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## ORCHID BEES (HYMENOPTERA: APIDAE) IN THE COASTAL FORESTS OF SOUTHERN BRAZIL: DIVERSITY, EFFICIENCY OF SAMPLING METHODS AND COMPARISON WITH OTHER ATLANTIC FOREST SURVEYS

VANESSA C. MATTOZO<sup>1</sup>  
LUIZ R.R. FARIA<sup>1,2</sup>  
GABRIEL A.R. MELO<sup>1,3</sup>

### ABSTRACT

Surveys of orchid bees at the Brazilian Atlantic forest have been restricted to a few regions, making difficult to understand latitudinal patterns of distribution and diversity of these bees. For this reason we sampled the euglossine fauna at Atlantic forest areas at the coastal region of São Paulo (Sete Barras, Faz. Morro do Capim: SP3) and state of Paraná (Antonina, Reserva Natural do Rio Cachoeira: PR3), in southern Brazil. In PR3, we also evaluated the efficiency of collecting methods for sampling the fauna, comparing bait traps with direct collecting using entomological nets on fragrance baits. The diversity and abundance of bees was very low: we caught only 39 males of eight species in SP3 (*Euglossa iopocila*, *Euglossa roderici*, *Eulaema nigrita*, *Euglossa annectans*, *Eulaema cingulata*, *Euglossa pleosticta*, *Euglossa viridis* and *Exaerete smaragdina*) and 254 males of six species in PR3 (*Euglossa iopocila*, *Euglossa annectans*, *Euglossa stellfeldti*, *Euglossa roderici*, *Euglossa pleosticta* and *Eulaema nigrita*). Comparing the sampling methodologies, use of insect nets on fragrance baits (six species; 221 specimens) was more efficient than bait traps (three species; 33 specimens). When comparing the faunas of these two areas with other surveys at the Atlantic forest sites, through a DCA analysis, we found that the two surveys presented in this paper were placed relatively close to each other, but apart from the other sites analyzed, not clustering with the southernmost survey at the subtropical Atlantic forest of Rio Grande do Sul or with the remaining surveys carried out at northern lowland sites of this biome.

KEY-WORDS: Euglossina; *Euglossa*; Atlantic forest; Euglossini; orchid bees.

### INTRODUCTION

The subtribe Euglossina (Hymenoptera, Apidae) includes bees with extraordinary long glossa

and, in most cases, metallic integument (Dressler, 1982; Cameron, 2004). About two hundred species are known, in five genera (Moure *et al.*, 2007). This group presents a primarily Neotropical distribution,

1. Laboratório de Biologia Comparada de Hymenoptera, Departamento de Zoologia, Universidade Federal do Paraná. Caixa Postal 19.020, 81531-980, Curitiba, PR, Brasil.

2. Current Address: Departamento de Ciências Agrárias e Biológicas, Centro Universitário do Norte do Espírito Santo (CEUNES/UFES). BR-101 Norte, Km 60, 29932-540, São Mateus, ES, Brasil.

3. Corresponding author. E-mail: garmelo@ufpr.br

from southern Brazil and northern Argentina (Wittmann *et al.*, 1988; Pearson & Dressler, 1985) to southern United States (Minckley & Reyes, 1996; Pemberton & Wheeler, 2006), and is more diverse in warm tropical forest areas (Moure, 1967; Roubik & Hanson, 2004). Male euglossine bees collect aromatic compounds on flowers and other non-floral resources, storing them in their hind tibia (Dressler, 1982; Cameron, 2004). Many authors agree that these substances play a role in the reproductive biology of the bees, but the process is not entirely understood (see Eltz *et al.*, 1999; Cameron, 2004). When it was clear that males were attracted to man-made aromatic compounds, it was possible to develop sampling methodologies specifically for this group of bees (Dodson *et al.*, 1969), increasing the number of known species and allowing studies on local fauna composition (see Ramírez *et al.*, 2002; Roubik & Hanson, 2004).

In the Brazilian Atlantic forest, euglossine surveys are geographically restricted and mostly performed at small fragmented areas (e.g. Sofia & Suzuki, 2004; Nemésio & Silveira, 2007a). In fact, only a few pristine areas were assessed for orchid bee fauna (e.g. Bonilla-Gómez, 1999; Nemésio & Silveira, 2006). Also, it is remarkable the latitudinal decrease in number of orchid bee species found in studies at the subtropical Atlantic forest of Rio Grande do Sul (five species; Wittmann *et al.*, 1988) when compared to more northern sites in southeastern Brazil (at least twenty species; see Bonilla-Gómez, 1999; Tonhasca *et al.*, 2002). Refining this pattern of latitudinal variation in richness seems to be an important task to improve our knowledge of the biogeography of this group of bees.

No standardized surveys have been carried out on coastal areas of southern São Paulo and Paraná states. In Paraná, two previous surveys have been conducted, both at small forest fragments in inland sites in northern Paraná, where the original vegetation has been heavily fragmented (Sofia & Suzuki, 2004; Sofia *et al.*, 2004). These two studies contributed to our knowledge of euglossine bee diversity along the inland portion of the Atlantic forest, but their results cannot be extrapolated to coastal Atlantic forest sites and therefore are insufficient to fill the gap of surveys existing between the subtropical Atlantic forest of Rio Grande do Sul and southeastern Brazil. Not only the Atlantic forest is heavily fragmented but it also possesses high levels of species richness and endemism (Fonseca, 1985) with expressive risk of short term extinctions (Mittermeier *et al.*, 1998). Particularly in the state of Paraná, the forest covering the coastal lowlands and the adjacent mountain slopes is one of the most representative remnants of the Brazilian Atlantic forest (Ferretti & Britez, 2006).

Previous results point out that some species (e.g. *Euglossa analis* Westwood, 1840 and *Euglossa sapphirina* Moure, 1968) (see Nemésio & Silveira, 2006; Ramalho *et al.* 2009) are found only in non-disturbed natural habitats, while other species (e.g. *Eulaema nigrita* Lepeletier, 1841) (Morato *et al.*, 1992; Peruquetti *et al.*, 1999; Nemésio & Silveira, 2007a) could be used as indicators of highly disturbed areas. In fact, surveying these bees in areas under different degrees of anthropogenic disturbance would also generate data related to the use of orchid bees as indicators of environmental quality.

It is also important to consider here the two main collecting methodologies deployed in euglossine surveys: the standard methodology, in which the males attracted to scent lures are directly captured with an insect net, versus bait traps, which retain the males attracted to scent baits placed inside them. Morato (1998) claims that the two methods are not comparable, and moreover that they could provide distinct results. Previous results suggest that bait trapping would be less effective than net collecting (Nemésio & Morato, 2004, 2006; Justino & Augusto, 2006), but more studies about the subject are necessary.

The present paper aims to assess the orchid bee fauna at two coastal areas of Atlantic forest in southern Brazil, including a large remnant of well-preserved Atlantic forest in Paraná, and to evaluate the efficiency of bait traps compared to direct collecting using entomological nets at one of the study sites. We also compare the composition of the orchid bee fauna in the studied region with data previously presented in other studies on the orchid bee assemblages in the Brazilian Atlantic forest.

## MATERIAL AND METHODS

### Study sites

The Reserva Natural do Rio Cachoeira (herein called PR3) is located in the APA of Guaraguêçaba, municipality of Antonina (48°39'W; 25°19'S; elevation: 54 m), state of Paraná, southern Brazil. The reserve is maintained by the Sociedade de Pesquisa em Vida Selvagem e Educação Ambiental (SPVS) and has about 8,700 ha, most of them covered with pristine Atlantic forest (Ferretti & Britez, 2006; Liebsch *et al.*, 2007). In this region, the main climate is the Cfa of Köppen, with mean annual precipitation ranging to 3,000 mm (Maack, 1981). In PR3, two similar sites inside the forest were chosen along the "Pinheiro" track. In one of these sites, insect nets were used for collecting bees (active sampling), while bait traps were used in the other site (passive sampling) (see below).

The Morro do Capim farm (herein called SP3) is located in the municipality of Sete Barras (47°58'W; 22°22'S; elevation varying from 27 to 150 m), in the "Vale do Ribeira" region, state of São Paulo, south-eastern Brazil. Morro do Capim has 644 ha (444 ha of old secondary forest and 200 ha of pastures). Mean temperatures in the dry (July to September) and wet seasons (October to June) vary, respectively, between 12-22°C and 20-32°C (Marchi, 2008). One sample site was chosen inside the forest near the border of the larger fragment of SP3. Only active sampling at scent lures was carried in SP3.

### Sampling

In PR3, four aromatic compounds were offered to bees: 1,8-cineol, eugenol, methyl salicylate and vanillin. Sampling was carried out from 09:00 to 15:00 h at least once a month, totalizing thirteen samples per site, during one year (December, 2006 to November, 2007). In SP3, five scents were utilized to attract bees: the four compounds utilized in PR3 and  $\beta$ -ionone. Dial sample effort were the same performed in PR3. In SP3, collecting took place during seven separate days between September, 2008 and April, 2009.

In the active collecting sites (one at PR3 and one at SP3), pads of absorbent paper were soaked up with the attractive substances (one compound per pad) and offered to bees hung from plant branches at about 1.5 m above ground and distant at least 2 m from each other. Regarding the passive collecting, bait traps were made with plastic bottles (detailed below). Traps were also hung from plant branches and exposed in the site from 09:00 to 15:00 h. In both situations, bees attracted to those lures were killed with ethyl acetate and kept apart in paper bags. Time of day and substance were recorded, and for identification the bees were mounted in entomological pins.

Bees were identified with help of taxonomic keys (Rebêlo & Moure, 1996; Faria & Melo 2007; Nemésio, 2009) and by comparison with specimens previously identified. All the bees collected are deposited in the "Coleção Entomológica Pe. Jesus Santiago Moure" (DZUP), Department of Zoology, Universidade Federal do Paraná, Curitiba, Brazil.

### Scent traps

Bait traps used were primarily based on models proposed by Campos *et al.* (1989) and Nemésio &

Morato (2006). The trap consisted of plastic bottles with 29.5 cm of height and 9.5 cm of diameter. In each bottle, two pairs of holes, one pair opposite to the other, were made in different heights and with different diameters: the first pair had two opposite holes with 2 cm of diameter, made at 8 cm from the upper end; in the other pair, the holes, also in opposite sides of the bottle, were placed at 14 cm from the upper end of the trap. The orientation of the holes made possible to have openings in four different directions. Funnels cut from the upper end of small plastic bottles were glued to each hole, leaving a short neck (*ca.* 0,5 cm) projecting inside the trap as a landing platform for the approaching bees (see Nemésio & Morato, 2006). Inside the traps, paper pads, soaked with the specific aromatic compound, were offered to bees hanging at 7 cm of the upper end.

### Comparison of the orchid bee fauna in the Brazilian Atlantic forest

In order to compare the orchid bee fauna resulting from our surveys at the coastal region of São Paulo and Paraná with surveys in other Brazilian Atlantic forest sites, a presence-absence matrix was built with original data presented here and with data previously published in the following studies (in square brackets are the abbreviations used in the DCA analysis): Wittmann *et al.* (1988) [RS1]; Rebêlo & Garófalo (1991) [SP1]; Neves & Viana (1997) [BA1]; Rebêlo & Garófalo (1997) [SP2]; Bonilla-Gómez (1999) [ES1]; Peruquetti *et al.* (1999) (data only from the survey at Viçosa) [MG1]; Bezerra & Martins (2001) [PB1]; Tonhasca *et al.* (2002) [RJ1]; Viana *et al.* (2002) [BA2]; Nemésio (2004) [MG4]; Sofia & Suzuki (2004) [PR1]; Sofia *et al.* (2004) [PR2]; Nemésio & Silveira (2006) [MG2]; Nemésio & Silveira (2007a) [MG3]; Aguiar & Gaglianone (2008) [RJ2]; Farias *et al.* (2008) [PB2]; Nemésio (2008) [MG5]; Ramalho *et al.* (2009) [RJ3]. The surveys from the present study, in Morro do Capim and Reserva Natural do Rio Cachoeira, were indicated by the codes SP3 and PR3, respectively. The selection of works to the DCA analysis was based on criteria suggested by Sydney *et al.* (2010), with a slight modification regarding the sampling time during the year: (i) euglossine male bees collected using scent baits, either through traps and/or active collecting; (ii) at least three compounds used, one of them being, necessarily, cineole or eucalyptol; (iii) fieldwork for at least eight months, with monthly, fortnightly or seasonal samples, but comprising necessarily the rainy season. When a paper

presented results obtained in different but nearby sites, data were assembled to represent a single sample. This procedure is justified considering our aim of investigating broader faunistic patterns along the Atlantic forest.

Unidentified species, presented in a given work as “sp.”, were not taken into consideration. Also, some species here are referred under a name different from that used in the original papers: *Euglossa chalybeata* Friese, 1925 was considered as *Euglossa iopoecila* Dressler, 1982, following Faria & Melo (2007), and *Euglossa despecta* Moure, 1968 was considered as *Euglossa violaceifrons* Rebelo & Moure, 1996. The taxonomical arrangement considered when constructing the matrix (and also in the discussion section) follows Moure *et al.* (2007). The data matrix was submitted to a detrended correspondence analysis (DCA) using the software Past (Hammer *et al.* 1999-2011). Ordination techniques were used since they allow the reduction of the information related to the differences among areas, with respect to its species composition, to a few variation axes, which facilitates the visualization and interpretation of the results (see Sydney *et al.*, 2010).

## RESULTS

### Reserva Natural do Rio Cachoeira (PR3)

A total of 254 males belonging to six species were collected in the two sites (Table 1) during 156 baiting hours (78 hours of active insect netting and 78 hours of passive sampling). *Euglossa iopoecila* was the most abundant species, followed by *Euglossa annectans* Dressler, 1982. Two species, *Euglossa pleosticta* Dressler, 1982 and *Eulaema nigrita*, contributed with only one specimen each.

Comparing the collecting methodologies, 221 specimens were actively collected with insect nets at scent lures (87%), while only 33 bees (13%) were captured in the bait traps. Three of the six species collected with hand nets in the area were not caught in bait traps: *Eulaema nigrita*, *Euglossa pleosticta* and *Euglossa stellfeldi* Moure, 1947. No species was sampled solely in the bait traps.

Eugenol was the most attractive scent, with about 54% of the male bees collected on it, followed by 1,8-cineole (31.8%). Vanillin attracted only *Euglossa iopoecila*, while *Euglossa roderici* Nemésio, 2009

**TABLE 1:** Orchid bee species captured in the Reserva Natural do Rio Cachoeira site (Antonina, Paraná, Brazil). Values in the cells represent the number of specimens by collecting method and type of bait scent. C: 1,8-cineol; E: eugenol; S: methyl salicylate; V: vanillin.

Species	Bait traps				Direct collecting				Total
	C	E	S	V	C	E	S	V	
<i>Euglossa (Euglossa) pleosticta</i>	0	0	0	0	1	0	0	0	1
<i>Euglossa (Euglossa) roderici</i>	0	0	11	0	0	0	14	0	25
<i>Euglossa (Glossura) annectans</i>	1	3	0	0	4	25	0	0	33
<i>Euglossa (Glossura) iopoecila</i>	8	9	0	1	65	81	7	3	174
<i>Euglossa (Glossurella) stellfeldi</i>	0	0	0	0	1	19	0	0	20
<i>Eulaema (Apeulaema) nigrita</i>	0	0	0	0	1	0	0	0	1
Total per method/scent	9	12	11	1	72	125	21	3	
Grand total individuals		33				221			254

**TABLE 2:** Orchid bee species captured in the Morro do Capim site (Sete Barras, São Paulo, Brazil). Values in the cells represent the number of specimens by type of bait scent. B:  $\beta$ -ionone; C: 1,8-cineol; E: eugenol; S: methyl salicylate; V: vanillin.

Species	B	C	E	S	V	Total
<i>Euglossa (Euglossa) pleosticta</i>	0	0	1	0	0	1
<i>Euglossa (Euglossa) roderici</i>	2	0	0	6	0	8
<i>Euglossa (Euglossella) viridis</i>	0	0	1	0	0	1
<i>Euglossa (Glossura) annectans</i>	1	1	4	0	0	6
<i>Euglossa (Glossura) iopoecila</i>	0	3	12	0	0	15
<i>Eulaema (Apeulaema) cingulata</i>	0	1	0	0	0	1
<i>Eulaema (Apeulaema) nigrita</i>	0	6	0	0	0	6
<i>Exaerete smaragdina</i>	1	0	0	0	0	1
Total per scent	4	11	18	6	0	39

visited only pads and traps baited with methyl-salicylate (Table 1).

**Morro do Capim (SP3)**

Only 39 orchid bee males, belonging to eight species, were collected at Morro do Capim farm (Table 2) during 42 baiting hours. As in PR3, the most collected species was *Euglossa iopoecila* (15 males; 38.5%). The second most abundant species was *Euglossa roderici* (eight males; 20.5%) followed by *Eulaema nigrita* and *Euglossa annectans* (both represented by six specimens; 15.4%). The other four additional species, *Eulaema cingulata* (Fabricius, 1804), *Euglossa pleosticta*, *Euglossa viridis* (Perty, 1833) and *Exaerete smaragdina* (Guérin, 1844), were each represented by a single male.

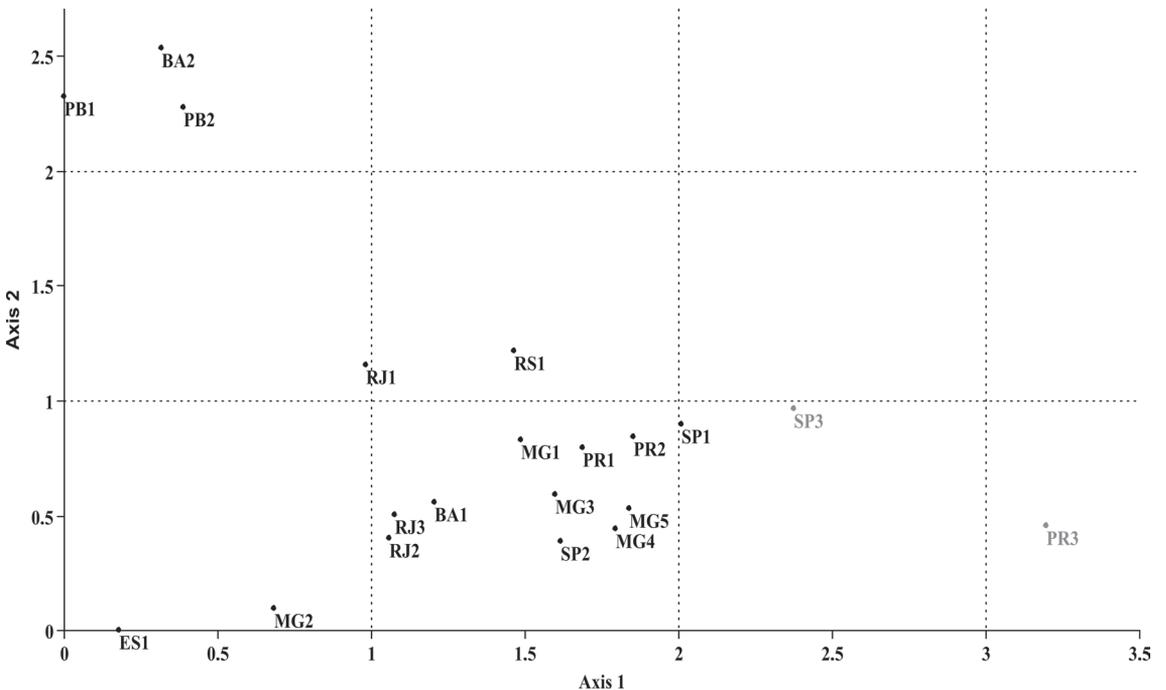
Eugenol was the most attractive scent (18 males and four species) followed by 1,8-cineole (11 males; four species). No species was attracted to vanillin at this site (Table 2).

**Correspondence analysis**

The first two axes of the DCA analysis explained almost two thirds of the variation (DCA1: 0.413;

DCA2: 0.236). Graphic representation of this analysis is depicted in Fig. 1. The faunal surveys presented here – PR3 and SP3 – were placed apart from other surveys carried out in Atlantic forest lowland areas (PB1, PB2, BA1, BA2, RJ1, RJ2, RJ3, ES1). The PR3 site was the most distinctive and came out far apart from all the other sites. SP3, on the other hand, had an intermediate position and was placed somewhat close to inland sites in São Paulo (SP1) and Paraná (PR2). Also, PR3 and SP3 were only distantly associated to the southernmost survey considered in the analysis, carried out in the Rio Grande do Sul state (RS1).

Bee assemblages from studies in lowland coastal sites from more northern localities in the Atlantic Forest (PB1, PB2, BA1, BA2, RJ1, RJ2, RJ3, ES1) showed lower values in the axis 1 and were placed more dispersed along the axes. The areas from the northernmost sites (mainly PB1, PB2 and BA2) also came out far apart from the other analyzed areas. It is noticeable that the fauna from a large remnant of lowland forest in Minas Gerais state (MG2) appeared more closely associated with the coastal sites. On the other hand, the faunas from inland areas of Atlantic forest (MG1, MG3, MG4, MG5, SP1, SP2, PR1, PR2 AND RS1) were more similar to each other and grouped in a more cohesive way.



**FIGURE 1:** DCA analysis of the orchid bee assemblages along the Brazilian Atlantic forest (see text for site codes; the two sites from the current study, PR3 and SP3, are shown in gray).

## DISCUSSION

### Orchid bee richness and abundance

Species richness and specimen abundance found in PR3 and SP3 were remarkably low, with values smaller than what is commonly found northwards in lowland areas at the Brazilian Atlantic forest. Surveys carried out in large forest remnants in the states of Minas Gerais (Nemésio & Silveira, 2006), Espírito Santo (Bonilla-Gómez, 1999) and Rio de Janeiro (Tonhasca *et al.*, 2002) presented at least twenty species of euglossine bees. The number of species collected in PR3 was lower than that found in a site within the Cerrado (Nemésio & Faria, 2004), a humid savanna biome known to have a low number of orchid bee species (Moure, 1967; Nemésio & Faria, 2004). Even when compared to the species richness found in forest fragments inside an urban matrix (Nemésio & Silveira, 2007a), the number of species found in PR3 is quite small.

Data from additional brief euglossine sampling and from generalized bee surveys carried out in the region suggests, however, a slightly higher diversity, totaling 12 species. Brief sampling carried out in the municipality of Morretes, also in Paraná (48°48'25"W; 25°30'20"S; elevation: 13 m), detected the presence of a seventh species, *Eulaema cingulata* (V.C. Mattozo, L.R.R. Faria & G.A.R. Melo, unpublished data) and three species of the genus *Eufriesea* Cockerell, *E. dentilabris* (Mocsáry, 1897), *E. musitans* (Fabricius, 1787) and *E. smaragdina* (Perty, 1833), and two additional species of *Euglossa*, *E. mandibularis* Friese, 1899 and *E. anodorhynchi* Nemésio, 2006, are known to be present in the coastal lowlands of Paraná (see Schwartz-Filho & Laroca, 1999; Moure *et al.*, 2007; Maia, 2008; Nemésio, 2009).

The absence of *E. cingulata* in the PR3 survey may be explained by its low local abundance, which is probably a consequence of its distribution pattern, since the study region represents the southernmost limit of this species (see Oliveira, 2007). Regarding the species of *Eufriesea*, one possible explanation for their absence may be related to their putative association to areas covered with restinga vegetation, being apparently absent from the coastal forests growing on Tertiary soils (unpublished data). We cannot dismiss, however, the hypothesis that the absence of *Eufriesea* results from its species being highly seasonal, usually active during a few months in the rainy season (Kimsey, 1982; Cameron, 2004), or that they are poorly attracted to the usual aromatic baits employed in this type of survey (see Nemésio & Silveira, 2004). In the case of both species of *Euglossa*, no synthetic

fragrance is known to attract their males. The record of  $\beta$ -ionone as an attractant for *E. mandibularis* in Nemésio (2009) seems to represent a fortuitous case, since this substance was used in the SP3 site, and no male of this species was attracted to the baits (see Table 2), despite being present in the area (unpublished results).

In SP3, the number of species found was slightly higher than in PR3, but lower compared to the diversity found in inland areas of semi-deciduous Atlantic forest in the states of São Paulo and Paraná (Rebêlo & Garófalo, 1997; Sofia & Suzuki, 2004) and at more northern coastal Atlantic forest areas (Bonilla-Gómez, 1999; Tonhasca *et al.*, 2002; Souza *et al.*, 2005). The most surprising result from the study at the SP3 site, however, concerns its overall low abundance of euglossine bees. Even the abundance of PR3, much lower than that found in more northern coastal areas (*e.g.* Tonhasca *et al.*, 2002) and comparable to what was found in a brief survey in a Cerrado area (Nemésio & Faria, 2004), is considerably higher than the number of bees found in SP3. Differently from the PR3 site, which is immersed in a large, well-preserved remnant of Atlantic forest (see Maia, 2008), the SP3 site is within a much more fragmented landscape. Habitat fragmentation seems to affect local euglossine faunas (reviewed in Nemésio & Silveira, 2007a, 2010) and therefore, lower values for richness and abundance of these bees in particularly fragmented places, as in SP3, conform to the expected trend.

The difference in the relative abundance of *Eulaema nigrita* between the two study sites is probably also a consequence of their distinct degree of preservation. In PR3, only one male of *E. nigrita* was collected, an unusual result, since this species is known to be attracted to the bait scents utilized (Ramírez *et al.*, 2002). Even in areas with a small number of orchid bees species, the low abundance of *E. nigrita* may be related to the integrity of the forest in the region, since some authors defend that the presence and/or high abundance of *E. nigrita* may be an indicative of environmental modifications in fragmented habitats (Morato *et al.*, 1992; Peruquetti *et al.*, 1999; Nemésio & Silveira, 2007a; Ramalho *et al.*, 2009). On the other hand, the low abundance of *E. nigrita* may reflect just the expected abundance of a species that is near its distribution limits.

### Species composition

The orchid bee fauna at SP3 and particularly at PR3 proved to be quite distinct from that in other

lowland coastal areas situated northwards in the Atlantic forest, as well as from inland Atlantic forest areas. The distinctiveness of the sites studied here is evident in the results from the correspondence analysis, both of them exhibiting relatively high values in axis 1 (Fig. 1). The presence of *Euglossa roderici* and *E. stellfeldi*, and the absence of *Euglossa cordata* (Linnaeus, 1758) and *Eufriesea violacea* (Blanchard, 1840) in both sites seem to be the main reason for this pattern.

Explaining the role of *Euglossa roderici* in placing SP3 and PR3 in a different group is straightforward. This species, which appeared with considerable abundance in both areas, is restricted to the southern portion of the Atlantic forest, in the coast (Nemésio, 2009), reaching its northern limit in the coastal forests of northeastern São Paulo and being absent from the other areas northward. *Euglossa stellfeldi* is restricted to coastal areas in eastern Brazil, including mountain slopes in Espírito Santo (Faria & Melo, 2007). This species, however, has not been found in previous faunal surveys conducted within its distribution range (Bonilla-Gómez, 1999; Tonhasca *et al.*, 2002), and it is here reported for the first time in a systematized orchid bee survey. The absence of *E. stellfeldi* in surveys at coastal areas in Bahia state is also remarkably, since the northernmost record of *E. stellfeldi* is in Alagoas, northeastern Brazil (Faria & Melo, 2007). In PR3 we found a relative abundance of 9% for this bee species (20 specimens), a fact that appears to be expressive. Surprisingly, no males of *E. stellfeldi* were present in SP3, suggesting perhaps that this species might be more sensitive to anthropic disturbances compared to other sympatric species of *Euglossa*.

Also, the somewhat isolated position of PR3 and SP3 may be related to the fact that *Euglossa cordata*, a species with considerable environmental plasticity (Silva & Rebêlo, 1999, 2002; Viana *et al.*, 2002; Farias *et al.*, 2008), and the dominant species in most of the surveys in Atlantic areas northwards (discussed above; Nemésio & Silveira, 2007b), was absent in both studied sites (SP3 and PR3). This species is very common in coastal areas of northeastern and southeastern Brazil (*e.g.* Bezerra & Martins, 2001; Tonhasca *et al.*, 2002) and is also present in inland areas including the state of Paraná (Sofia & Suzuki, 2004), but always with lower abundance than that reported in coastal areas. The presence and abundance of *E. cordata* seem to be an important factor clustering the faunas of lowland areas in eastern Brazil, as suggested previously by Nemésio & Silveira (2007b).

The distribution pattern of *Euglossa iopoecila* also deserves attention. Data presented by Faria & Melo (2007) suggest that *E. iopoecila* is restricted to

coastal areas from Bahia to Santa Catarina. The relative abundance of this bee, however, varies significantly between the present study and other surveys in which it was present (Bonilla-Gómez, 1999; Tonhasca *et al.*, 2002; identified as *E. chalybeata*; see Faria & Melo, 2007 for details). In those studies, the relative abundance of *E. iopoecila* reached, respectively, 6.1% and 5.2%, while in our data it had a relative abundance of 78.7% in PR3 (considering the data from direct collecting with entomological nets, the same methodology used in the other works) and 38.5% in SP3. Taking into consideration that the dominance degree of an euglossine species seems to be inversely related to the number of species found (Nemésio, 2007; Storck-Tonon *et al.*, 2009), it is not surprising that the dominance of *E. iopoecila* is pronounced especially in PR3, the site with the lowest number of species.

The other species collected in PR3 and SP3 have the following distribution patterns: *Eulaema nigrita* and *Exaerete smaragdina* are found along the entire Neotropical region, *Eulaema cingulata* is found in most warm forested regions of the Neotropics, *Euglossa viridis* is found both in the Amazon basin and in the Atlantic forest while *E. pleosticta* is widely distributed in coastal and inland forests along the Atlantic forest domain (see Moure *et al.*, 2007 and Nemésio, 2009 for details).

Composition of the orchid bee fauna also changes considerably when comparing the coastal areas of southern São Paulo and northern Paraná with the state of Rio Grande do Sul. It is important to note that *Eufriesea violacea* (Blanchard), the orchid bee most frequently found in Rio Grande do Sul (97.8% of collected bees), is typical of inland areas, seeming to be associated to semi-deciduous forests in the countryside regions of Brazil (Sofia & Suzuki, 2004; Sofia *et al.* 2004; see also Nemésio & Silveira, 2007b and Nemésio, 2009). In this sense, it is outstanding that the euglossine fauna of Rio Grande do Sul represents only a subset of the fauna found at the inland forests of Paraná (see Sofia & Suzuki, 2004 and Sofia *et al.*, 2004), lacking the species from the coastal forests of Paraná.

Our data reinforces the general pattern that the euglossine fauna of the Atlantic forest changes considerably between inland and coastal areas at the same latitude (see Nemésio & Silveira, 2007b; Nemésio, 2009). Only four of the nine species found in PR3 and SP3 – *Euglossa annectans*, *E. pleosticta*, *Eulaema nigrita* and *Exaerete smaragdina* – are shared with inland forests of Paraná (Suzuki *et al.*, 2002; Sofia & Suzuki, 2004; Sofia *et al.*, 2004) and São Paulo

(Rebêlo & Garófalo, 1991, 1997). Both species of *Euglossa* are associated mainly with the Atlantic forest of southeastern Brazil, *E. annectans* showing higher abundance in coastal areas and *E. pleosticta* exhibiting a reversed pattern (e.g. Bonilla-Gómez, 1999; Nemésio & Faria, 2004; Sofia & Suzuki, 2004). In some inland areas in São Paulo (Rebêlo & Garófalo, 1991, 1997) and Paraná (Suzuki *et al.*, 2002), *E. pleosticta* is the most common species, with relative abundance near or above 50%, while in coastal areas it appears in low relative abundance (Bonilla-Gómez, 1999; Tonhasca *et al.*, 2002). Nemésio & Silveira (2007b) considered this species as typical of semi-deciduous forests. Its occurrence in lowland areas along the coast may be a more recent event in the evolution of its geographic distribution.

In conclusion, considering both diversity and species composition, the orchid bee fauna found in the coastal forests of Paraná and southern São Paulo is a impoverished subset of the fauna found in northern coastal areas of Atlantic forest, with a few endemic species present. Only *Euglossa roderici* is here considered to be endemic of southern São Paulo and Paraná coastal areas, although *E. anodorhynchi* may also prove to be an additional endemic. It is known to inhabit the studied region (Maia, 2008; Melo *et al.*, 2008), but was not collected in our surveys.

### Sampling methodology

The results found in the present study vary greatly when both collecting methods are compared. A total of 221 males (87%) were collected with the direct method, while only 33 bees were captured by the scent traps. Also the number of species, their abundance, and the community structure varied depending on the methodology used.

With direct collecting, all the six species in PR3 attracted to synthetic scents were sampled. The three most abundant species were collected with the following percentage: *Euglossa iopoeila* was, by far, the most abundant species (78.7%), followed by *E. annectans* (14.9%) and *E. roderici* (11.3%). Using bait trapping we found different relative abundances: *E. iopoeila*, 54.5%; *E. roderici*, 33.3%; and *E. annectans*, 12.1%. It is interesting to note that the relative abundance of *E. roderici* was three times higher in bait traps, while the abundance of *E. iopoeila* decreased considerably. One possible explanation for this result refers to the aggressiveness of some *Euglossa* species when approaching the bait trap (see Nemésio & Morato, 2006). Males of *E. iopoeila* seem to be aggressive and

when more than one male arrived to the scent pads, agonistic behavior was noticed. On the other hand, males of *E. roderici* seem to be more cautious when approaching the scent lures and may be repelled by the presence of the collector.

Besides the relative abundance, the number of species caught on bait traps was half that caught with the direct method. Two species, *Eulaema nigrita* and *Euglossa pleosticta*, both represented by singletons, were caught only with insect nets. Due to the low abundance of these species, it is difficult to compare the effectiveness of the bait trap. The most interesting case refers to *Euglossa stellfeldi*, which had twenty specimens caught with hand nets, and was not captured in the traps. In this case, the species seems to be common at the sampled area and its absence in the traps may reflect the inefficiency of the method.

Nemésio & Morato (2004, 2006) also suggested that individuals of *Euglossa* escape more frequently from traps when compared to males of *Eulaema*, a situation that may be related to the smaller size of the species of *Euglossa*, allowing them to pass more easily through a hole (see Nemésio & Morato, 2004, 2006). Since the fauna we surveyed is formed almost only by species of *Euglossa*, this could explain why the traps showed low efficiency. Other factors related directly to how the traps work should also be taken into consideration. The most significant seems to be how the trap design interferes with the volatilization of the scents. There are no conclusive data about how much interference occurs, but it is reasonable to think that the spread of the scents becomes more difficult through relatively small holes in the trap wall, when compared to a paper pad suspended directly in the air.

Our results point out that bait traps should be used carefully and the use of this method solely should be avoided. Bait traps should be considered a complementary method together with the direct method (Nemésio & Morato, 2004, 2006; Storck-Tonon *et al.*, 2009). The lower efficiency of traps, however, should be considered carefully, because community parameters (as abundance and species composition) may influence the level of bias introduced by traps. Also, the use of traps remains an important method for increasing per capita sampling effort, in particular when large areas and a large number of sites are surveyed simultaneously.

### RESUMO

*Os levantamentos da fauna de abelhas euglossíneas realizados até o momento ao longo da floresta Atlântica*

são restritos a poucas regiões e não permitem um entendimento mais aprofundado dos padrões latitudinais de distribuição e diversidade dessas abelhas. Por este motivo, duas áreas de floresta Atlântica do sul do Brasil, na planície costeira de São Paulo (Sete Barras, Faz. Morro do Capim: SP3) e do Paraná (Antonina, Reserva Natural do Rio Cachoeira: PR3), tiveram sua fauna de euglossíneos amostrada. Em PR3, foi avaliada também a eficiência de dois métodos alternativos de coleta, fazendo-se a comparação entre armadilhas plásticas, iscadas com fragrâncias, e coleta direta com rede entomológica em iscas odoríferas. A diversidade e abundância das abelhas foram muito baixas: apenas 39 machos de oito espécies foram coletados em SP3 (*Euglossa iopocila*, *Euglossa roderici*, *Eulaema nigrita*, *Euglossa annectans*, *Eulaema cingulata*, *Euglossa pleosticta*, *Euglossa viridis* e *Exaerete smaragdina*) e 254 machos de seis espécies em PR3 (*Euglossa iopocila*, *Euglossa annectans*, *Euglossa stellfeldi*, *Euglossa roderici*, *Euglossa pleosticta* e *Eulaema nigrita*). A comparação entre os métodos de amostragem mostrou que a coleta direta (seis espécies; 221 espécimes) foi mais eficiente do que o uso de armadilhas (três espécies; 33 espécimes). A análise de correspondência (DCA) mostrou que os dois levantamentos apresentados aqui se posicionaram relativamente próximos entre si, porém afastados dos outros locais comparados, não se agrupando com o levantamento mais meridional nos domínios da floresta Atlântica do Rio Grande do Sul, nem com aqueles conduzidos em áreas de terras baixas mais ao norte nesse bioma.

PALAVRAS-CHAVE: Euglossina; *Euglossa*; Floresta Atlântica; Euglossini; Abelhas.

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