Behavioral ecology of euglossine bees of the Atlantic rain forest

Jacquelyn Lee Blackmer

Ph.D. in Entomology from The Ohio State University, Ohio Agricultural Research and Development Center, Wooster, Ohio, USA; Currently - Research Entomologist, US Arid-Land Agricultural Research Center, Agricultural Research Service, United States Department of Agriculture, Maricopa, Arizona, USA

Athayde Tonhasca Jr.

Ph.D. in Entomology from The Ohio State University, Ohio Agricultural Research and Development Center, Wooster, Ohio, USA; Currently -Invertebrate Specialist, Scottish Natural Heritage, UK

Gilberto S. Albuquerque

Ph.D. in Entomology from Cornell University, Ithaca, New York, USA; Currently - Associate Professor, Universidade Estadual do Norte Fluminense, Campos dos Goytacazes, RJ, Brazil

Abstract

Before the colonization of Brazil, the Atlantic rain forest extended from Rio Grande do Norte to Rio Grande do Sul. Today, however, the Atlantic forest has been reduced to only 8% of its original size and is highly fragmented. Because of its biological diversity, endemism and number of endangered species, the Atlantic forest is considered to be one of the most threatened habitats in the world. Forest fragmentation can have profound effects on the flora and fauna with the most obvious outcome being a reduction in species diversity. Euglossine bees are the exclusive pollinators of ~ 700 species of orchids in the New World tropics and it is believed that such intricate associations are highly vulnerable to habitat loss. The objective of our study was to compare the abundance and diversity of euglossine bees within a highly fragmented area of the Atlantic forest and test the hypothesis that fragmentation and forest degradation affect species composition. Synthetic compounds resembling orchid fragrances or other natural substances were used to attract male bees. More than 3,600 bees belonging to at least 21 species were collected. Certain species such as *Euglossa analis* and *Eulaema nigrita* demonstrated distinct preferences for secondary undisturbed forest and disturbed forest, respectively; however, when all species were considered we found no clear difference in diversity or abundance across nine locations that varied in degree of disturbance. Based on these results, we proposed that the long-established fragments surrounding the Desengano area are functionally interconnected with regard to bee dispersal.

Correspondence:
Rua Salvador Correa, 139 - Centro
28035-310 - Campos dos Goytacazes - RJ
Phone number:+55 (22) 2726.2727
Fax: +55 (22) 2726.2720
www.isecensa.edu.br
e-mail: isecensa@isecensa.edu.br

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Ecologia behaviorista de abelhas "euglossine" da Mata Atlântica

Jacquelyn Lee Blackmer

Ph.D. em Entomologia pela Universidade Estadual de Ohio, Pesquisa em Agricultura em Ohio e Centro de Desenvolvimento, Wooster, Ohio, EUA; Atualmente - Entomologista em Pesquisa, EUA Centro de Pesquisa Agricultural Terra-Árida, Serviço de Pesquisa em Agricultura, Departamento de Agricultura dos Estados Unidos, Maricopa, Arizona.

Athayde Tonhasca Jr.

Ph.D. em Entomologia pela Universidade Estadual de Ohio, Pesquisa em Agricultura em Ohio e Centro de Desenvolvimento, Wooster, Ohio, EUA; Atualmente - Especialista em Invertebrados, Herança Natural Escocesa, Reino Unido.

Gilberto S. Albuquerque

Ph.D. em Entomologia pela Universidade de Cornel, Ithaca, Nova York, EUA; Atualmente - Professor Associado, Universidade Estadual do Norte Fluminense, Campos dos Goytacazes, RJ, Brasil

Resumo

Antes da colonização do Brasil, a Mata Atlântica estendia-se do Rio Grande do Norte ao Rio Grande do Sul. Atualmente, entretanto, a Mata Atlântica foi reduzida a apenas 8% de seu tamanho original e está altamente fragmentada. Por causa de sua diversidade biológica, endemismo e número de espécies em extinção, a Mata Atlântica é considerada um dos habitats mais ameaçados do mundo. A fragmentação da floresta pode ter conseqüências profundas na fauna e na flora tendo como resultado mais óbvio, a redução na diversidade das espécies. As abelhas "Euglossine" são coletoras exclusivas de pólen – 700 espécies de orquídeas nos trópicos do Novo Mundo. Acredita-se que associações intricadas estão altamente vulneráveis à perda do habitat. O objetivo de nosso estudo foi comparar a abundância e a diversidade das abelhas "Englossine" dentro de uma área altamente fragmentada da Mata Atlântica e testar a hipótese de que a fragmentação e a degradação da floresta afetam a composição das espécies. Compostos sintéticos assemelhando-se a fragrâncias de orquídeas ou outras substâncias naturais foram usados para atrair abelhas machos.

Mais de 3.600 abelhas pertencentes à no mínimo 21 espécies foram coletadas. Certas espécies tais como *Euglossa analis e Eulaema nigrita* demonstraram preferências distintas pela floresta secundária intocada e floresta tocada, respectivamente; entretanto, quando todas as espécies foram consideradas, nós não encontramos uma diferença clara na diversidade ou abundância de um lado a outro de nove locais que variavam em graus de transtorno. Baseados nesses resultados, nós propomos que fragmentos amplamente estabelecidos ao redor da área de Desengano são funcionalmente interconectados com respeito à dispersão da referida abelha.

Palavras-chave

orquídeas, abelhas, polinização, fragmentação, diversidade das espécies.

Correspondência:
Rua Salvador Correa, 139 - Centro
28035-310 - Campos dos Goytacazes - RJ
Telefone: +55 (22) 2726.2727
Fax: +55 (22) 2726.2720
www.isecensa.edu.br
e-mail: isecensa@isecensa.edu.br

Introduction

In pre-colonial times, the Atlantic Forest extended from Rio Grande do Norte to Rio Grande do Sul, an area covering more than 1,000,000 km^2 (Miller & Warren 1994, Tonhasca 2005). As late as 1832, Charles Darwin described it as "a forest which in the grandeur of all its parts could not be exceeded" (Miller & Warren 1994). It is now highly fragmented and reduced to only 8 % of its former size (Fig. 1). The states of Bahia and Espírito Santo have only 1-2 % of their rain forests remaining (Saatchi et al. 2001). One bright spot in this dismal trend is that approximately 20 % of the rain forest still stands in the state of Rio de Janeiro (Anonymous 2001). The Atlantic Forest consists of very diverse habitats from the mangroves and restingas along the coastal reaches, the dense forests inland, to the high altitudinal grasslands that exceed 1800 m in elevation (Tonhasca 2005). The mangroves and restingas are under great threat from coastal development and the ombrophilous dense forests have only remained because the steepness and remoteness of many of these areas make it difficult to harvest timber or practice agriculture.

Because of its biological diversity, endemism and number of endangered species, the Atlantic Forest is now considered to be one of the most threatened habitats in the world (Anonymous 2001). There are 34 such areas globally and they are referred to as ecological 'hot-spots'. These areas are designated as such because of their high degree of endemism of species that are under threat of extinction. Ecological 'hot-spots' constitute only 2.3 % of the earth's landmass but encompass 75% of all species on the planet. In Brazil, non-governmental organizations such as SOS Mata Atlântica, have put a spotlight on the need to conserve such areas. Probably the best-known reserve in Brazil protects the Golden Lion tamarin, *Leontopithecus rosalia*. In 1970, there were less than 200 of these beautiful monkeys surviving in the wild. The Golden Lion Tamarin Association was founded in 1992, and due to captive breeding programs at various zoos, there are now more than 1,500 in the wild. The woolly spider monkey, *Brachyteles arachnoides*, and several other species of mammals, birds and trees are similarly threatened in the Atlantic Forest. Many of these species are found nowhere else on earth. It has been estimated that there are approximately 250 species of mammals, 55 of which are endemic; 340 species of amphibians, 90 of which are endemic; more than 1,000 species of birds, 188 of which are endemic; and more than 20,000 species of trees, one-half of which are endemic to the Atlantic Forest (Fonseca et al. 1996, Stotz et al. 1996, Anonymous 2001, Tonhasca 2005). The tremendous biodiversity in the Atlantic forest is believed to be a consequence of the geographical and geological differences in the region. An important source of variation has to do with the latitudinal span from Rio Grande do Norte to Rio Grande do Sul. The second source is the change in elevation from sea level to more than 1,800 meters and thirdly, the considerable differences between inland forests and coastal forests (Anonymous 2001). As for the high degree of endemism, scientists believe that during the Pleistocene, conditions were much drier, resulting in the contraction of the forest areas in the Amazon and Atlantic Forest regions. This led to island refuges in which only a few species found favorable conditions. Over time, the long periods of isolation resulted in species differentiation (Anonymous 2001).

The flora of the Atlantic Forest is incredibly rich and the orchids are one of the most diverse groups. In the high mountain areas (> 1,000 m) of Macaé de Cima RJ, over 230 orchid species from 26 genera have been described (Miller & Warren 1994) and

it is estimated that more than 300 species in 70 genera probably exist in this region alone. Diversity at lower elevations is likely to be similar and it is at these lower elevations of the Atlantic Forest and elsewhere in the New World Tropics, that a group of approximately 700 species of orchids are exclusively pollinated by orchid bees. These orchids produce fragrances that attract males of one or a few species of bee. Their morphology is often very complex and when the bee arrives at the lip of the orchid to collect the essential oils, it accidentally acquires the orchid pollinia. When it visits the next flower, the pollinia are transferred from the bee to the orchid and pollination occurs. These orchids offer no nectar, pollen, or other rewards to the male bees.

Orchids require a microenvironment with specific humidity, light, heat and air movement – not to mention specific pollinators (Miller & Warren 1994). Once pollinated, the seeds within the fruit, while numerous, have no sustenance. For the seed to prosper, it must fall beside a symbiotic fungus. The fungus inoculates the seed and transfers to it sugars and nutrients essential for growth. Disturb the environment and any one of these vital elements (i.e., environment, bee, symbiotic fungi) might be eliminated.

The orchid bees belong to the order Hymenoptera, which includes the honeybee, ants and wasps (Borror et al. 1992). They are closely related to the honeybee, and fall within the tribe Euglossini. One of their most distinguishing features is their very long tongue, which is used for collecting nectars. Many species are beautifully colored, ranging from coppery green to blue and red. Other species resemble bumblebees and are covered with branched, colorful hairs. These bees are generally solitary, but some species exhibit rudimentary levels of sociality (i.e., communal nesting), and consequently have been studied in attempts to understand the evolution of social behavior in bees (Cameron

& Ramírez 2001). There are five genera in the Euglossini tribe – *Euglossa, Eulaema, Eufriesia, Exaereta* and *Aglae*. The latter two are cleptoparasitic on *Eulaema* and *Eufriesia*, respectively. *Exaereta* and *Aglae* bees wait until *Eulaema* and *Eufriesia* bees have constructed and provisioned their nests with nectar and pollen. They then usurp the nest and lay their own eggs inside. Currently there are approximately 200 known species of euglossine bees, mostly in the *Euglossa* genus. Approximately one-half of all described species occur in Brazil. These bees occur only in the New World moist tropics and are concentrated from southern Mexico to northern Argentina. However, a few species have made it into the USA (Minckley & Reyes 1996), and one species, *Euglossa viridissima*, is now well established in southern Florida (Skov & Wiley 2005, Pemberton & Wheeler 2006). Euglossine bees are important pollinators of at least 30 plant families, some of which are economically important crops.

The Euglossini engage in complex behaviors, some of which are not fully understood. Most of what we know is based exclusively on the males' behavior. They are attracted to aromatic compounds that are released from a variety of plants. Once the bee arrives at the odor source, it lands and begins biting the substrate, applying in the process a drop of fatty lipids released from its labial glands (Whitten et al. 1989, Cameron 2004). The non-polar fragrance compounds are dissolved in the lipids and then are mopped up by the bee's modified fore-tarsal brushes (Fig. 2). At this point, the bee will fly off the substrate and hover, during which it transfers the mixture from the front legs to the hind legs. Bees collect for several minutes before moving on to the next flower. It has been determined that species-specific blends of compounds are collected from a variety of hosts, and each hind tibia may contain 20-70 compounds (Eltz et al. 1999, 2003, 2006;

Pemberton & Wheeler 2006). The hind legs are highly modified for storage (Fig. 3 A & B), and it is here that the plant essential oils are absorbed and perhaps chemically altered. A variety of hypotheses have been formulated to explain why these bees collect fragrances. Dobson et al. (1969) proposed that the compounds are used to attract males to form leks (clusters of males), but while some males do cluster, there has been little direct support for this hypothesis. Later it was suggested that the aromatic compounds are transferred to the haemolymph from the hind tibia and modified chemically to produce a sex pheromone (Dressler 1982, Williams & Whitten 1983). It has also been suggested that the male presents these compounds to the female as a nuptial gift during courtship, or that they serve in anti-predator defense (Roubik 1989). More recently, Peruquetti (2000) suggested that these compounds are collected and used to enhance male aggregations, which then led to females being attracted to the congregation area. Roubik & Hanson (2004) suggested that these compounds are used to mark territories. Most recently, Eltz et al. (2005) provided some support for the idea that males present these compounds to the females as a sexual perfume during courtship, and that perhaps she is able to judge the quality of the male based on his odor makeup. Video links of this behavior can be found at http://www.uni-duesseldorf.de/MathNat/ Zoologie/eltz/index.htm.

Very little was known about these bees until the 1960s, when a group of orchidologists began testing synthetic baits as attractants (Dobson et al. 1969). The baits were highly attractive to males of many of the species. Since then the number of described species has risen rapidly and we have learned a great deal about their periods of activity, distributions, orchids they pollinate based on pollinia attached to them, as well as preference for certain baits. These compounds have helped to establish that species diversity and abundance are high in Central America and the Amazon basin, but we still know little about euglossine bees in the Atlantic Rain Forest (Nemésio 2003).

This research was conducted in the vicinity of Desengano mountain range, the largest remnant of Atlantic Forest in northern Rio de Janeiro State (*ca* 22,000 ha). Desengano is characterized by an edge of secondary vegetation that gradually shifts into a semi-deciduous primary tropical rain forest. The forest is surrounded by a heterogeneous landscape of cattle ranches, abandoned pasture, and forest fragments of varying sizes and stages of preservation. This fragmented landscape could impact the dispersal of important pollinators such as euglossine bees. Our objectives were to determine the effect of forest fragmentation on euglossine bee diversity and abundance and we hypothesized that these variables would have greatest values in contiguous forests and lowest in fragments. Furthermore, we expected that adjacent sites would be more similar in terms of bee diversity and abundance than more distant sites, and that bees would be less likely to disperse between fragments or sampling sites that were separated by pastures or open landscapes.

Methods & materials

Euglossine bee diversity and abundance (for more details see Tonhasca et al. 2002)

To determine euglossine bee diversity and abundance we sampled nine sites located within an area of *ca* 230 km^2 around the village of Sossego do Imbé (21°53'S, 41°48'W; Fig. 4), in the municipality of Santa Maria Madalena. Sites 1 to 4 (= forest) were located in well-developed, second-growth forest contiguous with the main forest. The

remaining sites were located in disturbed forest with high densities of *Attalea humilis*, a palm species common on regenerating slopes in this region of the Atlantic Forest (Souza et al. 2000). Sites 5 and 6 (= disturbed forest) were located in an area contiguous with the main forest that was comprised of a mosaic of second-growth and scrubby vegetation with no clear transition between vegetation types. Sites 7, 8, and 9 (= forest fragments) were forest fragments surrounded by pasture, and their sizes were 200, 156, and 14 ha, respectively. According to local people, these areas had not been disturbed for the last 20–30 years.

These nine sites were sampled over a 10-day period every two months during 12 field trips, which were conducted from September 1997 to July 1999. Sampling occurred twice during the first field trip and once thereafter. Usually three sites were sampled on the same day, and weather permitting, all sites were sampled during three consecutive days. Site 4 was not sampled during September 1998 and site 7 was added to the study beginning in May 1998. Sampling sites were set up 30–100 m inside the forest. At each site, *ca* 10 m of string was tied *ca* 1.5 m above the ground between two trees, with the string perpendicular to ground declivity the direction of prevailing winds. Six pieces of blotter paper (7 x 7 cm) were fixed at equal distances along the string, and each was impregnated with 2 ml of one of the following chemicals: cineole, eugenol, methyl salicylate, methyl cinnamate, skatole, and vanillin. The last three chemicals are crystals and were dissolved in ethanol until reaching saturation point. These synthetic products mimic natural components found mainly in orchid flowers and are collected by male euglossine bees (Dressler 1982). Fragrances were replenished every 1.5 h except for cineole, which was replenished every 30 min. These intervals were based on our previous observations of the chemicals' volatility. Male euglossine bees landing on the pieces of paper were captured with insect nets. Sampling started between 9:00 and 10:00 h and continued until 15:00 h, weather permitting. Voucher specimens were deposited at the entomological museum of the Universidade Estadual do Norte Fluminense.

To compare habitats (forest, degraded forest, and fragments) we conducted analyses of variance (ANOVAs) for the total number of bees collected and for each of the six most abundant species, which comprised 88.7 percent of all samples (see Results). For the analysis, we averaged the number of bees collected per field trip and transformed the data to log $(x + 1)$ to homogenize variances. Statistical power was expected to be low in these analyses because of the small number of replicates; therefore, we did not correct the experiment-wise error resulting from multiple tests (so that power would not be further reduced) and considered a critical level of a = 0.10.

To compare bee diversity in the three habitats, we plotted the log number of individuals against their rank to obtain estimates of species richness and evenness (Whittaker 1965). To compare the number of species among sites, we added the samples from the 12 field trips and used bootstrapping (Simon 1999) to generate 1000 samples from each site and obtain 95% confidence intervals for the number of species (richness). Diversity was expressed as the Simpson–Yule diversity index, *D,* calculated as $D = (1)/(S(Ni/NT)2)$, where Ni is the number of individuals in the i*th* species and NT is the total number of bees at each site (Magurran 1988). Therefore, *D* can be used as a measure of dominance and distribution of individuals among species in a community.

The community similarity between sites was estimated with the Morisita–Horn index, *MH,* calculated as $MH = (2$ *(ani* x

bni)/((*da* + *db*) x *Na* x *Nb*), where *ani* and *bni* are respectively the number of individuals in communities *a* and *b*, *Na* and *Nb* are the total number of individuals in each community, and *da* = (S*an2i*)/(*Na*), *db* = (S*bn2i*)/(*Nb*) (Southwood & Henderson 2000). *MH* approaches 1.0 when species in both communities have similar ranking and densities, and it is zero when there is no species overlap between communities.

Dispersal of Euglossine bees across an open landscape (for more details see Tonhasca et al. 2002, 2003)

This study was conducted at two sites, respectively 5 and 30 km from the town of Conceição de Macabu (22o05'S, 41o51'W), Rio de Janeiro State. The sites are part of a landscape of forest fragments, pastures and areas under regeneration surrounding the Desengano mountain range. Both sites were representative of the region: rolling rangeland with disturbed remnants of moist forest on hilltops. There were two adjacent forest fragments of at least 50 ha at each site, and euglossine bees were sampled simultaneously at two sampling locations on opposite sides of the pasture that divided the fragments. The spatial arrangement of the sampling stations at both sites is depicted in Figure 5. Direct routes from sampling station 2 to stations 1 and 5 at site 1 (Fig. 5 A) were blocked by a hill, likewise for routes from station 2 to sampling stations 3 and 4 at site 2 (Fig. 5 B).

The sampling stations were set up 10 to 30m inside the forest in a manner similar to that described above. Each piece of blotter paper was impregnated with 2 ml of one of the following chemicals: cineole, methyl cinnamate, skatole and vanillin. Fragrances were replenished every 1.5 h except for cineole, which was replenished every 30 min.

Euglossine bees attracted to the fragrances were captured with insect nets and marked on the thorax and on one of the forewings with Outliner marking pens (Sakura Color Products, Japan) or acrylic paint (Hobby Cores, Brazil) and released. A different color was used at each station. Euglossine bees are very adept at rubbing off paints and tags from their bodies, so we carefully placed marks in the scuto-scutellar suture (Stephen et al. 1969). Although most of the markings had been removed in recaptured bees, remains of paint could be seen in the suture or on the wing. Recaptured bees were released if they had been previously marked at the same sampling station, marked for a second time and released if they originated from a station in the same fragment, or retained if they came from the opposite side of the pasture. Sampling started between 9:00 and 10:40 h, and continued until. 14:00 to 15:00 h.

Results and discussion

Euglossine bee diversity and abundance (for more details see Tonhasca et al. 2002)

We collected 3653 euglossine bees representing at least 21 species (Table 1). Bees classified as *Euglossa cordata* may actually have belong to more than one species (R. Dressler, pers. comm.). This sampling nearly doubled the number of previously identified euglossine bee species in the state of Rio de Janeiro. The ANOVAs indicated no significant differences in the total number of bees or in the number of the dominant species except for *Eulaema nigrita* ($F = 4.37$, $P = 0.06$, df = 2, 6). *Eulaema nigrita* was the most common species, but it was concentrated (42.5% of the total) in the two disturbed forest sites (Table 1). Rank abundance curves for the three habitats were topologically equivalent, indicating similar diversity. Richness estimates generated by

bootstrapping demonstrated that random samples of euglossine bees were likely to contain more species at site 2, whereas all the other sites had equivalent richness. The diversity index accentuated the differences among sites, with lower values for sites 1, 5, and 6. The strong dominance by *E. nigrita* was responsible for the considerably lower diversity index values at sites 5 and 6. A similar pattern occurred at site 1, where the second most abundant species, *E. cordata*, comprised a relatively larger proportion of the total number of individuals compared to the dominant species of the remaining sites. Similarity decreased with increasing distances between sites, but values of the Morisita–Horn index were relatively high for most site combinations. The species list in Table 1 clearly shows the similarity among sites in relation to euglossine bees. Excluding the rare species, species composition and their relative abundance were alike at all sites. These results demonstrated that, as a whole, the euglossine bee community in the three habitats was essentially the same. There was greater variability among sites at the species level, and in two cases (*E. nigrita* and *Euglossa analis*) differences among sites are likely to be associated with habitat types. Despite the lack of significance, none of the 178 *E. analis* were collected from degraded forest or the two smallest fragments, suggesting that this species is mostly restricted to large, relatively undisturbed forests. For most other species, densities were comparatively high at specific sites regardless of the habitat type, such as *Euglossa pleosticta*. The accumulation of individuals at particular sites was not the result of preference throughout the season, but rather originated from momentary local increases in the number of bees. Considerable seasonal variation in euglossine bee abundance is expected (Roubik 2001), and in this study it probably was caused in part by weather patterns. Although the

number of bees per site or field trip was not correlated with temperature or relative humidity, the field trips with the fewest dominant species (and lowest total number of bees) were the coldest; average temperatures during sampling were below 22°C. Considerable variation in species capture over the seasons supports Roubik's (2001) view that short-term census of euglossine bee populations can be misleading; although *E. cordata* and *Euglossa securigera* were among the dominant species, their numbers during the first year of sampling corresponded to only 3.3 and 8.7 % of the total, respectively.

Although it is reasonable to assume that similar habitats might have similar bee communities, to consider sites as habitat replicates is not appropriate because of differences in site size, vegetation type and density, degree of disturbance, and topography. Reduced or improper replication, and consequently low power, are among the most serious limitations of experiments with forest fragmentation (Crome 1997). Despite these caveats, our results revealed some aspects of euglossine bee responses to habitat quality. Forest fragmentation has been associated with population decline of euglossine bees (Powell & Powell 1987) and native bees in general (review in Cane 2001), but we found no clear association between abundance and richness of the euglossine bee community and habitat type. The total number of bees was lowest in the smallest fragment, but this result was determined mostly by the relative low abundance of two species. Although the results suggest that numbers of some euglossine bees may decline in fragments or disturbed forests, most species appear to respond to specific and ephemeral local conditions. Euglossine bee populations have relatively high stability, but substantial seasonal fluctuations occur possibly because of nesting and adult emergence patterns (Roubik & Ackerman

1987, Roubik 2001). Brown (1991) proposed that many insects could be used as indicators of habitat quality, and Peruquetti et al. (1999) attributed this role to *E. nigrita* because of its high abundance in disturbed areas (Rebêlo & Cabral 1997, Peruquetti et al. 1999). Our data suggest that *E. nigrita* and *E. analis* are the most likely candidates for indicator species of disturbed and primary (or less disturbed) areas, respectively. Euglossine bees are able to locate isolated food and fragrances and use resources far away from their reproductive sites due to their exceptional flight capacity and plant fidelity (Dressler 1968, Janzen 1971, Janzen et al. 1982). Bee numbers and species composition in different sites are probably similar because their capacity to disperse and locate fragrances compensate for possible negative effects of forest isolation or forest degradation.

Dispersal of euglossine bees across an open landscape (for more details see Tonhasca et al. 2002, 2003)

The number of bees collected, marked and recaptured at sites 1 and 2 is indicated in Tables 2 and 3. The five identified species that moved across fragments are among the most abundant of the 21 species recorded in our previous study. There were $5.6 \pm 4.8\%$ (mean $\pm$ 1 SD) recaptures from the 17 samples that resulted in movements across clearings. We measured temperature and relative humidity and estimated wind speed with the Beaufort scale every 30 min at each sampling station and none of the variables could be associated with the absence of bee traffic across open areas on 10 and 15 April. On the other hand, the crossing of only one bee across the pasture on 20 January can be attributed to temperature, which was the highest of all sampling dates (median of 30.6 °C). Euglossine bee flight activity is sharply reduced when temperatures reach 30 °C (pers. obs.)

Powell & Powell (1987) monitored euglossine bees before and after the isolation of forest patches at the Biological Dynamics of Forest Ecosystem Project in the Amazon and observed a decline in the number of bees in isolated fragments, supposedly because of their inability to cross open areas. This study is frequently cited as evidence for the negative effect of fragmentation on euglossine bee dispersal and consequently their pollinating services (Kearns et al. 1998, Power 1996, Turner 1996, but see Cane 2001). However, Powell & Powell's study was conducted shortly after fragmentation occurred, which could have affected temporarily the local euglossine bee populations (Cane 2001.). In a later study in the same area, Becker et al. (1991) did not find a significant reduction in euglossine bee visits to fragments except for the smallest fragments. Euglossine bees, particularly the large *Eulaema* spp., have exceptional flight capacity (Janzen 1971). Even the smaller *E. cordata* was able to travel 4 km over non-forested areas in a 30-d period (Raw 1989). Because of their dispersal capacity, euglossine bees can utilize resources far away from their reproductive sites (Janzen 1981, Janzen et al. 1982). Our observations indicate that cleared areas do not prevent some euglossine bee species from moving between patches of Atlantic Forest in search of fragrances, even to and from very small forest fragments. In fact, more bees were recaptured across openings than inside the forest, although it was not possible to evaluate the significance of these results because samples were conducted at different sites and times of the year. Nonetheless, it is possible that the vegetation inside the forest makes it more difficult for bees to locate fragrances. Our results reveal two additional characteristics of euglossine bee dispersal. First, steep hills between sampling sites did not prevent bees from locating baits on the opposite side from where they were released.

Second, simultaneous recaptures at most stations indicate that bees locate baits regardless of their relative position in the landscape. This suggests that possible variations in fragrance concentration caused by the irregular terrain of the Atlantic Forest do not affect the ability of bees to find the fragrance source.

Our observations indicate that bees are able to move within and between forest patches, at least at the spatial scale of this study. Based on these results, we propose that the long-established fragments surrounding the Desengano area are functionally connected with regard to euglossine bee dispersal.

Conclusions

In 1988, the Atlantic Rainforest was declared a national heritage and the government prohibited any further cutting or clearance. However, in the six years from 1990-1995, more than 500,000 ha were destroyed (http://www.iracambi.com). Satellite images from SOS Mata Atlântica Foundation suggest higher rates of deforestation from 1996-2000 than in previous years. The most intense deforestation in the state of Rio de Janeiro is now concentrated in some municipalities of Angra dos Reis, Carmo, Santa Maria Madalena, and Campos dos Goytacazes (Anonymous 2001). Although we found few differences in euglossine bee diversity and abundance in the fragmented landscape of Desengano, there are certainly other species of plants and animals that have been affected by habitat loss in this region. For posterity these remaining remnants of the Atlantic Forest should be preserved. In addition to the incredible diversity of flora and fauna that would be preserved, these areas are perhaps the best protector of natural springs and serve as an important watershed area for the region around Campos dos Goytacazes.

Acknowledgements

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Table 1. Number of bees collected at nine sampling sites (Fig. 4) in the Desengano region, Brazil, during 12 field trips from September 1997 to July 1999. From Tonhasca et al. 2002.

Habitat type:	Secondary forest				Disturbed forest		Forest fragments		
Species Site:	1	2	3	4	5	6	7	8	9
Euglossa nigrita Lepeletier	100	100	66	100	143	292	89	76	56
Euglossa cordata (L.)	140	95	112	52	34	125	106	99	74
Euglossa sapphirina Moure	32	140	94	43	62	61	51	5	17
Eulaema cingulata (F.)	56	95	43	40	15	37	38	41	38
Euglossa securigera Dressler	6	26	23	6	18	74	30	56	24
Euglossa chalybeata Friese	9	51	20	10	22	26	34	19	0
Euglossa analis Westwood	17	21	59	40	0	0	41	0	0
Euglossa pleosticta Dressler	1	17	9	2	7	17	2	25	5
Euglossa ignita F. Smith	0	24	1	0	0	1	2	7	10
Euglossa cf. *deceptrix*	1	7	3	1	0	12	4	6	1
Euglossa fimbriata Rebêlo & Moure	1	4	2	0	2	10	0	1	0
Euglossa viridis (Perty)	0	3	0	2	1	1	2	1	1
Euglossa cybelia Moure	2	3	0	1	2	1	1	0	0
Euglossa cf. *nigropilosa*	0	2	3	3	2	0	0	0	0
Exaerete smaragdina (Guérin)	0	1	0	0	0	0	0	1	5
Eufriesea brasilanorum Friese	1	0	0	0	0	0	3	0	0
Eufriesea ornata (Mocsáry)	0	1	0	0	0	0	1	1	0
Euglossa truncata Rebêlo & Moure	0	0	0	0	0	0	0	0	1
Eulaema meriana (Oliver)	0	1	0	0	0	0	0	0	0
Euglossa townsendi Cockerell	0	0	0	0	0	0	0	0	1
Eufriesea surinamensis (L.)	0	0	0	0	0	0	0	1	0
Total	366	591	435	300	308	677	404	339	233

Table 2. Summary of euglossine bees movement between sampling statio ns at site 1 (Fig. 5A).
The number of marked bees at station 4 on 1 4 April and at station 5 on 15 April were 73 and 52, respectively.
Asterisks indicate bees marked on the previous day. From Tonhasca et al. 2003.

Date	Capture Site	Number marked	Recapture site	Species and number recaptured
20 Jan	1	36	2	*Euglossa* sp. 1
	2	112	3	*Eulaema nigrita* 1
	3	67	4	*Euglossa* sp. 3
	4	67	3	*E. nigrita*, 2 *Euglossa* sp. 1
21 Jan	1	34	2	*Euglossa cordata* 1
			4	*E. cordata* 1
	2	84	1	*E. cordata* 1
			3	*E. cordata* 1 *E. nigrita* 1 *Euglossa securigera* 1
			4	*E. cordata* 1
	3	64	4	*E. cordata* 8
	4	59	2	*E. cordata* 2
			3	*E. cordata* 4
14 Apr	1	54	2	*E. nigrita* 1
			4	*E. nigrita* 1
	2	91	1	*Euglossa* sp. 1
			4	*Euglossa* sp. 1 *E. nigrita* 1
			5	*E. nig rita* 1
	5	47	2	*E. nigrita* 1
15 Apr	1	19	2	*E. nigrita* 1*
			4	*E. nigrita* 1*
			5	*E. nigrita* 2*
	2	69	4	*Euglossa* sp.1*
			5	*E. nigrita* 1*
	4	67	2	*Euglossa* sp. 1*
			5	*Euglossa* sp. 1*

Table 3. Summary of euglossine bees movement between sampling stations at site 2 (Fig. 5B).
Asterisks indicate bees marked on the previous day. There were 138 bees marked on 10 April, but there were no same-day recaptures. From Tonhasca et al. 2003.
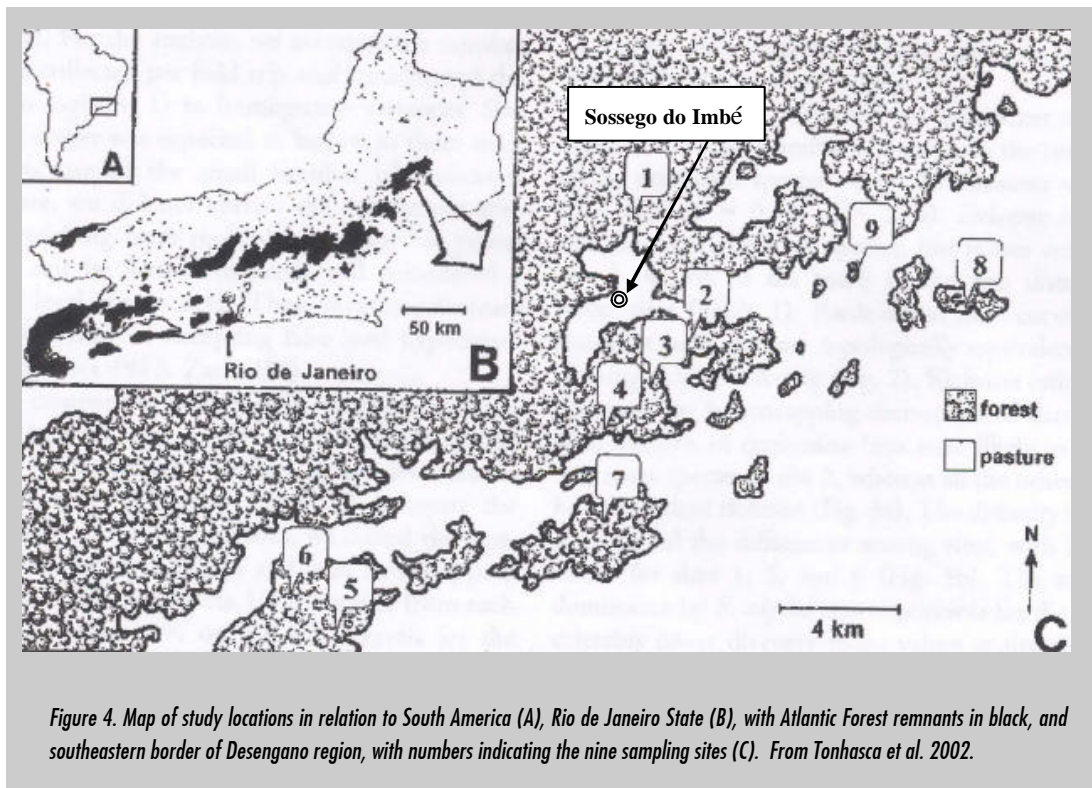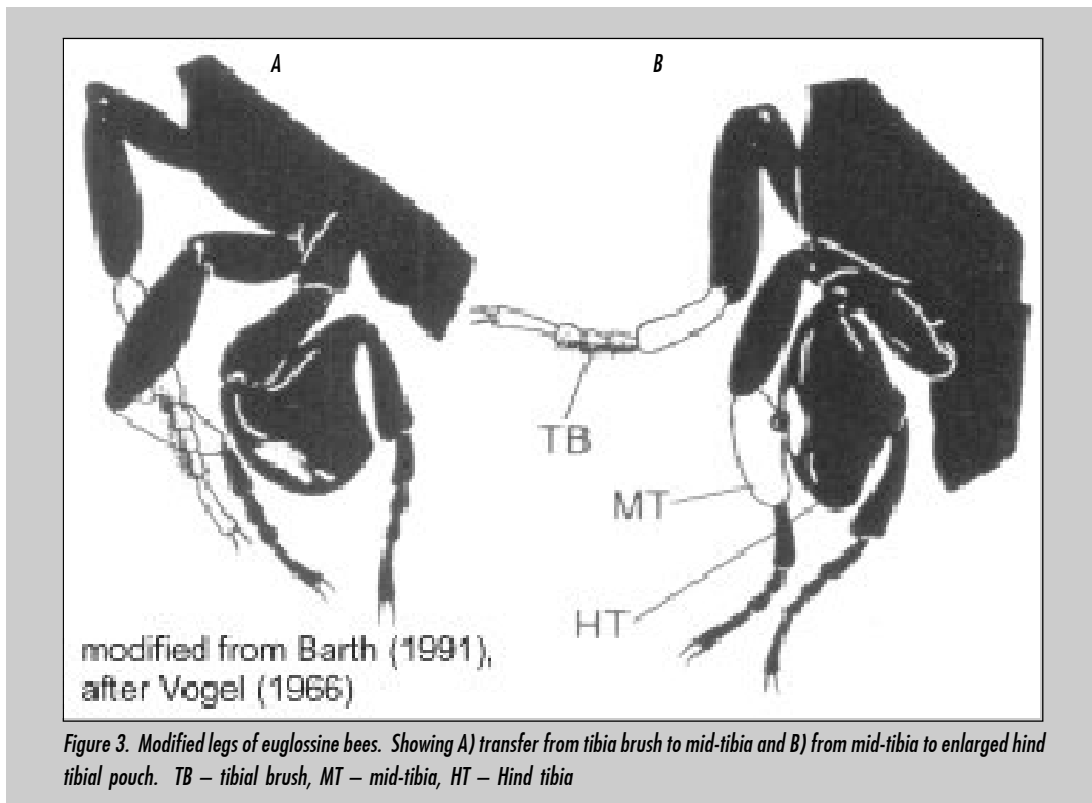
Date	Capture Site	Number marked	Recapture site	Species and number recaptured
19 Jan	1	84	3	*Euglossa cordata* 5 *Euglossa analis* 1
	2	97	1	*E. cordata* 4
			3	*E. cordata* 4
			4	*E. cordata* 2
	3	44	1	*E. cordata* 3
			4	*E. cordata* 1
	4	37	3	*E. cordata* 1
11 Apr	1	24	2	*Euglossa sapphirina* 1*
			3	*E. sapphirina* 1*
			4	*E. cordata* 1
	2	25	3	*E. sapphirina* 1
	3	31	4	*Euglossa* sp.1 *E. sapphirina* 1*
	4	26	2	*E. analis* 1
			3	*E. sapphirina*, 1 *E. sapphirina* 2*
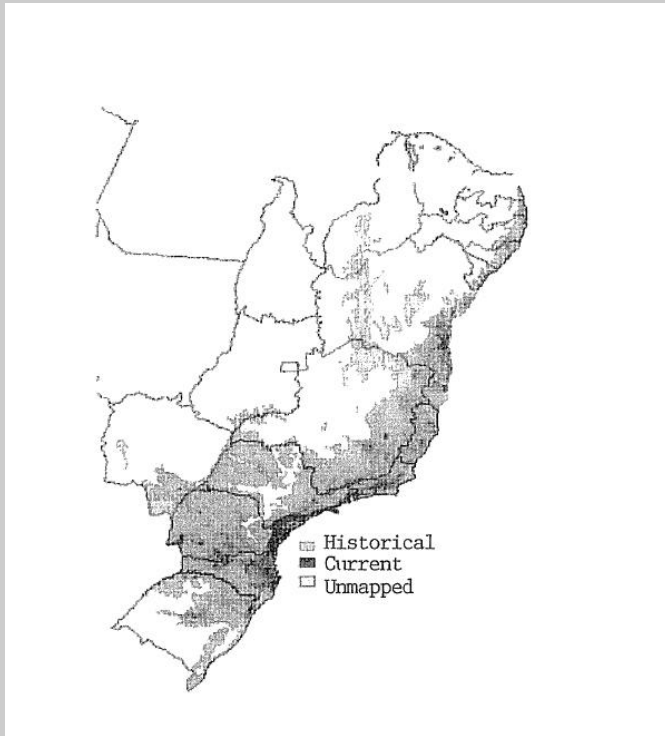
Figure 1. Pre-colonial and current Atlantic forest domain (from Critical Ecosystem Partnership Fund 2001)

Figure 2. Scanning electron micrograph (SEM) of the tibial brushes on the fore legs of a euglossine bee [from http://www.uni-dusseldorf.de/MathNat/ Zoologie/eltz/morphology.htm]

Figure 3. Modified legs of euglossine bees. Showing A) transfer from tibia brush to mid-tibia and B) from mid-tibia to enlarged hind tibial pouch. TB – tibial brush, MT – mid-tibia, HT – Hind tibia

Figure 4. Map of study locations in relation to South America (A), Rio de Janeiro State (B), with Atlantic Forest remnants in black, and southeastern border of Desengano region, with numbers indicating the nine sampling sites (C). From Tonhasca et al. 2002.
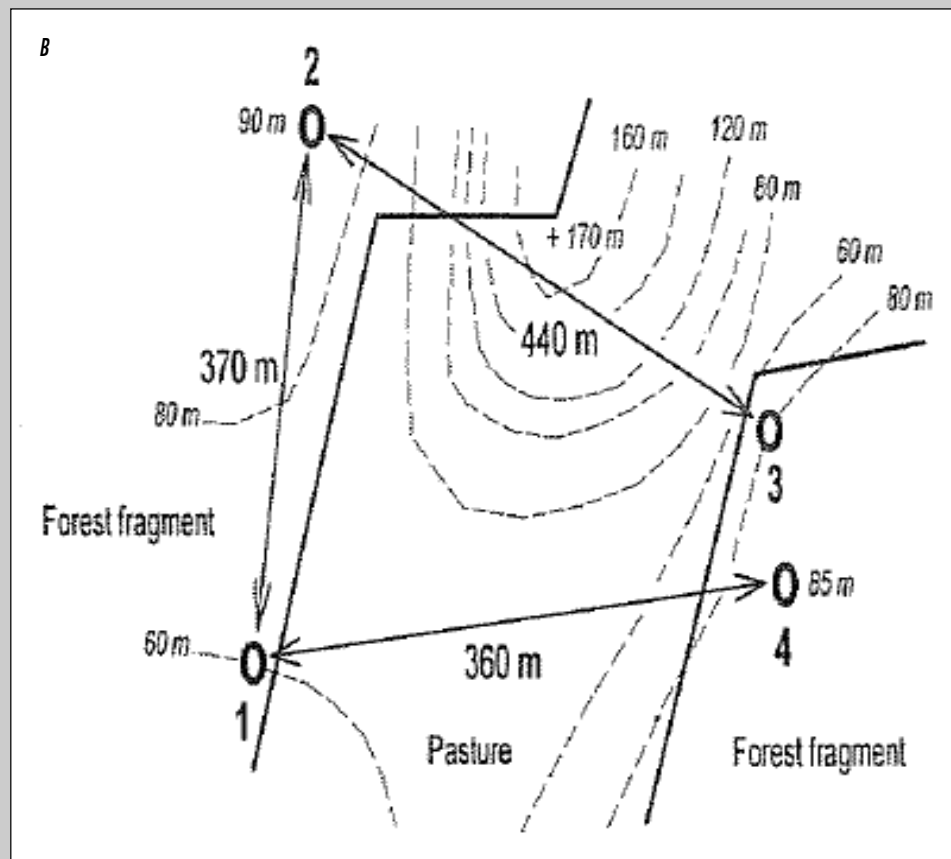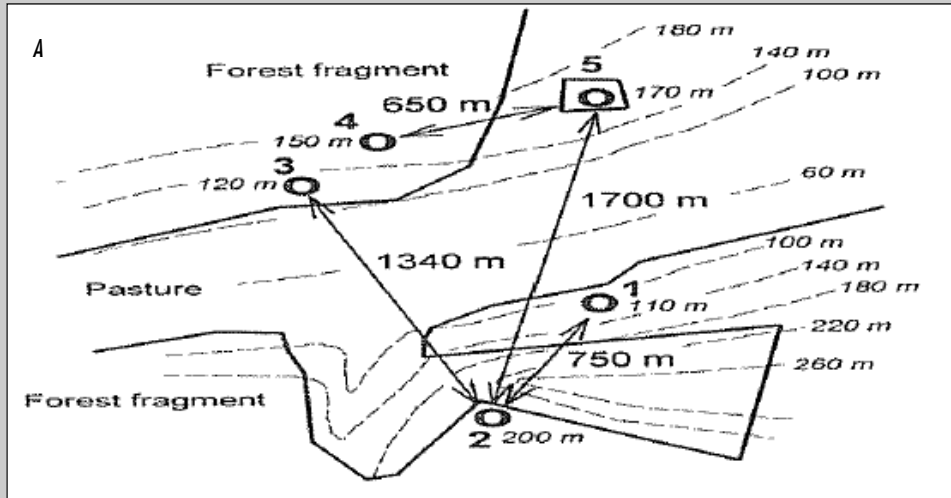
Figure 5. Diagram of the sampling stations (1-5) at site 1 (A) and (1-4) at site 2 (B) in the Desengano region. Distance among sites is indicated in meters. Dashed lines are contour lines and smaller-sized numbers in italics are elevations. From Tonhasca et al. 2003.