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SHORT NOTE

First record of the occurrence of *Partamona ailyae* Camargo (Hymenoptera: Apidae) in Atlantic Forest

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Abstract
This is the first record of the stingless bee *Partamona ailyae* Camargo in an area of the Atlantic Forest in Brazil. The morphological identification and COI and 12S sequences indicated that the samples collected in the Atlantic Forest and other areas of the range of distribution belong to the same biological species. The data revealed low intraspecific variation and high interspecific divergence, with no overlap of the taxa compared.

The tribe Meliponini clusters 417 valid species of stingless bees in the Neotropical region (Camargo & Pedro, 2003). Recent data demonstrate 244 species described and another 89 undescribed species for Brazil (Pedro, 2014). Stingless bees have broad geographic distribution, a large number of individuals per colony and generalist feeding habits (Roubik, 1989).

Species of the genus *Partamona* Schwarz are distributed over a large geographic range, with records from central Mexico to southern Brazil (i.e. Roubik, 2006). These stingless bees are found in forests, savannas, semiarid regions and locations with an altitude of more than 2000 m above sea level (Pedro & Camargo, 2003; Camargo & Pedro, 2003). They use a wide variety of substrates for nest building, but most nest in termite mounds (Camargo, 1980; Camargo & Pedro, 2003).

Thirty-three valid species have been described for the genus *Partamona* (Camargo & Pedro, 2003). Twenty-three of these species occur exclusively in Brazil (Pedro, 2014), three of which have previously been recorded in areas of the Atlantic Forest (hereafter AF): *Partamona criptica* Pedro and Camargo, *Partamona helleri* Friese and *Partamona sooretamae* Pedro and Camargo (Camargo & Pedro, 2003).

Partamona ailyae Camargo is known to nest in epigeal termite mounds. This bee has broad distribution, occurring from the rainforest in southeastern Amazonia to savannas in central Brazil and semiarid areas of the state of Piauí in the northeastern region of Brazil (Camargo, 1980; Camargo & Pedro, 2003). However, there are no previous records of this species in AF areas.

This paper reports the first record of *P. ailyae* in the Atlantic Forest, broadening its distribution to the Atlantic portion of southeastern Brazil.

Twelve *P. ailyae* nests were located in the upper portion of the south of the state of Bahia, in an area of approximately 100,000 m² in AF (13°27’14’’ S and 39°25’15’’

W), within the limits of the Bom Jesus, Jacarandá and Riacho do Louro farms (Tancredo Neves municipality); additionally, in the southern region of this state, two nests were located in rural areas of the municipalities of Uruçuca and Una (Table 1, Fig 1). The regions have characteristic secondary Atlantic Forest vegetation in different stages of regeneration: low vegetation, high vegetation and secondary forest. According to the Köppen classification (Veloso et al., 1991), the climate is Af (humid tropical or equatorial), with the temperature ranging between approximately 22 and 31º C. Mean annual precipitation in AF is 1600 mm, with rains distributed throughout the year.

The first observation of a specimen of *P. ailyae* in the region occurred in 2004 during a bee sampling from flowers with an insect net. In 2008, the first colony was found using an artificial feeder containing sugar water. The localization of the colony was possible due to successive movements of the feeder along a straight line, approximately five meters at a time, following the flight path of the worker bees. Subsequently, other nests were located through active searches and georeferenced. Thirteen *P. ailyae* nests (12 in Presidente Tancredo Neves and one in Uruçuca) were found on clay soil in epigeal termite mounds of the subfamily Apicotermitinae and one nest was found in Una at the base of a tree.

Specimens from each colony were sent to the Laboratório de Abelhas Solitárias e Ecologia de Ecossistemas da Faculdade de Tecnologia e Ciências (LABEE/FTC), pinned, labeled and dried. The morphological identification of *P. ailyae* was performed by Dr. Silvia Regina de Menezes Pedro (FFCLRP-USP). Voucher specimens were deposited in the Zoology Museum of the Faculty of Technology and Science (MZ/FTC) and Prof. J.M.F. Camargo Collection (RPSP), (FFCLRP-USP).

Specimens from 10 out the 14 *P. ailyae* nests were analyzed through molecular markers for the confirmation of their taxonomic status. DNA extraction, PCR amplification, amplicon purification, sequencing of the gene regions cytochrome C oxidase (COI) and 12S ribosomal DNA, as well as the edition and analysis of the sequences were performed according to previous reports (Miranda et al., 2016).

Table 1. *Partamona ailyae* Camargo nests sampled in municipality of Presidente Tancredo Neves (nests 1-12) in south lowlands, state of Bahia, and municipalities of Uruçuca and Una (nests 13-14) in southern portion of state of Bahia. Some nest characteristics are presented. "H" = height of nest entrance in the termite nest. nc = non-collected datum.

Nest	Localities	Farm	Coordinates	Termite nest height x diameter (cm)	H
1	PTN	Bom Jesus	13º26'57.2" S, 39º30"26.4" W	93 x 203	37
2		Jacarandá	Nc	72 x 175	29
3		Bom Jesus	13º26'28.3" S, 39º29'57.3" W	80 x 210	38
4		Bom Jesus	13º26'57.2" S, 39º30'26.4" W	84 x 198	43
5		Jacarandá	13º26'40.5" S, 39º30"35.0" W	60 x 170	17
6		Jacarandá	13º26'37.3" S, 39º30"36.0" W	81 x 208	50
7		Riacho do Louro	Nc	52 x 140	8
8		Bom Jesus	Nc	83 x 196	10
9		Jacarandá	13º26'57.2" S, 39º30"26.7" W	70 x 150	20
10		Jacarandá	13º26'28.2" S, 39º29"27.3" W	73 x 202	13
11		Jacarandá	Nc	75 x 195	15
12		Jacarandá	Nc	85 x 208	42
13	URU	nc	14°37'51.6"S, 39°16'48.9"W	nc	nc
14	UNA	nc	15°11'41.1"S, 39°03'00.6"W	nc	nc

Samples from the Atlantic Forest and other areas of distribution were compared through an analysis of 629 base pairs of the COI gene. The optimum threshold (OT) was estimated considering 12 haplotypes of *P. ailyae* [three obtained from the AF samples and nine from samples of the known distribution area, as well as nine haplotypes from *Partamona rustica* Pedro and Camargo and five haplotypes from *Partamona cupira* Smith as controls]. Using the "local minima" function of the SPIDER (Species IDentity and Evolution in R) package, the estimated OT was 1.95% between intraspecific and interspecific variation. The Kimura two-parameter model of the MEGA 7 program demonstrated lower intraspecific and higher interspecific variation values for all combined pairs in comparison to the OT, indicating that the three species are well differentiated. Moreover, the 12S haplotypes of the *P. ailyae* AF samples revealed the same insertion of five base pairs beginning at position 25 of the sequence (417 bp) previously described by Cardoso (personal communication, September 13, 2016) in samples of *P. ailyae* from geographically distinct origins.

Thus, the molecular analyses confirmed the morphological identification of the samples. It is very unlikely that *P. ailyae* arrived to the Atlantic Forest in Bahia by human hands. We believe that *Partamona* species cannot be maintained by beekeepers or common people for three main reasons: i) these species do not produce much honey and it is inappropriate for human consumption (some *Partamona* species collect feces); ii) they can hardly be kept in rational boxes because most of them are termitophilic species and their nests are hosted by specific termite species, and they are not managed by humans yet; iii) *P. ailyae* has a strong defensive behavior and bites hard when its nests are opened. Within the Meliponini tribe, *Partamona* is an excellent study model for population genetics and biogeographic studies because their colonies are rarely manipulated.

Our findings demonstrate the occurrence of *P. ailyae* in an area of AF in Brazil, broadening the known area of the distribution of the species (Fig 1).This new record makes this species particularly interesting for future phylogeographic studies, as it is found in the Amazon forest, savannas, semiarid regions and AF in Brazil. Such studies could contribute toward clarifying evolutionary relations in the biota of different biomes of the Neotropical region.

Fig 1. New records of occurrence of *Partamona ailyae* in Atlantic Forest (diagonal lines). PTN = municipality of Presidente Tancredo Neves in upper portion of the south of the state of Bahia; URU = municipality of Uruçuca and UNA = municipality of Una, both located in southern portion of state of Bahia. These records broaden the known distribution of the species (in yellow) proposed by Camargo and Pedro (2003).

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