O. stricta* was evaluated on 28–29 April 1997 (sunny day and clear night) through a 24 h census carried out on 10 tagged plants (1.0–1.8 m tall). Monitored cacti had nine to 13 flower buds and were at least 10 m apart from each other. Nectar-gathering ants were censused at 1 h intervals and samplings at each tagged plant consisted of recording the number of workers from different species during a standard period of 20 s. Air temperature (1 m above ground) was recorded at each sampling session during the whole census period. Ant morphospecies are numbered in accordance with the species checklist of La Mancha (P. Rojas, unpublished data).

#### ANT-EXCLUSION EXPERIMENTS, HERBIVORE ACTIVITY AND PLANT REPRODUCTION

The protective role of visiting ants against herbivores of *O. stricta* was tested with ant-exclusion experiments. By early June 1997, before the beginning of the wet season, 19 experimental plant pairs of *O. stricta* (0.6–2.0 m tall) were tagged along nearly 500 m of stabilized dune vegetation. Plants within a pair were approximately the same height, 0.5–20 m apart from each other, and in the same phenological state (no buds, flowers or fruits). Each plant in a given pair was randomly designated by the flip of a coin as a treatment or control plant, and had one branch selected for the monthly records of herbivore and ant activity, as well as fruit production (see below). Experimental branches in a given plant pair had equal numbers of cladodes (one to four). Ants were prevented from climbing on experimental branches of treatment plants by applying to their base a sticky barrier of tree Tanglefoot® (Tanglefoot Co., Grand Rapids, MI, USA). Grass bridges providing aerial access of ants to treated plants were regularly pruned. Ants had free access to control individuals of *O. stricta*.

Herbivore activity on experimental branches of control and treatment plants was measured monthly from June 1997 to March 1998 (no record in

December). Infestation levels by *Narnia* and *Hesperolabops* sucking bugs were determined by counting the number of nymphs and adults present on the branches. Damage by mining dipterans to each cladode of experimental branches was estimated visually prior to ant treatment (June 1997) and after 10 months of ant-exclusion (March 1998). Intensity of miner attack is expressed as the percentage surface area covered by miner tunnels. Four categories were established: none (0%), low ( $\leq 30\%$ ), medium (30–60%), and high ( $\geq 60\%$ ). Infestation by the pyralid moth was evaluated by counting the number of external marks left by caterpillars on cladodes and flower buds of experimental branches. Pyralid damage was measured only in February and March 1998, when bud emission by *O. stricta* and egg-laying activity by the moths were most intense.

The number of ants visiting the EFNs of experimental branches of control plants was counted monthly, simultaneously with the records of herbivore activity (see above). Fruit production by control and treatment branches was determined at the end of *O. stricta*'s fruiting period at La Mancha (March 1998). To account for unknown effects of size among experimental branches, the number of ripe fruits in a given branch was divided by the number of cladodes that formed it.

#### STATISTICAL ANALYSES

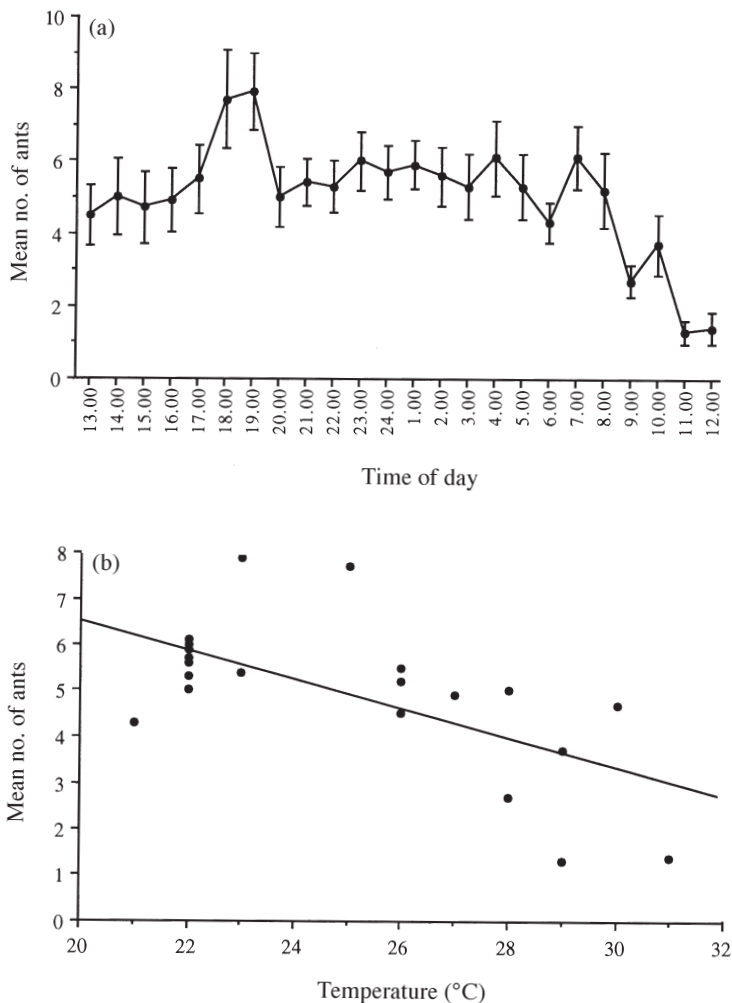
Hemipteran activity (pooled for *Narnia* and *Hesperolabops*) on experimental plant pairs along successive months was analysed by repeated-measures ANOVA after square-root transformation of the data (to normalize the distribution). Damage by mining insects was compared using chi-square tests (Yates correction) on the proportion of experimental cladodes falling under different categories of miner attack before (June 1997) and after 10 months of ant-exclusion (March 1998). Because the data on pyralid damage were not normally distributed, larval external marks to experimental plants pairs were compared using a Mann-Whitney *U*-test. Data on fruit production by control and treatment branches were analysed by a two-tail paired *t*-test after passing the normality assumption.

## Results

#### ANT VISITATION TO *O. STRICTA*'S EFNS

Ants visited the EFNs of *O. stricta* on a round-the-clock basis and at any given sampling period  $9.5 \pm 1.1$  plants were occupied by ants (mean  $\pm$  SD; 10 plants checked at 1 h intervals). Average visitation rate ranged from 1.4 to 7.9 ants per plant, and the activity rhythm of nectarivorous ants was negatively and significantly related with temperature (Fig. 2). The ant assemblage associated with the EFNs of *O. stricta* was formed by

nine species distributed in four subfamilies, as follows: Formicinae, *Camponotus planatus*, *Camponotus abdominalis*, *Camponotus* sp. 10, *Paratrechina longicornis*; Myrmicinae, *Crematogaster brevispinosa*, *Monomorium cyaneum*; Dolichoderinae, *Forelius* aff. *pruinosis*; Pseudomyrmecinae, *Pseudomyrmex* sp. 5 (*pallens* group), *Pseudomyrmex* sp. 7 (*pallidus* group). Although some ant species were seen at EFNs both day and night, the species composition of the principal ant visitors changed markedly from one period to the other (Fig. 3). *Camponotus planatus*, *P. longicornis*, *Cr. brevispinosa* and *Pseudomyrmex* sp. 5 were the most frequent species on plants during daytime samplings, whereas the dominant *C. abdominalis* was by far the most frequent visitor at night, followed by the less common and exclusively nocturnal *Camponotus* sp. 10 (Fig. 3a). The contrasting activity rhythms of the two most dominant ants at EFNs, *C. planatus* and *C. abdominalis*, are shown in Fig. 3b for the entire census period. No aggressive interaction between EFN-gathering ants was ever observed on *O. stricta*.



**Fig. 2.** (a) Daily activity of ants at the extrafloral nectaries (EFNs) of *O. stricta* in the coastal sand dunes of Veracruz, México. Data are means  $\pm$  1 SE ( $n = 10$  plants). (b) Relationship between temperature and ant activity at *O. stricta*'s EFNs (regression equation:  $y = 12.8 - 0.316x$ , Adj.  $r^2 = 0.383$ ,  $P < 0.001$ ).

#### ANT-EXCLUSION EXPERIMENTS AND HERBIVORE INFESTATION LEVELS

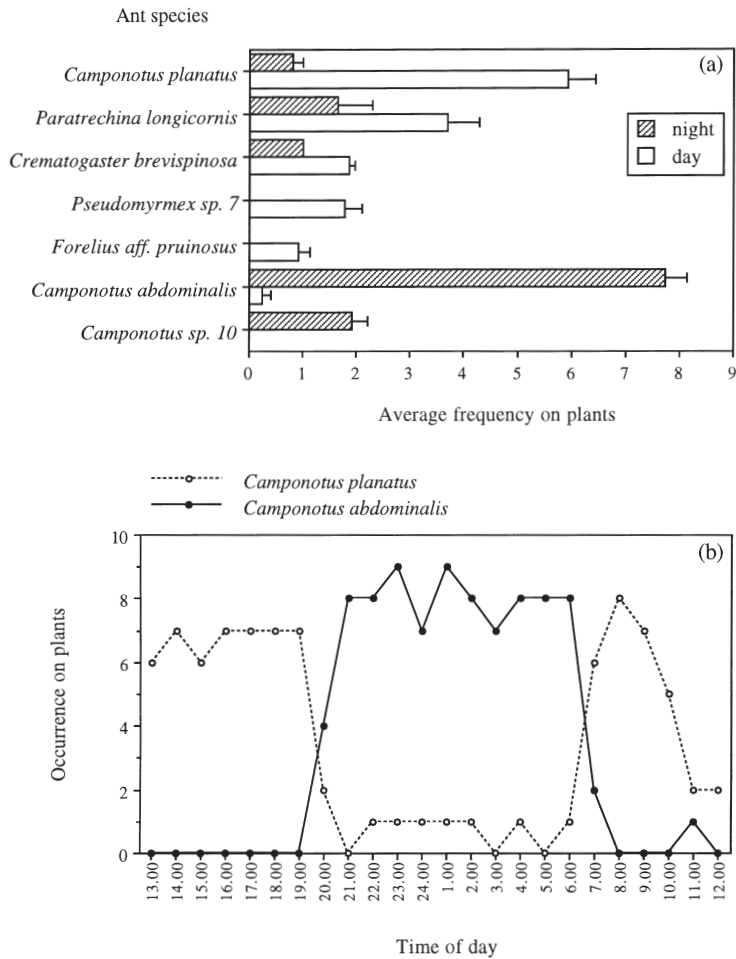
Control and ant-excluded plants of *O. stricta* were equally infested by *Narnia* and *Hesperolobops* sucking bugs throughout the field experiment (Fig. 4; repeated-measures ANOVA,  $F_{1,36} = 0.067$ ,  $P = 0.797$ ). Visiting ants were never observed attacking the hemipterans and seemed to ignore them on control plants. Similarly, ant visitation to EFNs had no effect on the infestation levels by mining insects; the proportion of infested cladodes was similar for either experimental plant class, both before ( $\chi^2 = 1.279$ ,  $df = 2$ ,  $P = 0.734$ ) and after ant treatment ( $\chi^2 = 0.973$ ,  $df = 2$ ,  $P = 0.807$ ) (Fig. 5). On the other hand, damage by the pyralid moth was significantly higher on treatment than on control plants. External marks left by caterpillar burrowing activity within cladodes and flower buds were more numerous on ant-excluded than on ant-visited plants (mean  $\pm$  SD =  $0.84 \pm 1.92$  vs  $0.10 \pm 0.3$ , respectively; Mann-Whitney  $U$ -test,  $U = 893.0$ ,  $P < 0.001$ ,  $N = 19$ ).

#### ANT EFFECTS ON PLANT REPRODUCTION

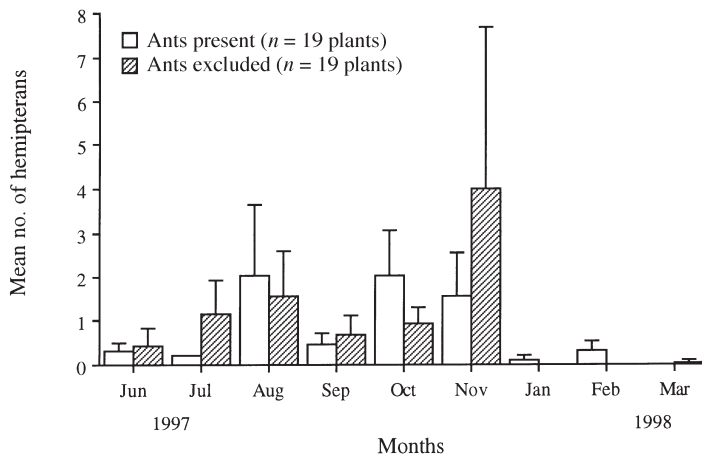
Ant visitation to *O. stricta*'s EFNs translated into a 50% increase in the plant's reproductive output: the mean number ( $\pm$  1 SD) of fruits per cladode produced by experimental branches was  $3.62 \pm 1.80$  for ant-visited plants vs  $2.40 \pm 0.34$  for ant-excluded ones (paired  $t$ -test,  $t = 2.564$ ,  $df = 18$ ,  $P = 0.0195$ ). Moreover, fruit production by control branches was positively and significantly associated with the mean rate of ant visitation to EFNs along the successive months of the study (Fig. 6).

#### Discussion

Extrafloral nectar is regarded as a general ant reward capable of attracting to the plant a wide diversity of ants (Carroll & Janzen 1973; Beattie 1985). Surprisingly, however, most studies dealing with ant-plant systems mediated by EFNs have focused on the role of 'key' ant species, and relatively few investigate the associated ant assemblage through daytime and night censuses (see Oliveira & Brandão 1991; and included references). In general the emerging pattern from 24 h censuses at EFNs reveals a clear turnover of ant species through time, with quite distinct diurnal and nocturnal assemblages of visiting ants (Bentley 1977b; Blom & Clark 1980; Oliveira & Brandão 1991; Oliveira, Klitzke & Vieira 1995). In some cases, however, little daily turnover in ant species composition has been reported, with nectar sources being continually visited by one or a few dominant ant species (e.g. O'Dowd 1979; Beckmann & Stucky 1981). The segregation of daily foraging schedules among sympatric ant species is especially common in tropical areas and generally results from distinct humidity and



**Fig. 3.** (a) Daily turnover in species composition of the principal ant visitors to the extrafloral nectaries of *O. stricta* in Veracruz, México. Frequency is expressed as the mean number of plants ( $\pm 1$  SE) on which the species was recorded during diurnal (07.00–19.00 h) and nocturnal (20.00–06.00 h) samplings. Ten plants were checked at 1 h intervals. The ants *Monomorium cyaneum* and *Pseudomyrmex* sp. 5 (*pallens* group) were each recorded just once during daytime and are not represented in the graph. (b) Daily species replacement by the two dominant ants at the extrafloral nectaries of *O. stricta* in Veracruz. Occurrence is expressed as the number of plants on which the species was recorded at hourly samplings.

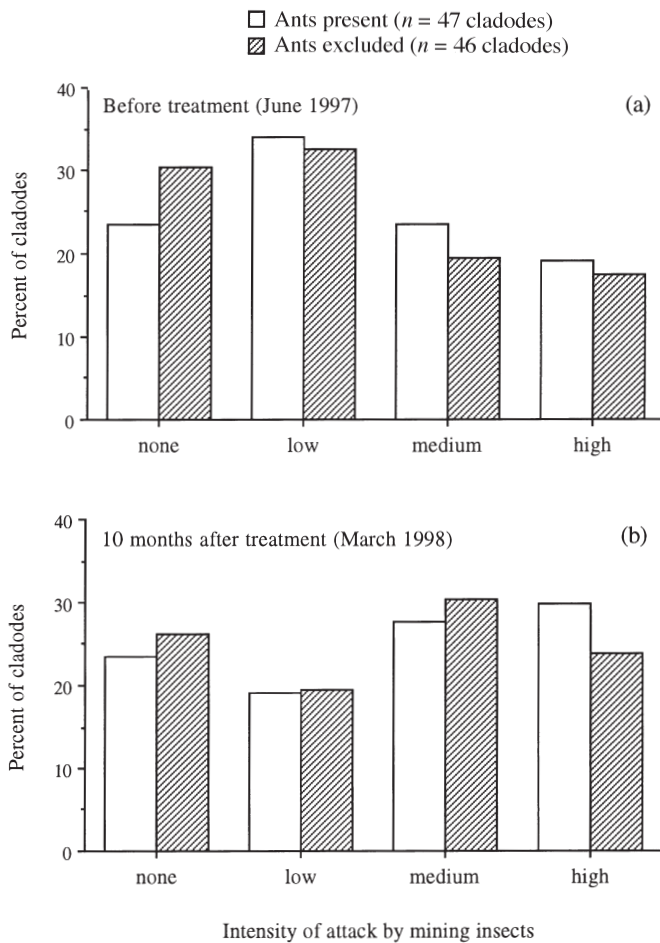


**Fig. 4.** Infestation levels by *Narnia* and *Hesperolabops* sucking bugs (nymphs and adults) on experimental plant pairs of *O. stricta* through time. Herbivore activity is not affected by ant treatment (repeated-measures ANOVA,  $F_{1,36} = 0.067$ ,  $P = 0.797$ ). Data are means  $\pm 1$  SE.

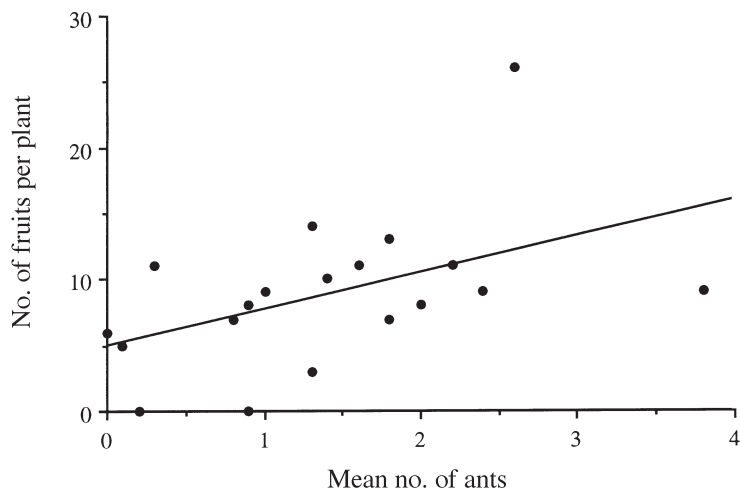
temperature ranges tolerated by different species (Levings 1983; Hölldobler & Wilson 1990; Cerdá, Retana & Cros 1998). The round-the-clock census of the ant assemblage at EFNs of *O. stricta* revealed a clear species turnover within the 24 h period and a significant relationship between ant activity and temperature. The pattern here reported for coastal sand dunes has also been registered for nectarivorous ant assemblages living in other similarly exposed environments such as deserts (Blom & Clark 1980) and savannahs (Oliveira & Brandão 1991; Oliveira *et al.* 1995), where ant foraging schedules are apparently more closely linked with marked daily oscillations in temperature (see Bernstein 1979; Moreno-Casasola 1982). Although such differences in daily foraging rhythms may be proximately adjusted by the species' physiological properties, they can allow temporal partitioning of persistent liquid resources such as extrafloral nectar (Oliveira & Brandão 1991), and have also been reported for ant assemblages harvesting homopteran honeydew (Del-Claro & Oliveira 1999) and lepidopteran secretions (DeVries 1991).

Two species of *Camponotus*, *C. planatus* and *C. abdominalis*, comprised most of the ant observations at *O. stricta*'s EFNs, followed by *P. longicornis* and *Cr. brevispinosa*. These four ant species, but most especially *C. planatus* and *Cr. brevispinosa*, are very common in Mexican coastal sand dunes, where they forage for several types of plant-derived food resources, including floral and extrafloral nectar from many plant species, as well as homopteran and lepidopteran secretions (Rico-Gray 1989, 1993; Rico-Gray & Thien 1989; Rico-Gray & Castro 1996; Rico-Gray *et al.* 1998). These data confirm the dominance of *Camponotus* and *Crematogaster* ants on tropical foliage (Wilson 1987), and on plants bearing EFNs worldwide (see Oliveira & Brandão 1991; and included references).

Although ant visitation to EFNs of Cactaceae has long been reported in the botanical literature (Lloyd 1908), only more recently has this interaction been studied in greater detail (Pickett & Clark 1979; Blom & Clark 1980; Ruffner & Clark 1986; Pemberton 1988). Cacti extrafloral nectar may represent an extremely rich food source for ants because it contains not only sugars (fructose, glucose and sucrose) but also several amino acids which are thought to be essential for insect nutrition (Pickett & Clark 1979; Ruffner & Clark 1986). The current study involving *O. stricta*, visiting ants and insect herbivores extends earlier reports by other authors by providing a more precise picture of the species interactions within the system through ant-exclusion experiments. The field observations and experiments in the coastal sand dunes of La Mancha demonstrated that round-the-clock visitation by ants to *O. stricta*'s EFNs can reduce herbivore damage to plant reproductive structures and increase by 50% the plant's fruit set under natural conditions. Moreover, the level of ant visita-



**Fig. 5.** Infestation levels by mining dipterans on experimental cladodes of *O. stricta* prior to ant treatment (June 1997), and after 10 months of ant-exclusion (March 1998). Intensity of miner attack is expressed as the percentage surface area covered by miner tunnels: none (0%), low ( $\leq 30\%$ ), medium (30–60%), and high ( $\geq 60\%$ ). Attack by mining insects was similar for either plant class before and after ant treatment ( $\chi^2$  tests,  $P > 0.70$ ).



**Fig. 6.** Relationship between ant visitation rates to extrafloral nectaries and fruit production by control branches of *O. stricta* ( $n = 19$  plants). Ant visitation is expressed as the mean number of ants recorded through monthly samplings, and fruit production is the number of fruits produced by experimental branches (regression equation:  $y = 4.94 + 2.77x$ , Adj.  $r^2 = 0.180$ ,  $P = 0.04$ ).

tion to the plants was also shown to positively affect fruit production. Although similar results were obtained in other EFN-mediated ant–plant systems (see Bentley 1977a,b; Inouye & Taylor 1979; Koptur 1979; Schemske 1980; Keeler 1981; Rico-Gray & Thien 1989; Del-Claro, Berto & Réu 1996) this is the first demonstration of ant protection leading to increased fruit set in the Cactaceae under natural conditions.

The results from the ant-exclusion experiments further corroborated other studies showing that the deterring capacities of visiting ants may not be equally effective against all types of herbivores infesting a plant and this may be owing to several factors. First, some herbivores may possess mechanisms to overcome ant predation and can feed on the plant despite the ants (e.g. Horvitz & Schemske 1984; Koptur 1984; Heads & Lawton 1985; Freitas & Oliveira 1992, 1996). Second, variation in the abundance, aggressiveness or size of ant visitors can affect their protective abilities against a particular herbivore species (Bentley 1977b; Inouye & Taylor 1979; Horvitz & Schemske 1984; Oliveira *et al.* 1987; Rico-Gray & Thien 1989). Third, herbivore vulnerability to ant predation can depend strongly on the type of plant tissue being consumed relative to the spatial and temporal distribution of extrafloral nectar within the plant crown (Bentley 1977a,b; Tilman 1978; Koptur & Lawton 1988; Oliveira 1997).

Although Pickett & Clark (1979) reported that *Crematogaster opuntiae* ants attack/kill captive *Chelinidea* sp. sucking bugs on *O. acanthocarpa*, the observations of Blom & Clark (1980) on *Ferocactus gracilis* confirm ours on *O. stricta* in that visiting ants (including *Crematogaster*) do not disturb *Narnia* or *Hesperolabops* sucking bugs on the plants. Foraging ants are known to attack and dislodge bud/fruit-feeding hemipterans on plants bearing EFNs near reproductive structures (Oliveira 1997). Although *Narnia* bugs are reported to feed on buds and fruits (Mann 1969), their feeding marks on *O. stricta* were mostly confined to well-developed cladodes (as well as those of *Hesperolabops*), where they presumably are less vulnerable to interference from the ants gathering at the EFNs of apical developing buds and cladodes (Fig. 1).

Insect larvae feeding on internal plant tissue, such as miners and borers, may live within their ‘shelters’ relatively safe from ants (Heads & Lawton 1985; Costa, Oliveira-Filho & Oliveira 1992). However, internal plant feeders can be negatively affected by ants if ant activity at a plant’s food source is likely to disturb ovipositing adults, as repeatedly shown for plants bearing EFNs on reproductive organs (e.g. Bentley 1977b; Inouye & Taylor 1979; Schemske 1980; Horvitz & Schemske 1984; Oliveira 1997). Results from the ant-exclusion experiments with *O. stricta* are clear-cut with respect to the two types of internal feeders tested: damage to cladodes by mining

dipterans was not affected by ant treatment, whereas the damage by pyralid burrowing larvae to flower buds was significantly higher on ant-excluded plants. The observed pattern most likely results from the more intense ant activity at the EFNs of floral buds than on cladodes. While ovipositing insects are known to be negatively affected by ant activity near a nectar source (Inouye & Taylor 1979; Freitas & Oliveira 1996), the spatial distribution of extrafloral nectar in *Opuntia* probably makes cladode mining dipterans less vulnerable to ants during oviposition. Similar results with ants vs internal feeders on another nectary plant are reported by Oliveira (1997).

In conclusion, the present study with *O. stricta* has shown that round-the-clock ant activity at EFNs can reduce damage to reproductive structures and increase the plant's reproductive output. Differential deterrence by ants toward the insect herbivores of *Opuntia* may result from several factors, but the spatial distribution of extrafloral nectar within the plant is possibly the most likely one. The fitness consequences of herbivore damage to vegetative tissue are frequently less clear and more difficult to measure than direct damage to plant reproductive structures (Marquis 1992). Although the consequences of damage by sucking and mining insects remain unclear for *Opuntia*, this study clearly shows how the association of EFNs with vulnerable reproductive plant organs can result in a direct ant-derived benefit to plant fitness.

### Acknowledgements

We are grateful to M. L. Martinez and J. López-Portillo for logistic support and discussions, and to M. Palacios-Rios for helpful suggestions on the manuscript. The final version was improved by suggestions from D. H. Janzen and an anonymous referee. Insects were identified by P. Rojas (ants), V. Meléndez (hemipterans), S. Passoa (lepidopterans) and A. Bonet (dipterans). Financial support to P.S.O. was provided by a research grant from the Conselho Nacional de Desenvolvimento Científico e Tecnológico, and by travel grants from the Fundação de Amparo à Pesquisa do Estado de São Paulo (no. 98/03833-1), and the Instituto de Ecología, A.C. (no. 902-16). V.R.-G., C.D.-C. and C.C.-G. were supported by the Instituto de Ecología, A.C. (no. 902-16).

### References

- Barton, A.M. (1986) Spatial variation in the effect of ants on an extrafloral nectary plant. *Ecology* **67**, 495–504.
- Beattie, A.J. (1985) *The Evolutionary Ecology Ant-Plant Mutualisms*. Cambridge University Press, Cambridge.
- Beckmann, R.L. & Stucky, J.M. (1981) Extrafloral nectaries and plant guarding in *Ipomoea pandurata* (L.) G.F.W. Mey. (Convolvulaceae). *American Journal of Botany* **68**, 72–79.
- Bentley, B.L. (1977a) Extrafloral nectaries and protection by pugnacious bodyguards. *Annual Review of Ecology and Systematics* **8**, 407–428.
- Bentley, B.L. (1977b) The protective function of ants visiting the extrafloral nectaries of *Bixa orellana* (Bixaceae). *Journal of Ecology* **65**, 27–38.
- Bernstein, R.A. (1979) Schedules of foraging activity in species of ants. *Journal of Animal Ecology* **48**, 921–930.
- Blom, P.E. & Clark, W.H. (1980) Observations of ants (Hymenoptera: Formicidae) visiting extrafloral nectaries of the barrel cactus, *Ferocactus gracilis* Gates (Cactaceae), in Baja California, Mexico. *Southwestern Naturalist* **25**, 181–196.
- Bronstein, J.L. (1998) The contribution of ant-plant protection studies to our understanding of mutualism. *Biotropica* **30**, 150–161.
- Carroll, C.R. & Janzen, D.H. (1973) Ecology of foraging by ants. *Annual Review of Ecology and Systematics* **4**, 231–257.
- Cerdá, X., Retana, J. & Cros, S. (1998) Critical thermal limits in Mediterranean ant species: trade-off between mortality risk and foraging performance. *Functional Ecology* **12**, 45–55.
- Coley, P.D. & Aide, T.M. (1991) Comparison of herbivory and plant defenses in temperate and tropical broad-leaved forests. *Plant-Animal Interactions: Evolutionary Ecology in Tropical and Temperate Regions* (eds P. W. Price, T. M. Lewinsohn, G. W. Fernandes & W. W. Benson), pp. 25–49. John Wiley & Sons, New York.
- Costa, F.M.C.B., Oliveira-Filho, A.T. & Oliveira, P.S. (1992) The role of extrafloral nectaries in *Qualea grandiflora* (Vochysiaceae) in limiting herbivory: an experiment of ant protection in cerrado vegetation. *Ecological Entomology* **17**, 362–365.
- Crawley, M.J. (1983) *Herbivory: the Dynamics of Animal-Plant Interactions*. University of California Press, Berkeley.
- Davidson, D.W. & McKey, D. (1993) The evolutionary ecology of symbiotic ant-plant relationships. *Journal of Hymenoptera Research* **2**, 13–83.
- Del-Claro, K. & Oliveira, P.S. (1999) Ant-Homoptera interactions in a neotropical savanna: the honeydew-producing treehopper *Guayaquila xiphias* (Membracidae) and its associated ant fauna on *Didymopanax vinosum* (Araliaceae). *Biotropica* **31**, 135–144.
- Del-Claro, K., Berto, V. & Réu, W. (1996) Effect of herbivore deterrence by ants on the fruit set of an extrafloral nectary plant, *Qualea multiflora* (Vochysiaceae). *Journal of Tropical Ecology* **12**, 887–892.
- DeVries, P.J. (1991) Evolutionary and ecological patterns in myrmecophilous riodinid butterflies. *Ant-Plant Interactions* (eds D. F. Cutler & C. R. Huxley), pp. 143–156. Oxford University Press, Oxford.
- Elias, T.S. (1983) Extrafloral nectaries: their structure and functions. *The Biology of Nectaries* (eds B. Bentley & T. S. Elias), pp. 174–203. Columbia University Press, New York.
- Freitas, A.V.L. & Oliveira, P.S. (1992) Biology and behavior of *Eunica bechina* (Lepidoptera: Nymphalidae) with special reference to larval defense against ant predation. *Journal of Research on the Lepidoptera* **31**, 1–11.
- Freitas, A.V.L. & Oliveira, P.S. (1996) Ants as selective agents on herbivore biology: effects on the behaviour of a non-myrmecophilous butterfly. *Journal of Animal Ecology* **65**, 205–210.
- González-Espinoza, M. & Quintana-Ascencio, P.F. (1986) Seed predation and dispersal in a dominant desert plant. *Opuntia*, ants, birds, and mammals. *Frugivores and Seed Dispersal* (eds A. Estrada & T. H. Fleming.), pp. 273–284. Dr W. Junk, Publishers, Dordrecht.
- Grant, B.R. & Grant, P.R. (1981) Exploitation of *Opuntia* cactus by birds on the Galápagos. *Oecologia* **49**, 179–187.

- Heads, P.A. & Lawton, J.H. (1985) Bracken, ants and extrafloral nectaries. III. How insect herbivores avoid predation. *Ecological Entomology* **10**, 29–42.
- Hölldobler, B. & Wilson, E.O. (1990) *The Ants*. The Belknap Press of Harvard University Press, Cambridge, MA.
- Horvitz, C.C. & Schemske, D.W. (1984) Effects of ants and ant-tended herbivores on seed production of a neotropical herb. *Ecology* **65**, 1369–1378.
- Inouye, D.W. & Taylor, O.R. (1979) A temperate region plant–ant–seed predator system: consequences of extrafloral nectar secretion by *Helianthella quinquinervis*. *Ecology* **60**, 1–7.
- Janzen, D.H. (1986) Chihuahuan desert nopaleras: defaunated big mammal vegetation. *Annual Review of Ecology and Systematics* **17**, 595–636.
- Keeler, K.H. (1981) Function of *Mentzelia nuda* (Loasaceae) postfloral nectaries in seed defense. *American Journal of Botany* **68**, 295–299.
- Koptur, S. (1979) Facultative mutualism between weedy vetches bearing extrafloral nectaries and weedy ants in California. *American Journal of Botany* **66**, 1016–1020.
- Koptur, S. (1984) Experimental evidence for defense of *Inga* (Mimosoideae) saplings by ants. *Ecology* **65**, 1787–1793.
- Koptur, S. (1992) Extrafloral nectary-mediated interactions between insects and plants. *Insect–Plant Interactions*, vol. 4 (ed. E. Bernays), pp. 81–129. CRC Press, Boca Raton.
- Koptur, S. & Lawton, J.H. (1988) Interactions among vetches bearing extrafloral nectaries, their biotic protective agents, and herbivores. *Ecology* **69**, 278–283.
- Levings, S.C. (1983) Seasonal, annual, and among-site variation in the ground ant community of a deciduous tropical forest: some causes of patchy species distributions. *Ecological Monographs* **53**, 435–455.
- Lloyd, F.E. (1908) Extra-floral nectaries in the cacti. *Plant World* **11**, 138–140.
- Mann, J. (1969) *Cactus-feeding insects and mites*. Smithsonian Institution Bulletin no. 256. Smithsonian Institution, Washington, DC.
- Marquis, R.J. (1992) Selective impact of herbivores. *Plant Resistance to Herbivores and Pathogens: Ecology, Evolution, and Genetics* (eds R. S. Fritz & E. L. Simms), pp. 301–325. The University of Chicago Press, Chicago.
- Moreno-Casasola, P. (1982) Ecología de la vegetación de dunas costeras: Factores físicos. *Biotica* **7**, 577–602.
- Moreno-Casasola, P., van der Maarel, E., Castillo, S., Huesca, M.L. & Pisanty, I. (1982) Ecología de la vegetación de dunas costeras: Estructura y composición. *Biotica* **7**, 491–526.
- Novelo, A. (1978) La vegetación de la Estación Biológica El Morro de la Mancha, Veracruz. *Biotica* **3**, 9–23.
- O'Dowd, D.J. (1979) Foliar nectar production and ant activity on a neotropical herb, *Ochroma pyramidale*. *Oecologia* **43**, 233–248.
- O'Dowd, D.J. & Catchpole, E.A. (1983) Ants and extrafloral nectaries: no evidence for plant protection in *Helichrysum* spp.–ant interactions. *Oecologia* **59**, 191–200.
- Oliveira, P.S. (1997) The ecological function of extrafloral nectaries: herbivore deterrence by visiting ants and reproductive output in *Caryocar brasiliense* (Caryocaraceae). *Functional Ecology* **11**, 323–330.
- Oliveira, P.S. & Brandão, C.R.F. (1991) The ant community associated with extrafloral nectaries in Brazilian cerrados. *Ant–Plant Interactions* (eds D. F. Cutler & C. R. Huxley), pp. 198–212. Oxford University Press, Oxford.
- Oliveira, P.S. & Leitão-Filho, H.F. (1987) Extrafloral nectaries: their taxonomic distribution and abundance in the woody flora of cerrado vegetation in Southeast Brazil. *Biotropica* **19**, 140–148.
- Oliveira, P.S. & Oliveira-Filho, A.T. (1991) Distribution of extrafloral nectaries in the woody flora of tropical communities in Western Brazil. *Plant–Animal Interactions: Evolutionary Ecology in Tropical and Temperate Regions* (eds P. W. Price, T. M. Lewinsohn, G. W. Fernandes & W. W. Benson), pp. 163–175. John Wiley & Sons, New York.
- Oliveira, P.S. & Pie, M.R. (1998) Interaction between ants and plants bearing extrafloral nectaries in cerrado vegetation. *Anais Da Sociedade Entomológica Do Brasil* **27**, 161–176.
- Oliveira, P.S., da Silva, A.F. & Martins, A.B. (1987) Ant foraging on extrafloral nectaries of *Qualea grandiflora* (Vochysiaceae) in cerrado vegetation: ants as potential antiherbivore agents. *Oecologia* **74**, 228–230.
- Oliveira, P.S., Klitzke, C. & Vieira, E. (1995) The ant fauna associated with the extrafloral nectaries of *Ouratea hexasperma* (Ochnaceae) in an area of cerrado vegetation in Central Brazil. *Entomologist's Monthly Magazine* **131**, 77–82.
- Pemberton, R.W. (1988) The abundance of plants bearing extrafloral nectaries in Colorado and Mojave desert communities of southern California. *Madroño* **35**, 238–246.
- Pickett, C.H. & Clark, W.D. (1979) The function of extrafloral nectaries in *Opuntia acanthocarpa* (Cactaceae). *American Journal of Botany* **66**, 618–625.
- Rico-Gray, V. (1989) The importance of floral and circumfloral nectar to ants inhabiting dry tropical lowlands. *Biological Journal of the Linnean Society* **38**, 173–181.
- Rico-Gray, V. (1993) Use of plant-derived food resources by ants in the dry tropical lowlands of Coastal Veracruz, Mexico. *Biotropica* **25**, 301–315.
- Rico-Gray, V. & Castro, G. (1996) Effect of an ant–aphid–plant interaction on the reproductive fitness of *Paullinia fuscescens* (Sapindaceae). *Southwestern Naturalist* **41**, 434–440.
- Rico-Gray, V. & Thien, L.B. (1989) Effect of different ant species on the reproductive fitness of *Schomburgkia tibicinis* (Orchidaceae). *Oecologia* **81**, 487–489.
- Rico-Gray, V., García-Franco, J.G., Palacios-Rios, M., Díaz-Castelazo, C., Parra-Tabla, V. & Navarro, J.A. (1998) Geographical and seasonal variation in the richness of ant–plant interactions in Mexico. *Biotropica* **30**, 190–200.
- Ruffner, G.A. & Clark, W.D. (1986) Extrafloral nectar of *Ferocactus acanthodes* (Cactaceae): composition and its importance to ants. *American Journal of Botany* **73**, 185–189.
- Schemske, D.W. (1980) The evolutionary significance of extrafloral nectar production by *Costus woodsonii* (Zingiberaceae): an experimental analysis of ant protection. *Journal of Ecology* **68**, 959–967.
- Schemske, D.W. (1983) Limits to specialization and coevolution in plant–animal mutualisms. *Coevolution* (ed. M. H. Nitecki), pp. 67–109. University of Chicago Press, Chicago.
- Schupp, E.W. & Feener, D.H. (1991) Phylogeny, lifeform, and habitat dependence of ant-defended plants in a Panamanian forest. *Ant–Plant Interactions* (eds D. F. Cutler & C. R. Huxley), pp. 175–197. Oxford University Press, Oxford.
- Smiley, J.T. (1985) *Heliconius* caterpillars mortality during establishment on plants with and without attending ants. *Ecology* **66**, 845–849.
- Stephenson, A.G. (1982) The role of the extrafloral nectaries of *Catalpa speciosa* in limiting herbivory and increasing fruit production. *Ecology* **63**, 663–669.
- Thompson, J.N. (1988) Variation in interspecific interactions. *Annual Review of Ecology and Systematics* **19**, 65–87.
- Tilman, D. (1978) Cherries, ants and tent caterpillars: timing of nectar production in relation to susceptibility of caterpillars to ant predation. *Ecology* **59**, 686–692.



- Whalen, M.A. & Mackay, D.A. (1988) Patterns of ant and herbivore activity on five understory Euphorbiaceous saplings in submontane Papua New Guinea. *Biotropica* **20**, 294–230.
- Whitman, D. (1994) Plant bodyguards: mutualistic interactions between plants and the third trophic level. *Functional Dynamics of Phytophagous Insects* (ed. T. N. Ananthakrishnan), pp. 207–248. Oxford and IBH

Publishing Co, PVT. Ltd, New Delhi, India.

- Wilson, E.O. (1987) The arboreal ant fauna of Peruvian Amazon forests: a first assessment. *Biotropica* **19**, 245–282.

*Received 29 September 1998; revised 28 January 1999;  
accepted 16 February 1999*