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Interactions among nectar robbing, floral herbivory, and ant protection in *Linaria vulgaris*

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Nectar robbers are often assumed to be plant antagonists; however, empirical data show that the impacts of these animals range from negative to positive depending on the system and ecological conditions. We experimentally evaluated the combined effects of nectar robbing and ant visitation on three indices of reproductive fitness in *Linaria vulgaris* in the Colorado Rocky Mountains, via indirect effects on flower- and seed-eating beetles (*Brachypterolus pulicarius* and *Gymnaetron antirrhinni*). Nectar robbing *Bombus occidentalis* leave holes in the nectar spurs, effectively creating "extra-floral nectaries" that attract ants. In a paired-plant experiment, ants were significantly more abundant on robbed than on unrobbed plants. Manipulation of ant access and nectar robbing showed that ant exclusion increased beetle attack and decreased female fitness. There was a significant ant-by-robbing interaction on flower damage. Patterns in the other two indices were suggestive of ant-by-robbing interactions, but these were not statistically significant. We also found correlations between spider occupancy on some plants and the mean number of ants (marginally negative) or beetles (significantly positive). Although the effect we report in this study may be highly dependent on spatial and temporal distributions of several interacting species, we discuss its potential role in mitigating the costs of floral parasitism, and its importance to the study of nectar robbing in general.

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Researchers of plant–animal interactions have typically focused on pairwise associations in a single kind of relationship (e.g. pollination, herbivory, seed-dispersal), despite the actual complexity of most systems, which include several kinds of interaction at once. More recently, a growing number of papers have explicitly recognized that an organism's fitness is the net result of all its interactions with other organisms (Herrera 2000, Mothershead and Marquis 2000, Bacher and Friedli 2002, Ehrlén 2002, Herrera et al. 2002, Bronstein et al. 2003, Stanton 2003, Cariveau et al. 2004). These studies show that conclusions from traditional two-species studies can change when the direct and indirect influences of other species are considered (Price et al. 1986, Yodzis 1988, Strauss 1991, Wootton 1994, 2002, Menge 1995, Krupnick et al. 1999, Herrera 2000, Ehrlén 2002, Herrera et al. 2002, Bronstein et al. 2003).

Pollination biologists have tended to focus on the direct mutualistic relationship between flowers and the animals that visit them for pollen and nectar. Recently, however, this view of pollination relationships has expanded to include the influence of other species on animal-mediated plant reproduction. Newer studies highlight the shortcomings of considering pollination separately from other types of interaction.

Nectar robbers, because their fitness effects on plants are generally mediated through changes in the behaviour

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of a third species (the pollinator), have typically been studied in the context of three-way interactions (robber–pollinator–plant; Roubik 1982, Roubik et al. 1985, Irwin and Brody 1999, 2000, Navarro 2000, 2001, Maloof 2001). Nectar robbers bite holes into flowers to obtain nectar, usually without effecting pollination. Although they are typically viewed as detrimental to the plants they attack (but see Zimmerman and Cook 1985, Navarro 2000, Maloof 2001), and to the pollinators that they compete with (Mainero and del Rio 1985, Irwin and Brody 1998), nectar robbers may also indirectly benefit other animals (including corruptible pollinators); the holes they bite make nectar more accessible to others, who become secondary robbers (Inouye 1980). For example, Roubik et al. (1985) found that some avian visitors to *Quassia amara* were limited to flowers that had previously been robbed. In *Linaria vulgaris* in the United Kingdom, some short-tongued bumblebees collect nectar only through the holes left by the hole-biting *Bombus terrestris* (Stout et al. 2000). Intuition suggests that these additional visitors will have further negative effects on plant fitness, either through pollinator avoidance of nectar-depleted flowers, or through defection of legitimate pollinators to secondary robbing.

Some secondarily robbing visitors, however, may offer benefits to the plants that offset the costs of robbing. In 2002, we noticed ants feeding on *Linaria vulgaris* nectar from holes created by nectar-robbing bumblebees. This observation led us to hypothesize that nectar robbing might have positive effects on *L. vulgaris* fitness, if ants defended it from its antagonists. Such defence is a reasonable expectation. Ant–plant protection mutualisms are widespread, often mediated by extrafloral nectar (reviewed by Agrawal and Rutter 1998, Bronstein 1998). This evolutionary pattern indicates that a plant may indeed benefit by providing small amounts of sugar to ants. Thus, nectar made available through holes made by robbers, might recruit ants as plant protectors, just as extrafloral nectar does in other systems.

Not all ant–plant associations are beneficial for both partners; in fact, ants adversely affect some plants by stealing nectar or damaging flowers (Galen 1983, 1999, Norment 1988), by eating plant tissues, or by protecting herbivores (Huxley 1991, Beattie and Hughes 2002). In some cases, specialized plant traits such as extrafloral nectaries or food bodies strongly suggest that ants have a net beneficial effect on the plants. In the absence of such traits, it is difficult to predict how ants will affect the plants they visit.

A thorough knowledge of the natural history of complex systems is required to make relevant predictions about indirect effects (Menge 1995, Raimondi et al. 2000). In this study, we observed the interactions among nectar robbing, ants, flower- and seed-eating beetles (hereafter called "herbivores"), and *L. vulgaris*, a plant without extrafloral nectaries or other ant-rewarding traits. We measured the effects of ant recruitment to robbed plants on several reproductive fitness components in *L. vulgaris*. We hypothesized that nectar robbers can act as interaction modifiers (sensu Wootton 1994) that indirectly change the behaviour of ants and consequently, the abundance or impact of herbivores. By depositing droplets of sugar solution on experimental plants, Bentley (1976) demonstrated opportunistic ant protection of plants that bear no ant-rewarding traits. Based on her results and our observations of ants on robbed plants, we posed the following questions: (1) do ants recruit to robbed flowers to obtain nectar? And (2) do plants exposed to ants suffer less from attacks by antagonistic beetles? We predicted that robbed plants exposed to ants benefit from reductions in herbivory.

Methods

Study site and organisms

The study was performed at the Rocky Mountain Biological Laboratory (RMBL) at Gothic, Colorado (106°59′15″N; 38°57′30″W; 2900 W; 2900 m), between July 7 and 24, 2003. The area comprises open alpine meadow, aspen woodland and spruce forest, and human habitation. Weather during the entire experimental period was remarkably consistent: no rain, and clouds, when they occurred, only in the afternoon.

Linaria vulgaris Mill. (Scrophulariaceae) is a long-lived, perennial, clonal herb introduced to eastern North America from Eurasia in the 1700s. It has since become a noxious weed invading pastures and other agricultural lands, especially in western North America (Saner et al. 1995). Ramets bear several racemose inflorescences with numerous yellow flowers (mean ± SD number of open flowers during daily surveys of this population: 9.45 ± 3.64; $n = 155$; range = 1–19) that present nectar in the end of long spurs (mean ± SD in this population: 13.37 ± 3.04 mm; $n = 673$). The flowers' corolla lips are closed and thus limit nectar access to insects that are strong enough to pry them open (generally bumblebees) or, rarely, animals whose mouthparts are long and thin enough to fit between them (hummingbirds and hawkmoths). Bumblebees effect pollination by contacting the sexual organs on the roof of the flower. Other visitors, such as very small bees, beetles and ants, can also fit or force themselves between the corolla lips (D. A. Newman, pers. obs.). *Linaria vulgaris* reproduces clonally by aggressive rhizomatous growth and sexually by prolific seed production.

Three studies have investigated the pollinator-mediated effects of nectar robbers on female fitness in *L. vulgaris* (Stout et al. 2000, Irwin and Maloof 2002, Nepi et al. 2002); none found significant reductions in seed set, presumably because so few legitimate visits are required to fertilize all available ovules (Arnold 1982). At the RMBL, seed set in *L. vulgaris* is not pollination limited, even in heavily robbed patches (Irwin and Maloof 2002, R. E. Irwin, pers. comm.). Attack by two beetle species, *Brachypterolus pulicarius* (L.), a flower-eating nitidulid, and *Gymnaetron antirrhinni* Paykull, a seed-eating weevil, however, is known to strongly affect female fitness (Smith 1959, Harris 1961, McClay 1992, Saner et al. 1995). The two beetles may compete for resources and may displace one another in some cases (Harris 1961). Of the two beetles, *B. pulicarius* is more common at the RMBL (D. A. Newman, pers. obs.). Adults of this species emerge in spring and feed upon young *L. vulgaris* stems; when flowers begin to form, they oviposit in the buds, where larvae subsequently feed on the anthers and ovaries. Older larvae are also known to eat seeds (Harris 1961). The larvae are able to move from flower to flower and can reduce seed set by 75% and seed weight by 60% (McClay 1992). *Gymnaetron antirrhinni* adults emerge in spring and eat young stems, then lay eggs in the ovaries during flowering. In the developing fruit, the larvae feed on seeds, then pupate, eclose, and overwinter as adults. This species can also reduce seed set by more than 50% (Harris 1961).

Experimental set-up

We worked in a 10 × 6 m plot that contained an early blooming patch of *L. vulgaris* plants. Because there were no nectar robbers foraging at the time of the experiment, we simulated hole-biting by piercing the middle of the spur with fine-nosed forceps whose tips had been bent inward. The location and size of the artificial robber holes matched those of real robbers observed in 2002 at the RMBL. Nectar did not leak out of the holes, but could be reached by ants that could fit their mouthparts, heads, or whole bodies through them. To control for the disturbance that piercing may have caused (e.g. frightening ants and beetles), we also snapped the tweezers shut over the spurs of all flowers in the unrobbed treatments, but without making holes. We pierced or sham-pierced all flowers on the experimental plants, including all flower buds that were developed enough to contain nectar.

Ant recruitment to robbed and unrobbed plants

Between July 7 and 12, in an area with high ant activity, we haphazardly chose five pairs of racemes with a large number of open flowers (mean ± SD: 21.16 ± 3.88) and pierced all the flowers on one inflorescence of each pair. Paired ramets most likely belonged to the same genet (we selected pairs that appeared to grow out of the same root system), and were near neighbours (~20 cm). Three times a day (morning, noon, and late afternoon) for a total of 11 sampling episodes, we scored the ant activity on each plant, as follows: number of ants of each type, behaviour (patrolling plant, drinking nectar), and location on the ramet (lower leaves, inflorescence, within or on flowers). To avoid problems associated with the interdependence of ant sightings over subsequent sampling periods, we averaged data for each plant over all sampling episodes.

Effects of ants and nectar robbing on flower and seed predators and seed set

On July 7 where enough racemes approaching flowering could be found, we chose four neighbouring ramets (~20 cm apart) to represent four treatments in a full factorial design. These were: (1) ant-excluded, pierced spurs; (2) ant-excluded, unpierced spurs; (3) ant-access, pierced; and (4) ant-access, unpierced (hereafter, treatment names for all experiments will be capitalized). We pierced or sham-pierced every flower on all experimental ramets. In all, we treated 160 ramets (n = 40). We excluded ants by removing the leaves from each plant's ~15 lower nodes and applying Tanglefoot® to the stem; on plants where ants were allowed, we removed leaves but did not apply Tanglefoot. We also removed any vegetation that touched or could touch the experimental plants to prevent ants from accessing them. The piercing treatment, including the sham piercing of flowers on the unpierced plants, was administered as described above. Ideally, we would have hand-pollinated the flowers to remove any pollinator preference for one treatment over another, but the disturbance that this would have caused for the insect community inhabiting the plants would have greatly affected the results of the study; hand-pollinating *L. vulgaris* closed flowers is invasive enough to cause the beetles to leave the plants (D. A. Newman, pers. obs.). We noticed no differences in either the behaviour or the frequency of bumblebee visitors among treatments.

Between July 8 and 24, we surveyed the experimental plants, once each morning (~09:00 to ~11:30 h) and once each afternoon (~14:00 to ~16:30 h). During these surveys, we pierced or sham-pierced new flowers, counted the number of beetles and ants visible on the plants, and scored their activities. In addition, we noted the presence of other animals on the plants (Results; Animals on *Linaria vulgaris*). Since both the nitidulids and the weevils sometimes hide inside flowers, their abundance on plants was probably underestimated. We also noted any behavioural interactions among ants, beetles, and any other animals.

Effect of beetles and ants on plant fitness indices

Every day, we counted the number of mature flowers, maturing flowers (large buds with developed spurs), and flower buds on every plant. We also collected wilted flowers before abscision, and inspected them for beetle frass and larvae. We collected the mature fruit on August 16, counted them, then removed the seeds to count and weigh them. We recorded the presence of larval and mature beetles in the fruit.

We measured eight variables (Table 1). Most represent components of female fitness, although male fitness may also be inferred from some variables (e.g. damaged or frass-soiled flowers may be less attractive to pollinators, and the quality or quantity of pollen in flowers with many larvae might be reduced; Quesada et al. 1995, Strauss 1997). We then created three fitness "indices", an herbivore-damage index (mean% damaged flowers), a beetle-attack index [(% frass-soiled flowers) × (mean number of beetle larvae/flower)], and a female fitness index [(number of fruits/flower) × (number of seeds/fruit) × (mean seed weight)] by combining variables shown in Table 1.

Plant, ant, and beetle densities appeared to vary greatly within the experimental plot. To reduce random effects due to this variability, plants were grouped in fours with their neighbours (i.e. each group included one plant of each treatment); within groups, the mean value was subtracted from each data point (Quinn and Keough 2002). This standardization had the additional benefit of normalizing residuals and homogenizing variances. The effects of ant-access, piercing and ant-access × piercing interactions on the fitness indices were tested with two-way ANOVAs in SPSS 10.0 (SPSS Inc. 1989–9999). By the end of the study, some individual plants had died, been severely damaged by rodents, or aborted all of their flowers. Sample sizes vary because some data were collected earlier in the season when fewer plants were damaged. Only groups represented by plants of all four treatments were included in the analyses. Unstandardized means and standard errors for the eight variables are presented in Appendix 1.

Table 1. List of variables measured for each plant. Asterisks denote variables that measure "negative" fitness components.

Variable
Number of flowers per plant
Number of damaged flowers*
Number of flowers with beetle frass*
Number of beetle larvae per plant*
Number of enlarged fruit
Number of unenlarged (failed) fruit*
Total number of seeds
Total seed weight

Results

Animals on *Linaria vulgaris*

Linaria vulgaris plants were host to a wide variety of arthropods ranging from pollinators to herbivores to ambush predators that used them as habitat. Pollinators included four bumblebees (*Bombus bifarius*, *B. flavifrons*, *B. appositus* and *B. californicus*), *Anthophora furcata-terminalis*, and, potentially, a small number of unidentified halictid bees.

Both *G. antirrhinni* and *B. pulicarius* frequently flew from plant to plant. Both were often observed mating on the flowers throughout the study and in the weeks following it. *Brachypterolus pulicarius* larvae typically resided in the flower vestibule (the cavity into which a pollinator's head and thorax fit while it reaches for nectar), although we also often found them in the spurs. Flowers infested with the beetle larvae were often soiled with frass and, in several cases, had shrunken or otherwise damaged anthers.

Three species of ants were frequently observed on *L. vulgaris*. These were *Formica lasioides* Emery, *F. fusca* sspp. and *Tapinoma* spp. *Formica fusca* sspp. and *Tapinoma* spp. were the most common. *Tapinoma*, the smallest of the ants observed (mean ± SD: 2.58 ± 0.349 mm), can squeeze between the closed lips of *L. vulgaris* and was often seen drinking nectar within the spur. The two larger species (mean ± SD: *F. fusca* sspp. = 5.08 ± 0.576 mm and *F. lasioides* = 3.87 ± 0.291 mm) occasionally entered flowers that were open either due to malformation or wilting. Ants were divided into large (both *Formica* species) and small (*Tapinoma* spp.) categories for some analyses because large and small ants can exert predation pressures on different life history stages of their prey (Cushman and Addicott 1991). *Tapinoma* spp. could fit through the holes in the spurs and often imbibed nectar from within the flowers; the larger ants typically drank nectar at the opening of the hole or pushed their heads within the spur. We also observed a small number of ants biting the spur tissue around the piercing, apparently enlarging the hole. Ants of a fourth taxon, *F. fusca neorufibarbis* Emery, although abundant in the study plot and known to thieve nectar from other systems in the area (Galen 1983, Norment 1988), never visited *L. vulgaris* plants.

Ants were patchy in their distribution and were only rarely seen in the vicinity of some of the plants; ants were never observed on 11 groups of plants. Conversely, some plants, especially those growing on a gravel slope on the west side of the plot, were consistently visited by ants.

Ants visiting the plants typically patrolled the stem and flowers and occasionally chewed on floral tissues.

Several plants also hosted spiders (salticids, thomisids, and theridiids), which were occasionally observed killing ants and, once, a nitidulid. Spiders typically remained on the same plants over the survey period.

Observations of behavioural and other species interactions

Although ants and beetles often inhabited the same plant at the same time, we rarely observed interactions between them during the five-minute surveys. In those cases, however, we did see ants attacking and, in two cases, driving away nitidulids or weevils. Ants also reacted aggressively to experimental tools (e.g. tweezers, human hands) and to visiting bumblebees, in one case driving a *Bombus bifarius* worker away from the plant.

Nitidulids and weevils rarely interacted because they were rarely found on the same plants; over the course of the survey period we saw plants with at least one beetle of either species 627 times, with nitidulids alone 571 times, and with weevils alone 20 times. We observed plants with both only 16 times, constituting a significant negative association ($\chi^2 = 5.268$, df = 1, p = 0.022). When we found both species together, they seldom reacted to each other's presence. In one case, however, a weevil charged a nitidulid, driving it off the plant.

Spiders occasionally attacked and killed ants, and the webs that enmeshed some inflorescences often contained several dead ants. One crab spider was also observed holding a dead nitidulid. Crab spiders were, like ants, quite aggressive. They attacked almost any object approaching the plant, including human fingers and pen tips.

Within the fruit, which we dissected in October 2003, we found in addition to larval and adult weevils a small number of adult parasitic wasps of the genus *Pteromalus*, a group known to attack cucurlionid beetles (S. Libenson, pers. comm.).

Ant recruitment to robbed versus unrobbed plants

We observed more ants per survey on robbed plants than on unrobbed plants. For the five plant pairs (data from the 11 sampling epidsodes are pooled for each plant), the number of ants was higher on the robbed individuals than on their unrobbed neighbours (mean ± SD: 3.636 ± 4.076 and 0.982 ± 0.360, respectively; Wilcoxon matched pairs test: Z = 2.023, p = 0.043, n = 5). There were significantly more large ants on the robbed plants than on the unrobbed plants (1.72 ± 0.342 and 0.709 ± 0.088, respectively; Wilcoxon matched pairs test: Z = 2.023, p = 0.043, n = 5) and no difference in the number of small ants among treatments. During the surveys, there were more instances of ants feeding (robbing or forcing their ways into flowers) on robbed plants, whereas ants on unrobbed plants were more likely to be patrolling or sitting motionless ($\chi^2 = 27.092$, df = 1, p < 0.00001).

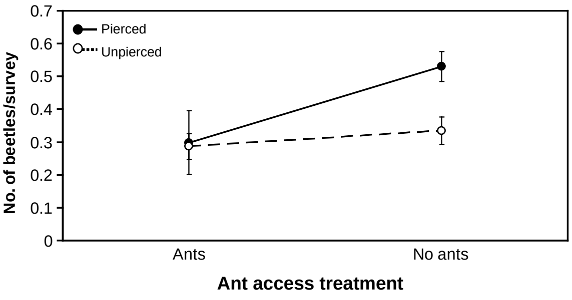

Fig. 1. Effect of ant access and spur piercing on beetle abundance on *Linaria vulgaris* ramets.

Ants and herbivores on *Linaria vulgaris*

There was no difference in mean (± SE) ant abundance between the two treatments that allowed ant access (ants-pierced = 0.301 ± 0.106, ants-unpierced = 0.311 ± 0.074; Mann–Whitney U test: z = 1.032, p = 0.302, n = 38); there was also no difference in recruitment of small or large ants to either treatment ($\chi^2 = 2.61$, df = 1, p = 0.106). There was, however, a difference in activity type among the ant-access treatments: on plants whose flowers were pierced, significantly more ants were seen on or in flowers, than in plants whose flowers were intact ($\chi^2 = 27.71$, df = 1, p < 0.00001).

Ant exclusion had a marginal negative effect on mean (± SE) number of beetles per plant per survey (ant-access = 0.292 ± 0.028, ant-excluded = 0.431 ± 0.054, Mann–Whitney U test: z = 1.83; p = 0.067; n = 155 Fig. 1), the hole treatment had no effect (holes = 0.414 ± 0.054, no holes = 0.310 ± 0.030; Mann–Whitney

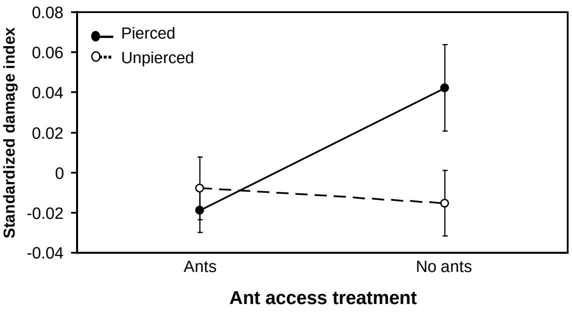

Fig. 2. Effects of the ant and piercing treatments on the herbivore damage index (% damaged flowers per plant per survey). Circles are means ± SE.

U test: $z = 0.992$, $p = 0.321$, $n = 155$). However, ant-excluded, pierced plants had significantly more beetles than any of the other treatments (Wilcoxon matched pairs tests; $Z > 2.482$, $p < 0.020$, Fig. 2).

The presence of spiders was not influenced by the treatments ($\chi^2 = 0.857$; df = 3; $p = 0.836$). However, spider occupancy (spiders per plant per observation) on plants was positively correlated with beetle abundance ($r_S = 0.280$, $p = 0.0004$, $n = 155$), and marginally negatively correlated with ant abundance ($r_S = -0.217$, $p = 0.058$, $n = 77$).

Effects of beetles and ants on plant fitness indices

Ant exclusion had significant effects on the beetle attack and female fitness indices, while the piercing and ant-access × piercing interaction did not (Table 2). Neither ant-access nor piercing significantly affected the flower-damage index, but the ants × piercing interaction was significant; holes in flowers appear to be detrimental when ants are excluded but positive when they are not (Fig. 2). Although not significant, this pattern is also found in the other two indices (Fig. 3, 4).

Mean observed ant abundance on plants was significantly correlated with more than half of the measured variables (Table 3); significant correlations with "negative fitness measures" were negative, and significant correlations with "positive fitness measures" were all positive. In addition, all correlation coefficients for the non-significant correlations were consistent in direction with our predictions about the positive effects of ants on plant fitness. Only number of frass-soiled flowers was significantly (and positively) correlated with mean beetle abundance (Table 3). Spider occurrence on plants was not correlated with any of the variables ($r_S < 0.15$; $p > 0.226$).

Table 2. Results of two-way ANOVAs on fitness indices in *Linaria* vulgaris. Sample sizes varied for analyses of different variables because plants did not always survive long enough to produce mature fruit.

Index	Treatment	df (effect, error)	F	p
Herbivore damage	ants	1, 128	2.647	0.106
	piercing		1.976	0.162
	ants × piercing		4.300	0.040
Beetle attack	ants	1, 116	10.28	0.002
	piercing		0.953	0.953
	ants × piercing		0.648	0.648
	ants	1, 92	9.702	0.002
	piercing		0.397	0.530
Female fitness	ants × piercing		2.411	0.124

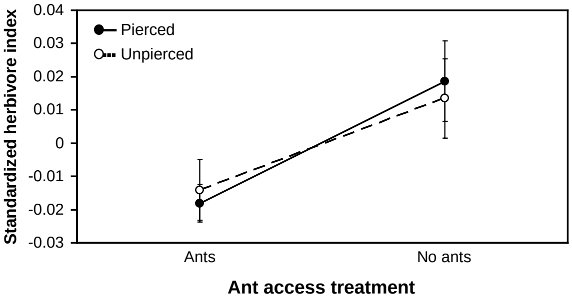

Fig. 3. Effects of the ant and piercing treatments on the beetle attack index ((proportion of flowers soiled with frass per plant) × (mean number of beetle larvae per flower)). Circles are means ± SE.

Discussion

Because they divert nectar from legitimate pollinators, nectar robbers can be seen as disrupting a mutualistic relationship between *Linaria vulgaris* and its pollinators. Species that disrupt mutualisms (aprovechados, Mainero and del Rio, 1985) are generally assumed to reduce the fitness of at least one of the two mutualistic partners. Although both negative and positive effects on plant fitness have been attributed to nectar robbers (reviewed by Maloof and Inouye 2000, Irwin et al. 2001,), most workers still consider robbers to be parasites of plant–pollinator mutualisms, and implicitly treat them as plant antagonists (Proctor et al. 1996, Traveset et al. 1998, Irwin and Brody 1999, 2000, Stout et al. 2000, Bronstein 2001, Lara and Ornelas 2001, Yu 2001, Anderson and Midgley 2002, Irwin 2003, Stanton 2003). Here we show that flower piercing may indirectly benefit a plant, *L. vulgaris*, by attracting secondary robbers (ants) that also reduce the impact of herbivores. Primary nectar robbers, by making holes in the nectar spurs, effectively create a plant trait that creates or strengthens a protective ant–plant interaction. Ants that normally explore the plants in low numbers may, upon finding access to nectar through holes in the spurs, recruit to robbed plants in much the same way as they would to plants bearing extra-floral nectaries. It is also plausible that

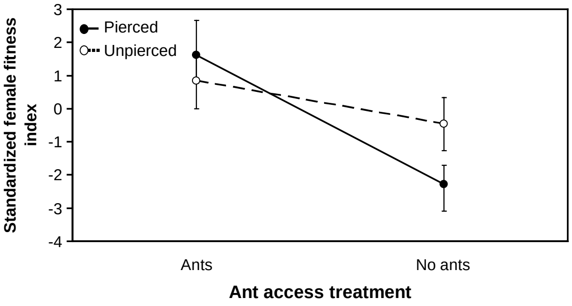

Fig. 4. Effects of the ant and piercing treatments on the female fitness index ((number of fruits/flower) × (number of seeds/fruit) × (mean seed weight)). Circles are means ± SE.

Table 3. Spearman correlations between mean ant and beetle abundances and plant fitness variables.

	Mean ant abundance		Mean beetle abundance	
	Spearman r	p-value	Spearman r	p-value
Number of damaged flowers*	−0.126	0.145	0.071	0.412
Flowers with frass*	−0.209	0.037	0.224	0.025
Number of larvae*	−0.294	0.005	0.120	0.264
Number of fruit	0.132	0.138	0.129	0.147
Number of failed fruit*	−0.040	0.655	0.136	0.126
Number of seeds	0.192	0.030	0.067	0.452
Mean seed weight	0.302	0.003	−0.026	0.804
Mean ant abundance			−0.064	0.458

ants can detect chemical indications of nectar robbing activity (i.e. the smell of nectar or other volatiles released by the corolla tissues). Our results also suggest that pierced flowers attract the beetles (Fig. 1, 2), but it appears in this case that the benefit of recruited ants exceeds the costs of recruited herbivores. In this study, ant access positively affected plant fitness components. It remains unclear whether ants act upon beetles primarily through consumptive or behavioural effects (Rudgers et al. 2003), and whether they act more upon adult beetles or larvae that can presumably be accessed through holes in the spurs (D. A. Newman, pers. obs.).

To our knowledge, this is the first study to show that nectar robbing can have indirect effects on plant fitness through mechanisms other than pollinator behaviour. Although the ant exclusion study did not reveal an effect of the flower piercing (artificial robbing) treatment on any fitness component, the ant recruitment study, conducted in a small subset of the plot with consistently high ant activity, did. Ants on pierced plants also behaved differently than those on unpierced plants; they were significantly more often feeding on nectar or patrolling flowers, where aggressive interactions with the herbivorous beetles were most likely to occur. In addition, the significant ant × piercing interaction effect found for the herbivore damage index, and the non-significant patterns that suggest such an interaction in the other two indices, support the prediction that holes may have effects contingent upon ants for at least some components of reproductive fitness. Our inability to show significant effects of the robbing treatment in the exclusion study is most likely due to spatial variation in ant recruitment within the study plot (see Bronstein 1998 on intraspecific variation in ant–plant protection mutualisms). Ant abundances are known to vary drastically within very small areas, depending in large part on the proximity of colonies (Cushman and Addicott 1991). Although the interactions we demonstrate may be strong only in some patches, they could still be ecologically important. Indeed, even if the average effect of ants and robbers (and spiders, for that matter) on the success of *L. vulgaris* is very small, as long as it is sometimes locally strong, as we have shown in this study, it could amount to more than a simple ecological curiosity (Berlow 1999). Indeed, if plants located close to ant colonies (which are often long-lived, and would thus be associated with the same *L. vulgaris* individuals for several years; Cushman and Addicott 1991) are consistently enjoying greater reproductive success than plants in ant-poor areas which may allocate more resources to vegetative growth than to sexual reproduction, a differential contribution of the ant-attended plants to future generations could be expected.

Nectar robbers can have indirect and counterintuitive effects on reproductive success in *L. vulgaris*. Their effects on pollinator behaviour and the resulting beneficial impacts on plant fitness have been well studied (reviewed by Maloof and Inouye 2000); here, we show that taxa unrelated to pollination may also benefit nectar-robbed plants. One of the important questions in community ecology is how indirect an effect can be without becoming completely negligible. Theorists have proposed that indirect effects should be attenuated when many species interact due to dilution of interaction strength, resulting from the magnification of noise as the number of links in the assemblage increases (Strauss 1991, Wootton 1994, Williams et al. 2002). However, theoretical and empirical studies have also shown that indirect effects may be as strong as, or stronger than, direct effects (Wootton 1994, Menge 1995, Berlow 1999, Williams et al. 2002). In this study, simulated nectar robbing, whose direct and pollinator-mediated indirect effects on female fitness are weak or non-existent, had highly indirect but significant effects on components of reproductive success. It is important to note that, unlike real nectar robbers, simulated robbing did not remove nectar from the spurs. However, we found that primary and secondary robbers did not always remove all the nectar from *L. vulgaris* at the RMBL, and that nectar production did not change in robbed flowers (D. A. Newman, pers. obs.). In addition, ants were even attracted to minute quantities of floral rewards, and would therefore still recruit to robbed plants whose holes allow access to traces of nectar; for example, we often observed ants feeding on immeasurably small amounts of sugar around the ovaries of recently abscised flowers.

It is possible that beneficial indirect effects of ants on *L. vulgaris* were tempered somewhat by negative direct effects of their foraging activities. Indeed, we observed ants chewing on floral tissues in both ant-access treatments; this may have detrimental impacts on flower attractiveness, flower longevity and fruit success (Krupnick and Weis 1998, Krupnick et al. 1999, Utelli and Roy 2001). Such conditionality in the consequences of species interactions is well known (Cushman and Addicott 1991, Bronstein et al. 2003, reviewed by Thompson 1988 and by Bronstein 1998) and could, depending on ecological circumstances, result in interactions that span from mutualism to commensalism to antagonism.

In addition to the main effects, we found suggestive evidence of an additional guild that, through yet another indirect step, may modify the observed robber–ant–beetle interactions. Plants that often hosted spiders hosted more beetles and almost significantly fewer ants than plants that hosted none or few. We saw spiders kill ants by ambush or in their webs. Gastreich (1999) found that theridiid spiders kill *Pheidole bicornis* ants visiting *Piper obliquum*, a plant with which they have a facultative protection mutualism; the resulting avoidance of plants with webs by ants caused an increase in herbivory. Due to the small number of plants that hosted spiders in this study, however, we could detect no significant correlations between spider occurrence and any of the fitness variables; it is likely that the effect of spiders in this system is highly incidental and very patchily distributed.

The effect we demonstrate in this study is almost certainly dependent on the floral morphology of plants that, like *L. vulgaris*, can deter nectar-seeking ants. Without closed corollas, which provide an effective barrier between ants and nectar, robbers would not modify the physical availability of "reward" and would therefore contribute no additional ant protection. The plant's natural history is also probably important. As discussed earlier, researchers have found that nectar robbing has no significant impact on the female fitness of *L. vulgaris* (Stout et al. 2000, Irwin and Maloof 2002, Nepi et al. 2002); in a species that suffered reductions in pollination due to nectar robbers, it is doubtful that ants attracted to robbed plants could counter the negative effects of pollen limitation.

One of the factors that has kept nectar robbing, despite its ubiquity and taxonomic breadth, from appreciation as an important phenomenon in plant evolutionary ecology (Irwin et al. 2001, Ehrlén 2002) is the lack of generality that researchers have found in the outcomes of plant–pollinator–robber interactions (Morris 1996, Maloof and Inouye 2000, Agrawal 2001, Irwin et al. 2001). For this reason, studies have been aimed primarily at demonstrating whether nectar robbers are parasites or mutualists, while very few (Lara and Ornelas 2001) have even implicitly addressed the potentially more interesting questions about how and why the magnitude and sign of their effects vary in time, in space, and among systems (Cushman and Addicott 1991, Irwin and Maloof 2002). In the system described in this study, we provide one of many potential mechanisms that may cause the effects of nectar robbing to vary; researchers interested in this and other ecological phenomena may benefit from considering species that do not appear to interact with their focal species. For the development of relevant questions and hypotheses in such complex systems, however, it is critical to base them not only on the rapidly growing body of theory dealing with indirect effects in ecological communities, but also on intimate knowledge of their natural history.

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Appendix 1. Means (±SE) for unstandardized variables.

	Ants, pierced	Ants, unpierced	No ants, pierced	No ants, unpierced
Mean number of ants per survey (n = 34)	0.155 (0.039)	0.182 (0.038)	0 (0)	0 (0)
Mean number of beetles per survey (n = 34)	0.265 (0.041)	0.257 (0.040)	0.480 (0.078)	0.298 (0.089)
Flower number (n = 34)	12.80 (1.424)	11.93 (1.222)	11.567 (1.053)	12.33 (1.139)
Number of damaged flowers (n = 34)	0.300 (0.063)	0.355 (0.074)	0.563 (0.105)	0.314 (0.089)
Flowers with beetle frass (n = 30)	1.733 (0.569)	1.567 (0.446)	2.400 (0.643)	3.233 (0.762)
Number of beetle larvae per plant (n = 30)	0.733 (0.235)	0.667 (0.237)	1.300 (0.413)	1.267 (0.392)
Number of fruit per plant (n = 32)	15.63 (1.344)	14.53 (1.638)	14.438 (1.279)	14.219 (1.536)
Number of failed fruit (n = 32)	6.938 (0.948)	7.281 (1.117)	9.125 (1.080)	8.250 (1.223)
Number of seeds per plant (n = 32)	1655.50 (186.08)	1752 (294.74)	1487.44 (221.91)	1445.00 (214.25)
Mean seed weight (mg) (n = 23)	0.149 (0.007)	0.139 (0.006)	0.128 (0.007)	0.129 (0.007)