

GLOSSOPHAGINE BAT POLLINATION IN *HELICTERES BARUENSIS* (STERCULIACEAE)

Otto von Helversen¹* & Christian C. Voigt

Zoologisches Institut II, Universität Erlangen-Nürnberg, Staudtstr. 5, 91058 Erlangen, Germany

Abstract. *Helicteres baruensis* Jacq. (Sterculiaceae) is a common shrub of the tropical dry forest in the Pacific lowlands of Costa Rica. We studied the floral characteristics of *Helicteres baruensis* and observed the feeding activity of animals at the inflorescences. Flowers of *Helicteres baruensis* were visited by moths and nectar-feeding bats, *Glossophaga soricina* (Phyllostomidae; Glossophaginae). Only glossophagine bats touched the anthers and stigma; pollen was deposited on the throat and belly when they approached and hovered in front of the flowers. Nectar secretion of the flowers was confined to a single night with a peak nectar secretion rate of *c.* 25 µl per hour around midnight and an average sugar concentration of 15% (weight/weight), which is in agreement with the syndrome of chiropterophily. Flowers of *Helicteres baruensis* are difficult for large-sized pollinators to exploit because: (1) they are located at the tip of pendulous branches, (2) they lack structures that enable a heavy pollinator to cling to them, (3) the androgynophore is oriented into the flight path of an approaching pollinator, and (4) nectar is offered deep in the calyx. Based on these floral characteristics we predicted that only glossophagine bats with their sophisticated ability for maneuvering and hovering flight would be able to exploit flowers of *H. baruensis*. In agreement with this prediction we found pollen of *H. baruensis* only on *Glossophaga soricina* and not on syntopic frugivorous bats of the subfamilies Stenodermatinae and Carollinae. Therefore, we suggest that *Helicteres baruensis* is adapted to pollination by glossophagine bats and that *Glossophaga soricina* is the main pollinator of *H. baruensis* in northwestern Costa Rica. The extrafloral nectararies in the vicinity of the flowers possibly manipulate the communication system of ants to prevent foraging on floral nectar. Accepted 23 February 2002.

Key words: Bat pollination, *Glossophaga*, extrafloral nectararies.

INTRODUCTION

Bar-pollinated plants are found predominantly in the tropical regions. Dobat & Peikert-Holle (1985) counted approximately 570 bat-pollinated plant species in the New World contrasting with 160 known for the Old World. Flowers visited by bats share some common characteristics, such as nocturnal anthesis, bell-shaped or pincushion-like corollas, whitish, greenish, or purplish colors, exposed position, large amounts of nectar and pollen, a sugar concentration of approximately 16% of nectar, and a musty to sour odor often dominated by sulfur compounds (e.g., Vogel 1958, Fægri & van der Pijl 1979, Dobat & Peikert-Holle 1985, von Helversen 1993, Knudsen & Tollsten 1995, von Helversen *et al.* 2000).

Vogel (1990) discussed the parallel radiation of Neotropical plants into different pollination modes, including chiropterophily, by describing, among other examples, the floral syndromes of the family Sterculiaceae and the corresponding pollinators. Among the

Sterculiaceae, members of the genus *Helicteres* provide an outstanding example of diversification in pollinator use within a single genus. Floral syndromes include psychophily, ornithophily and chiropterophily (Sazima & Sazima 1988, Vogel 1990); with bat-pollination confirmed in *Helicteres rekoii* (Vogel 1990) and *Helicteres ovata* (Sazima & Sazima 1988). Vogel (1954) mentioned *Helicteres jamaicensis* as being sphingophilous, but this species may also be bat-pollinated, as its scent is unpleasant, reminiscent of the smell of pumpkin (Vogel, pers. comm.). Bat-pollination in *Helicteres baruensis* was first proposed by Haber & Frankie (1989). Subsequently Ruiz *et al.* (1997) found pollen of *H. baruensis* in the feces and fur of *Glossophaga longirostris* in the arid zone of Tatacoa in Colombia. However, neither authors presented details of the floral characteristics or the specific mode of pollination.

Among the four phyllostomid subfamilies known to pollinate plants, Carollinae, Stenodermatinae, Phyllostominae, and Glossophaginae (Dobat & Peikert-Holle 1985), glossophagine bats are especially well adapted for flower exploitation and represent the

* To whom correspondence should be addressed

¹ e-mail: helver@biologie.uni-erlangen.de

most important mammalian pollinator of Neotropical plants. Von Helversen (1993) suggested that the parallel and independent evolutionary acquisition of bats as pollinators in Neotropical plant families (Vogel 1958, 1968/69, 1990; Dobat & Peikert-Holle 1985) was facilitated by the evolution of small-sized bats capable of sustained hovering flight. Glossophagine bats, with body masses of down to 6 or 7 g, can cover their energy demands with comparatively small portions of nectar – less than 10 μ l per visit – and their small size is also advantageous for energetically costly hovering flight (von Helversen 1986, 1993). Glossophagine bats expend less energy during hovering than moths and hummingbirds of similar size probably because of their larger wing span (Voigt & Winter 1999). Therefore, glossophagine bats may achieve a net energy gain from flower exploitation even if the single nectar reward is small. All these features in a potential new pollinator may have allowed many taxonomic groups of New World plants to switch from pollination by sphingids, birds, or terrestrial mammals to glossophagine bats (von Helversen 1993, Winter & von Helversen 2001). In the following we show that *Helicteres baruensis* belongs to the group of plants specifically adapted to the pollination by bats of the subfamily Glossophaginae. We describe some floral characteristics of this plant and the mode of flower exploitation by its predominant glossophagine pollinator.

MATERIALS AND METHODS

Helicteres baruensis Jacq. is a shrub of seasonal dry forests, growing up to 5 m in height and flowering at the end and at the beginning of the rainy season (M. T. Fernandez-Morillo, pers. comm.). We observed *Helicteres baruensis* and its pollinators in the Guanacaste Protection Area (Santa Rosa National Park), in northwestern Costa Rica (district of Guanacaste; 10°50' N, 85°37' W). The distinctly seasonal forest of the Pacific lowland in northern Costa Rica has been described by Holdridge *et al.* (1971), Janzen & Liepner (1980), Hartshorn (1983), and others. We visited the site in November 1995 and November/December 1996 at the end of the rainy season and found flowering shrubs of *Helicteres baruensis* at the border of secondary dry forests and in abandoned pastures surrounding the Santa Rosa Biological Station (50 to 100 m above sea level).

Nectar secretion rates of 21 flowers located on 10 plant individuals were measured at three-hour inter-

vals during two consecutive nights. Additionally, we also monitored the nectar secretion of each flower during the day. We refer to day 0 as the day of anthesis and day 1 and day-1 as one day prior and one day after anthesis respectively. We covered the inflorescences with plastic bags to prevent any visitation and extracted the nectar from the nectar containers with capillary tubes. Sugar concentration (% weight/weight) was measured with a refractometer (Atego) to an accuracy of 0.2%.

During five nights, from 18:00 to 22:00 or 23:00 h, we captured bats with mist nets near *Helicteres baruensis* bushes. We set two to three mist nets (6m, 50 D*2, 19 mm mesh) near to flowering bushes of *H. baruensis* where we expected the flightpath of approaching bats. Pollen samples were taken with gelatine cubes (Thomas 1988) from all regions of a bat's body. The pollen was stained with Fuchsin and all samples were mounted on slides and checked under a light microscope for the presence of *Helicteres baruensis* pollen.

To describe the mode of visitation in detail, flower visitors of *H. baruensis* were observed with a night-vision scope and videotaped with an infrared-sensitive camera (Sanyo). Infrared light was provided by a large array of IR-photodiodes that fired 0.5 ms flashes of 750 nm wavelength, triggered by the video camera. We converted the video sequences of interest into bitmap sequences using a video board and video editing software (Adobe premiere). To gain a higher time-resolution we split each picture into its two half-pictures and converted the resultant sequences again into a video sequence using a custom-written program.

RESULTS

(a) Floral Characteristics

Flower morphology. Flowers were exposed on the tips of branches in groups of one to five at a height of one to five meters. Only one or rarely two flowers per group opened during a single night. The appearance of the flowers was dominated by a large green calyx. The corolla was reduced to five inconspicuous, long, slender, whitish lobes (Fig. 1), curved back from the calyx. The green calyx had an average length of 3.5 cm, and opened with a diameter of 1.5 to 2 cm in the vertical axis and 0.7 to 1 cm in the horizontal axis, giving the flower a zygomorphous, monosymmetric shape. The androgynophore protruded from the ca-

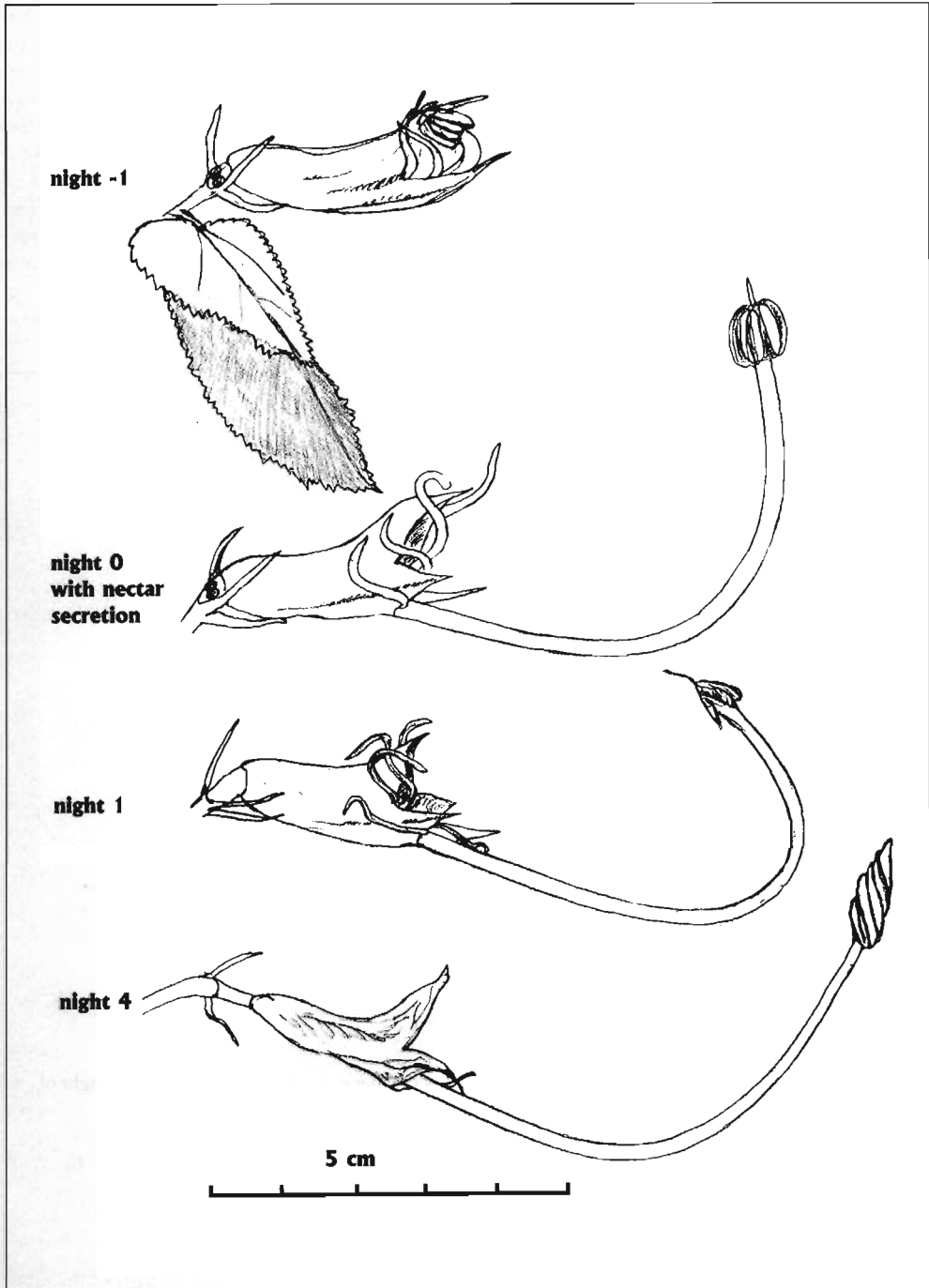


FIG. 1. Flowers of *Helicteres baruensis* on several consecutive nights. The flowering night, when nectar is secreted, was defined as "night 0."

lyx approximately 8 to 10 cm at anthesis. The anthers were located on the tip of the androgynophore, followed distally by the large pistil with its filamentous style. The overall coloration of the flower was whitish (corolla and androgynophore) to greenish (calyx). The flowers had a faint but distinctly chiropterophilous odour.

Buds opened slowly, usually starting one night before the onset of nectar secretion. Initially the curved androgynophore was hidden in the closed bud. When the bud opened, the medial part of the androgyno-

phore protruded from the calyx with the reproductive organs still hidden (Fig. 1, night -1). During the following day, the androgynophore grew out of the calyx and unfolded stretching out and upwards. Anthers dehiscid at dusk of the same day (night 0 in Fig. 1) and nectar secretion began. During the following day, the petals, pistil and anthers did not change their position but gradually became dry and brownish. A few days later the characteristic coiled, "helical" structure of the fruit – from which the genus name *Helicteres* originates – developed.

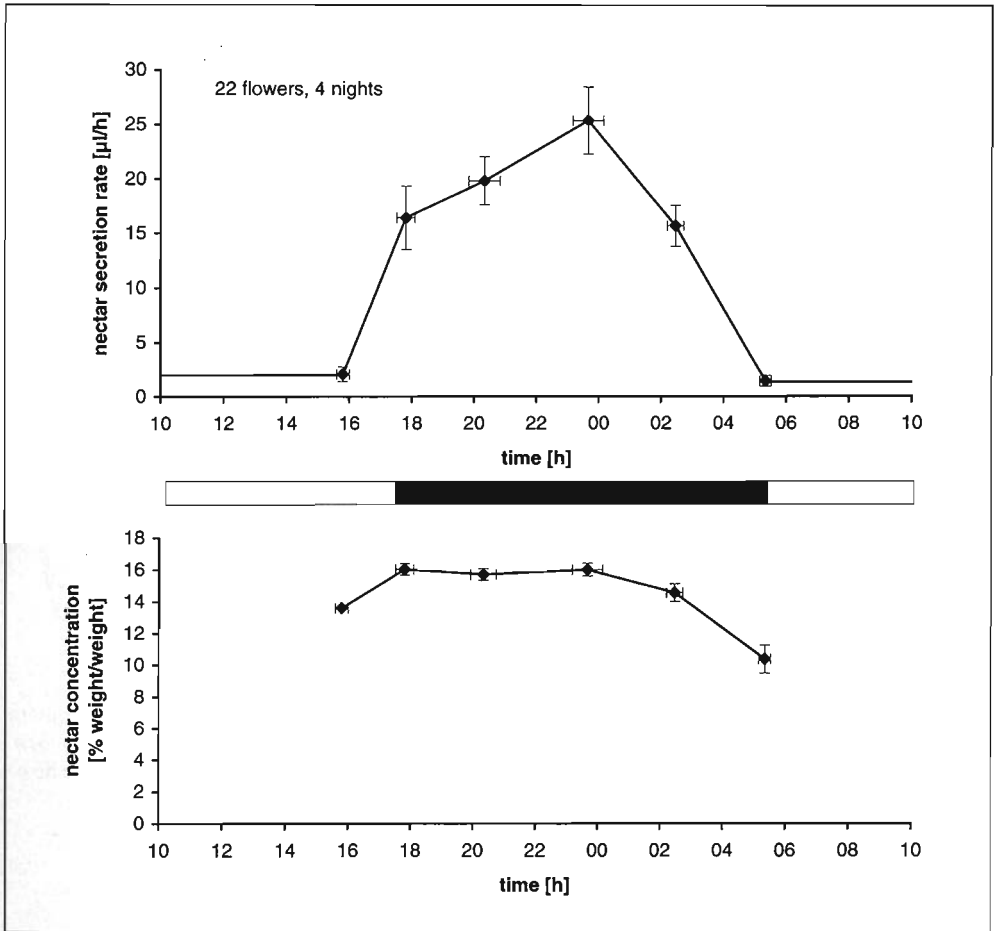


FIG. 2. (top) Mean secretion rate ($\mu\text{l} \times \text{h}^{-1}$) and (below) mean sugar concentration (% wt/wt) of the nectar of *Helicteres baruensis*. The rate of secretion was calculated by dividing the amount of extracted nectar by the time interval since the last extraction and was plotted against the mean of the time interval ($n=21$; average values for two-hour intervals; error bars give SE).

Nectar secretion. The average total nectar secretion per flower over 24 hours was 235 ml ($\pm 106 \mu\text{l}$ SD; $n = 21$; max. = 475 μl). Nectar secretion started between 16:00 and 17:00 h (Fig. 2). After a peak nectar secretion of ca. 27 $\mu\text{l h}^{-1}$ around midnight the secretion rate decreased, and nectar production stopped around 04:00 to 05:00 h in the morning. Each flower secreted nectar only during a single night. The sugar concentration of the nectar averaged 15% (weight/weight). During peak nectar secretion the concentration was slightly higher (mean 16%) (Fig. 2B). Nectar concentration dropped to approximately 10% in the morning. Sugar was probably reabsorbed actively by the flower in the morning hours as we found little or no nectar during the day, even in flowers that had been bagged during the whole preceding night.

(b) Behavioral observations

Mist netting. We caught the first bats shortly after sunset, around 18:00 h. We mist netted the following species in the vicinity of *Helicteres baruensis*: *Artibeus jamaicensis* (3 individuals), *Dermanura toltecus* (1), *Dermanura phaeotishwatsoni* (4), *Glossophaga soricina* (10), *Carollia perspicillata* (1), *Carollia subrufa* (1). Of the six species only *Glossophaga soricina* carried pollen of *Helicteres baruensis* (all individuals). The pollen of *H. baruensis* was concentrated in the fur of the throat and belly and on the proximal parts of the ventral side of the wings.

Videotaping. *Glossophaga soricina* usually approached the flower from above, maneuvering downward in a slow hovering flight (Fig. 3). The androgynophore was pushed down with the bat's body, and by coming closer to the calyx the anthers and the pistil slid ventrally along the bat's belly. In doing so, the pollen was deposited ventrally on the body and the underside of the wings near to the body. Still hovering, the bat pushed its snout into the opening of the calyx to reach the nectar with its tongue. Due to the pressure exerted by the bat, the slender, pendulous flower branches were pushed down, swinging up again when the bat left. Flower visits only lasted for approximately a half second.

Bats visited all flowers of the same bush or group of bushes consecutively, often within short time intervals of only 5 to 20 s. Flowers that were located high in the shrub were preferred. Individual flowers were visited every 15 to 40 minutes during the first half of the night.



FIG. 3. *Glossophaga soricina* approaching the flower of *Helicteres baruensis*. Single frame from a video recording made with an IR-sensitive camera and IR-emitting diode flashes (Nov. 26, 1996; Sta. Rosa National Park, Costa Rica).

We observed constant feeding activity of moths (mostly medium-sized noctuids) on *Helicteres baruensis* during the night. Moths approached the flower from the side or from below and landed on the calyx. We never saw moths touching either anthers or stigma. During the period of observation no sphingid moths ever visited *Helicteres baruensis*.

Robbing of flowers. During the day, especially at dawn and dusk, large black Meliponine bees robbed the flowers by biting a hole into the base of the calyx. The bees seemed to be more interested in the pollen, which they took from the hidden anthers one day prior to

flowering, than in the nectar. They may have also profited from nectar at those flowers that had not been visited by bats during the night.

Extrafloral nectaries. Starting at least one week before anthesis, two extrafloral nectaries developed at the base of each flower bud. The large green disks of the extrafloral nectaries were covered with small amounts of nectar during both the day and night and they dried when the flower withered. A single measurement revealed the same sugar concentration in the extrafloral nectaries as in the flower (c. 15%). Several ant species were attracted to the extrafloral nectaries and the species composition differed sometimes between day and night. Usually only a few (not more than 4 to 6) individuals visited an extrafloral nectary at the same time. Visitation rate at the extrafloral nectaries did not differ between days. We never observed ants invading the calyx in the flowering night and gathering flower nectar, although the flowers lacked any mechanical obstacles that could have prevented such an intrusion.

DISCUSSION

Bat pollination. Our observations reveal that *Glossophaga soricina* was the main nocturnal visitor of *Helicteres baruensis* flowers and that only *G. soricina* carried pollen of this plant in its fur. The sugar concentration of the nectar and other floral characteristics are typical for a bat-pollinated flower (von Helversen 1993). Based on our observations we conclude that *H. baruensis* is bat-pollinated and that members of the subfamily Glossophaginae are the predominant pollinators of *Helicteres baruensis* in the Guanacaste area of Costa Rica, at least during its flowering period at the end of the rainy season. In Tatacoa, Colombia, Ruiz *et al.* (1997) found that in the absence of *G. soricina* the congeneric similarly sized *G. longirostris* was a visitor to *H. baruensis*. More detailed investigations at other study sites and at other times of the year, e.g., at the beginning of the rainy season, will be useful for comparison.

Although sphingid moths visited neighboring sphingophilous plants and the chiropterophilous *Bauhinia unguolata* (see also Heithaus *et al.* 1975, C.C. Voigt & O. von Helversen, pers. observ.) we never observed or videotaped them at *Helicteres baruensis*. Sphingid moths may occasionally visit *Helicteres baruensis* but Haber & Frankie (1989) found pollen of *H. baruensis* on the proboscis of hawkmoths in only

one out of 546 studied specimens, suggesting that such visits are rare.

The floral syndrome of *Helicteres baruensis* is typical for "glossophagine" flowers (von Helversen 1993) and presumably excludes bats of the subfamilies Carrollinae, Stendermatinae, and Phyllostominae. We suggest that the exclusion of non-glossophagine bats is due to the following specific plant characteristics: (1) The flowers are situated on the tip of long and pendulous branches which immediately bend down when a bat tries to cling to them, (2) the long androgynophore reaches into the flight path of an approaching bat, which makes the handling of the flower more difficult, (3) the nectar is hidden in the calyx and therefore not reachable by short-tongued bats, (e.g., *Dermanura*, *Carollia*, *Artibeus*, and *Phyllostomus*), and (4) the small amount of nectar is sufficient only for small-sized bats. With a secretion rate of c. 25 $\mu\text{l h}^{-1}$ the energy gain is at the lower margin, even for a *Glossophaga* if the bat visits the flower 1 to 3 times per hour (von Helversen *et al.*, in prep.).

Convergent evolution towards bat pollination. The genus *Helicteres* is an example of the radiation of a plant taxon into different pollination modes, commonly found in many angiosperms (Vogel 1990). An evolutionary shift from one pollinator guild to another may occur when the resource usages by pollinators overlap and/or the supply of pollinators changes in time or space. Another driving force may be the need for prefertilization isolating mechanisms to avoid hybridization between related, syntopic species (Grant 1949). The ornithophilous species *Helicteres guazumaefolia* HBK commonly occurs syntopically with *H. baruensis* in Guanacaste. The flowers of *Helicteres guazumaefolia* are about half the size of those of *H. baruensis* and differ from them in possessing broad, red petals and a much shorter androgynophore. It flowers at the same time of the year and is visited by several species of hummingbirds during the day (Voigt & von Helversen, pers. observ.).

It is likely that *Helicteres baruensis* is derived from a moth-pollinated ancestor. The flowers share several characteristics with moth-pollinated flowers, especially the long, narrow, whitish petals. In this respect the flower morphology of *Helicteres baruensis* is very different from the other two bat-pollinated congeners. *Helicteres rekoii* has large, thick, waxy, white petals which open broadly (Vogel 1990) and *H. ovata* has a tube-like corolla with broad, deep red petals, resembling ornithophilous species like *H. guazumaefolia*.

lia. Thus the three species appear to have converged on chiropterophily independently, thus demonstrating the ease of an evolutionary change in the mode of pollination, especially from sphingid and bird pollination to glossophagine bat pollination (von Helversen 1993).

Extrafloral nectaries. In *H. baruensis* two large extrafloral nectaries located at the base of each flower are visited by several ant species. These extrafloral nectaries are visited throughout the day and night by a number of ant species that take the secreted nectar. It has been proposed that extrafloral nectaries serve to protect the plant by attracting guarding ants (e.g., Koptur 1992). However, several questions about extrafloral nectaries, especially regarding those that are located close to flowers, remain: (1) Do extrafloral nectaries protect flowers from "nectar thieves" (van der Pijl 1954) without deterring pollinators (Willmer & Stone 1997)?, and (2) how do plants prevent "guarding" ants from exploiting the flower nectar (Haber *et al.* 1981)?

In *Helicteres baruensis*, nectar-robbing Meliponine bees were not attacked by ants, which makes a protective role of ants unlikely (see also Fiala *et al.* 1996). Even more astonishing is that we never observed ants exploiting the floral nectar, although the extrafloral nectaries are in close proximity. Possibly ants are distracted from the flower nectar by some feature of the extrafloral nectaries. The first person who raised this idea was Kerner von Marilaun (1876), and he illustrated his idea with the example of *Impatiens tricornis*. Plants possibly exploit the communication system of the ants by simulating a poor nectar source. If the ants learn that a certain nectar source is insufficient for a large number of foragers, it is unlikely that they will recruit other foragers and, as a consequence, the colony as a whole is "discouraged" to initiate more foraging visits to this site. This hypothesis seems to be promising but also remains to be tested.

ACKNOWLEDGMENTS

We are grateful to Dagmar von Helversen, Rohini Balakrishnan, and Felix Matt for helping to measure nectar secretion rates during long nights, and to Carlos Drews and Mariate Fernandez-Morillo for help in the field, and to all of them, and especially to Stefan Vogel and to York Winter, for fruitful and critical discussions. We also thank Tigga Kingston for improving the English of this manuscript.

REFERENCES

- Dobar, K., & T. Peikert-Holle. 1985. Blüten und Fledermäuse. Frankfurt/Main.
- Fægri, K., & L. van der Pijl. 1979. The principles of pollination ecology. Oxford.
- Fiala, B., Krebs, S.A., Barlow, H.S., & U. Maschwitz. 1996. Interactions between the climber *Thunbergia grandiflora*, its pollinator *Xylocopa latipes* and the ant *Dolichoderus thoracicus*: the "nectar thief hypothesis" refuted? *Malayan Nature Journal*: 501–14.
- Grant, V. 1949. Pollination systems as isolating mechanisms in angiosperms. *Evolution* 3: 82–97.
- Haber, W.A., & G.W. Frankie. 1989. A tropical hawkmoth community: Costa Rican dry forest Sphingidae. *Biotropica* 21: 155–172.
- Haber, W.A., Frankie, G.W., Baker, H.G., Baker, I., & S. Koptur. 1981. Ants like flower nectar. *Biotropica* 13: 211–214.
- Hartshorn, G.S. 1983. Plants: Introduction. Pp. 118–157 in Janzen, D. H. (ed). *Costa Rican natural history*. Chicago.
- Heithaus, E.R., Opler, O. A., & H.G. Baker. 1975. Bar activity and pollination of *Bauhinia pauletia*. *Plant-pollinator coevolution*. *Ecology* 55: 412–419.
- v. Helversen, O. 1986. Blütenbesuch bei Blumenfledermäusen: Kinematik des Schwirfluges und Energiebudget im Freiland. *Biona-report* 5: 107–126. Akad. Wiss., Mainz; G. Fischer, Stuttgart, New York.
- v. Helversen, O. 1993. Adaptations of flowers to the pollination by glossophagine bats. Pp. 41–59 in Barthlott, W., Naumann, C.M., Schmidt-Loske, K., & K.-L. Schuchmann (eds.). *Plant-animal interactions in tropical environments*; Museum Koenig, Bonn.
- v. Helversen, O., Winkler, L., & H.J. Bestmann. 2000. Sulphur-containing "perfumes" attract flower-visiting bats. *J. Comp. Physiol. A* 186:143–153.
- Holdridge, L.T., Grenke, W.C., Harthaway, W.H., Liang, T., & J.A. Tosi. 1971. Forest environments in tropical life zones: a pilot study. Oxford.
- Janzen, D.H., & R. Liesner. 1980. Annotated check list of plants of lowland Guanacaste Province, Costa Rica, exclusive of grasses and non-vascular cryptogams. *Brenesia* 18: 15–90.
- Knudsen, J.T., & L. Tollsten. 1995. Floral scent in bat-pollinated plants: A case of convergent evolution. *Bot. J. Linnean Society* 119: 45–57.
- Koptur, S. 1992. Extrafloral nectary mediated interactions between insects and plants. Pp. 82–129 in Bernays, E. (ed.). *Insect-plant interactions*, Vol IV. Boca Raton.
- Marilaun, K. von. 1876. Die Schutzmittel der Blüten gegen unberufene Gäste. *Festschrift Zool. Bot. Ges. Wien*. Wien. 189–261.
- Pijl, L. van der. 1954. *Xylocopa* and flowers in the tropics. I–III. *K. Nederlandse ak. Wetens. Proc., C*, 57: 413–423, 541–562

- Ruiz, A., Santos, M., Soriano, P., Cavelier, J., & A. Cadena. 1997. Relaciones mutualísticas entre el murciélago *Glossophaga longirostris* y las cactáceas columnares en la zona árida de La Tatocoa, Colombia. *Biotropica* 29: 469–479.
- Sazima, M., & I. Sazima. 1988. *Helicteres ovata* (Sterculiaceae), pollinated by bats in Southeastern Brazil. *Botanica Acta* 101: 269–271.
- Thomas, D.W. 1988. Analysis of diets of plant visiting bats. Pp. 211–220 in Kunz, T. H. (ed.). *Ecological and behavioural methods for the study of bats*. Washington, D.C.
- Vogel, S. 1954. Blütenbiologische Typen als Elemente der Sipplengliederung. Jena.
- Vogel, S. 1958. Fledermausblumen in Südamerika. *Österreich. Bot. Zeitschr.* 104: 491–530.
- Vogel, S. 1968. Chiropterophilie in der neotropischen Flora. *Neue Mitteilungen I. Flora* 157: 562–602.
- Vogel, S. 1969. Chiropterophilie in der neotropischen Flora. *Neue Mitteilungen II., III. Flora* 158: 185–222, 289–323.
- Vogel, S. 1990. Radiación adaptativa del síndrome floral en las familias neotropicales. *Boletín Academia Nacional Ciencias Córdoba, Argentina* 59: 5–30.
- Voigt, C.C., & Y. Winter. 1999. Energetic cost of hovering flight in nectar-feeding bats (Phyllostomidae: Glossophaginae) and its scaling in moths, birds, and bats. *J. Comp. Physiol. B.* 169: 38–48.
- Willmer, P.G., & G.N. Stone. 1997. How aggressive antagonists assist seed-set in *Acacia* flowers. *Nature* 388:165–167.
- Winter, Y., & O. v. Helversen. 2001. Bats as pollinators: foraging energetics and floral adaptations. Pp. 148–170 in Chittka, L., & J.D. Thomson (ed.). *Cognitive ecology of pollination*. Cambridge.