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The orchid-bee faunas (Hymenoptera: Apidae) of 'Reserva Ecológica Michelin', 'RPPN Serra Bonita' and one Atlantic Forest remnant in the state of Bahia, Brazil, with new geographic records

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Abstract

The orchid bee faunas of two private natural preserves, 'Reserva Particular do Patrimônio Natural da Serra Bonita' (RSB) and 'Reserva Ecológica Michelin' (REM), and a forest fragment inside the campus of the 'Universidade Estadual de Santa Cruz', were surveyed for the first time. All three areas constitute Atlantic Forest remnants in the southern portion of the state of Bahia, Brazil. A total of 1,782 males belonging to 32 species were actively collected with insect nets during 90 hours of field work from November, 2009, to January, 2012. *Euglossa cyanochlora* Moure, 1996—one of the rarest orchid bee species—was found at RSB and REM, the latter representing the northernmost record for this species. *Euglossa cognata*, Moure, 1970 was found at RSB, the northernmost record for this species in the Atlantic Forest and the only recent record for this species at the northern border of Jequitinhonha river.

Keywords: Atlantic Forest, conservation, Euglossina, euglossine bees, Hexapoda,

A fauna de abelhas-das-orquídeas (Hymenoptera: Apidae) da Reserva Ecológica Michelin, RPPN Serra Bonita e um remanescente florestal de Mata Atlântica no baixo sul da Bahia, Brasil, com novos registros geográficos

Resumo

As faunas de abelhas-das-orquídeas de duas áreas florestais privadas, a Reserva Particular do Patrimônio Natural da Serra Bonita (RSB) e a Reserva Ecológica Michelin (REM), e de um remanescente florestal na área do *campus* da Universidade Estadual de Santa Cruz, foram amostradas pela primeira vez. Um total de 1.782 machos pertencentes a 32 espécies foi coletado com o auxílio de redes entomológicas durante 90 horas de esforço amostral entre Novembro de 2009 e Janeiro de 2012. *Euglossa cyanochlora* Moure, 1996—uma das espécies mais raras de Euglossina—foi coletada na RSB e na REM, esse último local representando o registro mais ao norte dessa espécie. *Euglossa cognata*, Moure, 1970 foi coletada na RSB, sendo esse o registro mais ao norte dessa espécie na Mata Atlântica e, também, o único registro recente da espécie ao norte do rio Jequitinhonha.

Palavras-chave: Abelhas euglossinas, conservação, Euglossina, Hexapoda, Mata Atlântica.

1. Introduction

Despite the pioneer inventories carried out in the state of Bahia in the late 1980s and 1990s (Raw, 1989; Neves and Viana, 1997, 1999), the orchid-bee fauna (Hymenoptera: Apidae: Euglossina) of the dense Atlantic Forest of the southern portion of the state remained largely understudied until very recently, when intensive collections were conducted in the largest forest remnants in the region (Melo, 2005; Nemésio 2011a, 2012b, 2013a, b; Nemésio et al., 2012). Only a brief list of species occurring in the Bahian Atlantic Forest was available (Neves and Viana, 2003), but it was based on sporadic collections and museum specimens. The Atlantic Forest of southern Bahia is probably the richest portion of the biome, with the highest levels of endemism for many taxonomic groups (e.g., Dean, 1995; Pacheco et al., 1996; Thomas et al., 1997, 1998; Sambuichi et al., 2008), including orchid bees (Nemésio, 2011c, d, e, 2012a, 2013a, b). Nevertheless, most of the original forest cover in southern Bahia was wiped out and only scattered fragments remain (Fundação SOS Mata Atlântica, 1993; Ribeiro et al., 2009), usually not exceeding 20,000 ha (Ribeiro et al., 2009). As a consequence of deforestation, populations of forest-dependent animal species usually decline (Collinge, 1996; Tocher et al., 1997), and species endemic in such fragmented biomes are more prone to

extinction. Since many orchid bees are forest-dependent insects, some of them rely on large forested areas to maintain viable populations (Tonhasca Jr. et al., 2002; Nemésio and Silveira, 2006; Nemésio, 2009, 2011b), a better sampling of the forest remnants in southern Bahia is important to provide information on the actual distributions of these bees and a tentative estimate of their population sizes.

The main goal of this study was to sample three forest remnants in southern Bahia for orchid bees for the first time, thus expanding our distributional and taxonomic knowledge on these bees and providing relevant data on their species.

2. Material and Methods

2.1. Study sites

This study was conducted at (i) the 'Reserva Ecológica Michelin' (REM), a 3,000-ha reservation situated in the municipalities of Igrapiúna and Ituberá, connected to approximately 10,000 ha of non-protected forested areas. The vegetation in the region is essentially dense Atlantic Rain Forest [Central Lowland Forest according to Thomas and Carvalho (2003)]. Most of the REM consists of secondary forests in various successional stages, immersed in a matrix of rubber tree plantation, but pristine forested areas can also be found in the reservation; (ii) the 'Reserva Particular do Patrimônio Natural da Serra Bonita' (RSB), in the municipality of Camacan, with some 1,000 ha of protected montane forest (up to 1,000 m a.s.l.) connected to approximately 5,000 ha of non-protected forested areas; and (iii) at a secondary forest patch located inside the *campus* of the 'Universidade Estadual de Santa Cruz' (UESC), with some 300 ha but connected to over 1,000 ha of forested areas in different successional stages (the area was formerly a cocoa plantation). Two sites were sampled at REM, and one site was sampled at RSB and UESC, but the site at RSB was sampled twice (January, 2010 and January, 2012) for comparison.

2.2. Sampling

Twenty hours of active sampling with insect nets were performed in each of the selected sites in the preserves (except the first sampling at RSB, only 10 hours), totaling 90 hours, following the methodology proposed by Nemésio (2010a, b, 2011a, b): REM site-1 (13°50'01"S, 39°14'56"W, *ca*. 300 m above sea level) was sampled on the 28th of November, 1st and 2nd of December, 2009; from 09:00h to 16:00h (except on 28th of November, from 09:00h to 15:00h); REM site-2 (13°50'44"S - 39°13'35"W, *ca*. 260 m above sea level) was sampled on the 27th and 30th of November, 2009 (from 07:00h to 17:00h); RSB (15°23'40"S - 39°34'01"W, *ca*. 920 m above sea level) was sampled on the 24th of January, 2010, from 07:00h to 17:00h, and from the 21st to the 24th of January, 2012, from 09:00h to 15:00h (except on the 24th, from 09:00h to 13:00h); UESC (14°47'43"S, 39°10'22"W, *ca*. 40 m above sea level), on 18th of November, 2009 (from 08:00h to 12:00h), 3rd of December, 2009 (from 07:00h to 12:00h), 22nd of January, 2010 (from 07:00h to 13:00h), and 20th of February, 2010 (from 11:00h to 16:00h). At each site, 17 scent baits were placed *ca.* 2.0 meters apart from each other at about 1.5 m above the ground. These baits were made of cotton wadding soaked with one of the following substances, known or believed to be attractive to orchid bees: benzyl acetate, benzyl alcohol, *r*-carvone, 1,8-cineole, *p*-cresol acetate, dimethoxybenzene, eugenol, β-ionone, methyl benzoate, methyl *trans*-cinnamate, heneicosane, methyl salicylate, skatole, tricosane, *p*-tolyl acetate, vanillin, and a mixture (1:1) of methyl *trans*-cinnamate and *p*-tolyl acetate. Baits with cineole, the most volatile compound, were recharged every hour. Bees arriving on the baits during the sampling period were collected with insect nets and killed with ethyl acetate. They were labelled as belonging to the project "Euglossina da Hileia Baiana" and were deposited at the Entomological Collection of the'Universidade Federal de Minas Gerais' (UFMG).

2.3. Data analysis

Diversity was estimated using the Shannon-Wiener diversity index (H'), as $H' = - \Sigma\, p_i \ln (p_i)$, where p_i is the proportion of total number of species made up of the ith species (Pielou, 1975). Evenness (E) was estimated through the formula $E = H'/ \ln (S)$, where S is the species richness. The similarity in faunistic composition among all sites was estimated by the percent similarity index of Renkonen, recommended by Wolda (1981) for small samples. Based on those similarities, the areas were grouped using UPGMA (Sneath and Sokal, 1973).

2.4. Taxonomy

Taxonomy follows Nemésio and Rasmussen (2011) and Nemésio (2012a).

3. Results

One thousand, seven hundred and eighty-two orchid-bee males belonging to 32 species in four genera were collected at the three forest remnants (Table 1). Abundance was the highest at UESC (29 specimens/hour), followed by RSB (22 specimens/hour) and REM (13 specimens/hour). *Euglossa ignita* Smith, 1874 was the most abundant species at both REM and UESC, and the second most abundant at RSB (where *Eulaema nigrita* Lepeletier, 1841 was the most abundant species). *Euglossa carolina* Nemésio, 2009 was the second most abundant species at REM and UESC (Table 1). Species of *Eufriesea* Cockerell, 1908 were only recorded at RSB. Thirteen species were represented by singletons or only two specimens (Table 1). Twenty species were recorded at each REM and UESC, whereas 23 species were recorded at RSB. UESC presented the lowest diversity (H' = 1.68) and evenness (E = 0.56), whereas diversity ranged from H' = 2.06 (site 1) to H' = 2.29 (site 2) at REM, and from H' = 2.01 (January, 2012) to H' = 2.31 (January, 2010) at RSB. Evenness was fairly constant (E = 0.78) at REM and ranged from E = 0.72 (2012) to E = 0.77 (2010) at RSB (Table 1).

Table 1. Diversity, species richness, evenness and relative abundance (%) of specimens of each orchid-bee species collected at 'Reserva Ecológica Michelin' (two sites, REM1 and REM2), at a forested area at the 'Universidade Estadual de Santa Cruz' (UESC), and at 'Reserva do Patrimônio Natural da Serra Bonita' (two collections, RSB2010 and RSB2012).

Species	REM1	REM 2	UESC	RSB2010	RSB2012	Total (N)
Eufriesea atlantica Nemésio, 2008	0	0	0	0.74	1.51	08
Eufriesea surinamensis (Linnaeus, 1758)	0	0	0	0	0.25	01
Euglossa amazonica Dressler, 1982b	5.28	6.91	0.17	1.49	1.00	37
Eg. aratingae Nemésio, 2009	0	0	0.17	0	0	01
Eg. adiastola Hinojosa-Díaz, Nemésio & Engel, 2012	2.48	1.38	3.65	0.74	0	34
Eg. bembei Nemésio, 2011	0	0	0	2.60	2.26	07
Eg. calycina Faria Junior & Melo, 2012	0.31	0	1.04	0.74	4.52	09
Eg. carinilabris Dressler, 1982a	0	0	0.17	0	0	01
Eg. carolina Nemésio, 2009	16.46	7.37	26.74	5.95	4.27	239
Eg. cognata Moure, 1970	0	0	0	0.74	0	02
Eg. clausi Nemésio & Engel, 2012	2.17	0.92	0.69	11.15	8.04	43
Eg. cyanochlora Moure, 1996	0.93	0.46	0	0.74	0	06
Eg. despecta Moure, 1968	9.01	11.98	1.74	0.74	0	67
Eg. fimbriata Moure, 1968	0	0	0	0.37	0	01
Eg. hemichlora Cockerell, 1917	0	0	0.17	0	0	01
Eg. ignita Smith, 1874	29.19	36.40	43.40	20.45	12.31	478
Eg. imperialis Cockerell, 1917	6.83	10.14	3.12	9.29	2.01	87
Eg. liopoda Dressler, 1982	0.31	0	0	0	0	01
Eg. marianae Nemésio, 2011	2.80	0	0	0	0	09
Eg. milenae Bembé, 2008	0	0	0.35	0	0	02
Eg. monnei Nemésio, 2012	0.31	0	0	0.37	0	02
Eg. pleosticta Dressler, 1982c	0.31	0	0	0	0	01
Eg. roubiki Nemésio, 2009	6.21	7.373	0.35	15.99	8.54	81
Eg. securigera Dressler, 1982c	0.31	0	0.17	0.74	0.50	04
Eg. stellfeldi Moure, 1947	0	0	0	0	0.50	02
Eg. viridis (Perty, 1833)	0	0	0	0.37	0	01
Eulaema atleticana Nemésio, 2009	5.28	5.07	12.5	1.49	0.75	104
El. marcii Nemésio, 2009	0.93	0.46	1.22	10.78	16.83	40
El. nigrita Lepeletier, 1841	8.07	7.83	2.95	14.50	36.43	99
El. niveofasciata (Friese, 1899)	2.80	3.23	0.87	0	0.25	21
Exaerete frontalis (Guérin-Méneville, 1844)	0	0.46	0.17	0	0	02
Exaerete smaragdina (Guérin-Méneville, 1844)	0	0	0.35	0	0	02
Total (N)	322	217	576	269	398	1.782
Species richness (S)	19	14	20	20	16	32
Shannon-Wiener diversity index (H')	2.29	2.06	1.68	2.31	2.01	2.25
Evenness (E)	0.78	0.78	0.56	0.77	0.72	0.65

The ordination of the sites according to their faunas showed a low overall similarity among them. Both sites at REM grouped together with ca. 83% similarity and both collections at RSB grouped together with ca. 66% similarity. REM and UESC, situated at the lowest altitudes, grouped together with ca. 63% similarity and then grouped to RSB with ca. 41% similarity.

4. Discussion

The strategy of intensive sampling, using the same methods as employed here, over a few days during the season when orchid bees are most actively foraging, has been already demonstrated to be adequate for surveys focusing on this group of bees and previously discussed (Nemésio, 2010b, 2011a, b, Nemésio et al., 2012).

Recent studies in southern Bahia have shown that around 30 orchid-bee species occur in the area (Nemésio, 2011a, 2013a, b) and the present study is in accordance with this figure. Two species recorded at the present study had not been collected in recent studies in southern Bahia: a single male *Euglossa carinilabris* Dressler, 1982a was collected at UESC, in Ilhéus, close to its type locality; two males *Euglossa stellfeldi* Moure, 1947 were collected at

RSB, representing the northernmost record of this species in the entire Atlantic Forest. Three other species also deserve further attention. All these species are discussed in detail below.

4.1. *Euglossa carinilabris* Dressler, 1982a

Euglossa carinilabris was described from Itabuna, only 10 km from the site where the single specimen collected at UESC comes from. The occurrence of this species at this site is, thus, absolutely no surprise. On the other hand, its rarity is outstanding, since old records of this species in southern Bahia did not suggest it was a rare species. Faria Jr. and Melo (2007) listed this species from Alagoas, in the north, to Espírito Santo, in the south. It is important here to notice that Faria Jr. and Melo (2007) treated *Eg. carinilabris* as a junior synonym of *Eg. stellfeldi*. Moreover, their interpretation of *Eg. stellfeldi* is different from that adopted here. Faria Jr. and Melo (2007) considered as *Eg. stellfeldi* the same bee Nemésio (2009) considered as *Euglossa solangeae* Nemésio, 2007, and the bee here treated as *Eg. stellfeldi* was treated by Faria Jr. and Melo (2007) as *Eg. annectans* Dressler, 1982a, which is, in turn, considered by Nemésio (2009) and Nemésio and Rasmussen (2011) as a junior synonym of *Eg. stellfeldi* (*sensu* Nemésio, 2009). All surveys in southern Bahia carried out recently (Melo, 2005; Nemésio, 2011a, 2013a, b), as well as in Alagoas (Nemésio, 2010b) and Espírito Santo (Nemésio, 2011b, 2013c) failed to record this species. Besides the integumental coloration and the gap in geographic distribution (from northern Espírito Santo to southern coastal São Paulo), the apparent rarity of *Eg. carinilabris* in northeastern Brazil (opposed to the relative commonness of *Eg. solangeae* in southern Brazil) is also suggestive that they may indeed be separate species, and not the same one as suggested by Faria Jr. and Melo (2007).

4.2. *Euglossa cognata* Moure, 1970

Euglossa cognata is apparently a forest-dependent species and Nemésio (2009: 114) suggested it should be considered as vulnerable according to IUCN criteria. Recent studies in southern Bahia, including collections in large forest remnants as 'Parque Estadual da Serra do Conduru', 'Reserva Biológica de Una' and 'Reserva Particular do Patrimônio Natural Estação Veracel', failed to record this species (Melo, 2005; Nemésio, 2011a, 2013a, b). The only area where specimens belonging to this species were recently (last five years) collected from is 'Parque Nacional do Monte Pascoal' and its vicinities (Nemésio, unpub. data). The record of two specimens of this species at RSB is important for two reasons: it is the first recent record of this species at the northern border of Jequitinhonha river and it is the first time this species is collected at relatively high altitudes (around 1,000 m a.s.l.), suggesting the distribution of this species might have been wider in the past and deforestation might have contributed for its current scattered distribution. It is noticeable, however, that this species was collected at RSB in 2010 but not seen in 2012. It is possible that populations of *Euglossa cognata* may be declining in areas where it occurs or had been previously recorded, a fact already observed for its close ally *Eg. marianae* (see Nemésio 2011b, Nemésio, 2013c).

4.3. *Euglossa cyanochlora* Moure, 1996

This conspicuous species, the largest member of *Euglossa* Latreille, 1802, was only known from two specimens until very recently, when intensive collections were carried out along its supposed distribution area and 24 specimens were collected (Nemésio et al., 2012). Nevertheless, its abundance is very low where it occurs and data on its biology are virtually non-existent – except for the scents males are attracted to (see Nemésio et al., 2012). The four specimens collected at REM represent the northernmost record of this species, extending its previous record from 'Parque Estadual da Serra do Conduru' (Nemésio, 2011a). Nevertheless, as shown by Nemésio et al. (2012), it is possible that its distributional range extends further north, and potentially suitable forested areas should be sampled in search for this species to correctly understand its actual geographic distribution. The two males collected at RSB represent the highest altitude (around 1,000 m a.s.l.) where *Eg. cyanochlora* has ever been recorded.

4.4. *Euglossa stellfeldi* Moure, 1947

Euglossa stellfeldi Moure, 1947 (*sensu* Nemésio, 2009) is a species with a remarkable geographic distribution: in southern Brazil it occurs at sea level, whereas in southeastern Brazil it is only recorded at higher elevations, usually above 850 m a.s.l. (Nemésio, 2009), a distribution pattern also known for other organisms, including bees (see Silveira and Cure, 1993). This species was recently recorded at 'Reserva Particular do Patrimônio Natural Duas Barras', at the border of the states of Minas Gerais and Bahia, also at altitudes ranging from 900 to 1,100 m a.s.l. (Nemésio, 2012b). The present record, however, extends its distribution northwards in at least 200 km and represents the northernmost record of this species.

4.5. *Euglossa viridis* (Perty, 1833)

Recent records of this species in southern Bahia (Nemésio, 2011a, 2013a), close to RSB, could indicate this is not a noteworthy record, especially when one considers that males of this species are not strongly attracted to the most commonly used scent baits. Nevertheless, this species had never been recorded at altitudes over 900 m a.s.l.

This record, together with the records of *Eg. cognata* and *Eg. cyanochlora*, as well as *Eulaema atleticana* Nemésio, 2009 and *El. niveofasciata* (Friese, 1899) – and also *Exaerete salsai* Nemésio, 2011c, *Eufriesea brasilianorum* (Friese, 1899) (see Nemésio, 2011e, 2012b) – show that most of the orchid-bee fauna of coastal Bahian Atlantic Forest do reach higher altitudes at the western limits of 'Hileia Baiana', where mean annual temperature is usually lower and average seasonal precipitation is higher, i.e., dry periods are usually longer than at coastal areas (data from WorldClim: http://www.worldclim.org/). The only noticeable exception is *Euglossa marianae*, but this may

be an artifact of environmental quality, since this latter species is apparently associated to the largest forest remnants in the Atlantic Forest (Tonhasca Jr. et al., 2002; Nemésio and Silveira, 2006; Nemésio, 2011b). Thus, its absence from higher altitudes might be more related to poor environmental quality (small forest remnants) than to the elevation itself, and this hypothesis is supported by the fact this species reach 'Parque Estadual do Rio Doce' (Nemésio and Silveira, 2006), far from the coast, deep inside the state of Minas Gerais, where humidity is lower than at the coast.

Although diversity and richness were reasonably similar among the three sampled forest remnants (Table 1), overall similarity was quite low. It is even noticeable that measured similarity between two samplings at RSB in a two-year interval was only around 66%, showing that either fluctuation from year to year can be relatively high or that the sampling methodology employed here is not sensitive enough. The strong fluctuation in the population of the two species of *Eulaema* belonging to the subgenus *Apeulaema*, particularly *El. nigrita* (see Table 1), however, supports the first hypothesis, since the similarity index used here takes in account the relative abundance of each species and more than 20% in the similarity loss can be attributed solely to the ups and downs of *El. nigrita* populations. Populations of this latter species tend to decline during the drier periods (Nemésio, unpub. data), and January, 2010 was exceptionally dry at RSB, at least partially explaining this low similarity between both samplings. REM and UESC, situated at lower elevations, showed higher similarities to each other than to RSB, situated at higher elevations. The high abundance of *El. nigrita* at RSB probably accounts for the low overall similarity between this area and the two other forest remnants.

Although they are not very large forest remnants in southern Bahia, all three studied areas presented rich and diverse orchid-bee faunas and apparently are big enough to maintain viable populations of many species, including some species rarely collected which population sizes in nature may not be too large. These areas are very important to be preserved, not only for housing these species, but also because they are strategically situated and connect larger forested areas, such as the 'Reserva Biológica de Una', which orchid-bee fauna has shown to be one of the richest and most diverse in the entire Atlantic Forest domain (Nemésio, 2013a). More private areas like those ones should be established, and it will be important if more public preserves are created in this region.

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