Neotrop Entomol (2020) 49:696–703
https://doi.org/10.1007/s13744-020-00765-y

SYSTEMATICS, MORPHOLOGY AND PHYSIOLOGY

A New and Rare *Actinote* Hübner (Lepidoptera: Nymphalidae: Heliconiinae: Acraeini) from Southeastern Brazil

AVL Freitas[1], RB Francini[2], OHH Mielke[3], AHB Rosa[1], LM Magaldi[1], KL Silva-Brandão[4]

[1]Depto de Biologia Animal and Museu de Zoologia, Instituto de Biologia, Univ Estadual de Campinas, Campinas, São Paulo, Brasil
[2]Lab de Biologia da Conservação, Univ Católica de Santos, Santos, São Paulo, Brasil
[3]Depto de Zoologia, Lab de Estudos de Lepidoptera Neotropical, Univ Federal do Paraná – UFPR, Curitiba, Paraná, Brasil
[4]Centro de Ciências Naturais e Humanas, Univ Federal do ABC, Santo André, São Paulo, Brasil

Keywords
Actinote eberti, *Actinote zikani*, Atlantic Forest, Itatiaia, Serra da Mantiqueira

Correspondence
A.V.L. Freitas, Depto de Biologia Animal and Museu de Zoologia, Instituto de Biologia, Univ Estadual de Campinas, Campinas, São Paulo13083-862, Brasil; baku@unicamp.br

Edited by Takumasa Kondo – CORPOICA

Received 6 December 2019 and accepted 6 February 2020
Published online: 28 May 2020

Abstract
The present paper describes a new species of *Actinote* (Nymphalidae, Heliconiinae, Acraeini), *Actinote keithbrowni* Freitas, Francini & Mielke **sp. nov.**, from southeastern Brazil, based on morphological and molecular data. The new species is very similar in wing pattern to *Actinote eberti*, within what we term the "light-gray mimicry complex." The host plants and immature stages of the new species are unknown. *Actinote keithbrowni* **sp. nov**. is one of the rarest species of *Actinote* from Brazil: in total, only eight individuals of this species are known from a narrow region of the Itatiaia massif despite years of collecting effort in that region since the 1940s.

Introduction

The butterfly genus *Actinote* Hübner [1819] contains a number of species that are particularly difficult to distinguish, especially females (e.g., D'Almeida 1925, 1935, Francini 1989, 1992, Penz and Francini 1996), often resulting in misidentifications and mixed series in collections that are identified as a single species (Francini 1989, Paluch 2006, Freitas *et al* 2018). This problem is particularly acute in montane areas of southeastern Brazil, where the genus reaches its greatest species richness and the wing patterns of some species are remarkably similar (D'Almeida 1935, Brown 1992, Lamas 2004, Francini & Penz 2006, Paluch 2006, Neild 2008, Silva-Brandão *et al* 2008, Willmott *et al* 2009). In recent years, however, several new species have been described from Brazil, clarifying some complexes of cryptic species, such as the "orangish red mimicry complex" (Penz & Francini 1996, Francini *et al* 2004, Paluch 2006, Freitas *et al* 2018). However, all these new species have been described based only on morphology; molecular data, although available for some species, have not been used so far.

On November 10, 1990 (about 10:00 am), a male *Actinote* with a wing pattern similar to a pale *Actinote eberti* Francini *et al* 2004 (at that time not yet described) was collected near the municipality of Penedo, a locality at the base of the Itatiaia massif, a high mountain in Rio de Janeiro State, southeastern Brazil. Subsequently, dissection of this individual revealed that the male genitalia were quite distinct from *A. eberti*, and very similar to that of *Actinote zikani* R.F. d'Almeida, 1951. Previously, therefore, that individual was recognized as belonging to a new taxon, but it has remained undescribed since then. Two decades later, on February 7, 2011, a trip to Itatiaia resulted in the collection of one female with a wing pattern very similar to that of the undescribed taxon, and subsequent searches in collections revealed a few additional specimens. Accordingly, the present paper describes this new species of *Actinote* from southeastern Brazil, based on both morphological and molecular data.

Material and Methods

Photographs of the male and female genitalia and some measurements were taken using a Zeiss® Discovery V20 Stereomicroscope. Dissections were made using standard techniques, where abdomens were soaked in hot 10% KOH solution for 10 min before dissection, and dissected parts were stored in glycerol. Taxonomic nomenclature follows Lamas (2004), modified after Wahlberg *et al* (2009). Male genitalia terminology follows Klots (1970). Nomenclature of venation used herein follows Wootton (1979) modified from Comstock & Needham (1898). Specimens of *Actinote* were examined in 13 public and private collections: CGCM, Coleção Carlos Guilherme Costa Mielke, Curitiba, Paraná, Brazil; CLDZ, Coleção de Lepidoptera, Departamento de Zoologia, Universidade Federal do Rio Grande do Sul, Porto Alegre, Rio Grande do Sul, Brazil; DZUP, Departamento de Zoologia, Universidade Federal do Paraná, Curitiba, Paraná, Brazil; IOC, Instituto Oswaldo Cruz, Rio de Janeiro, RJ, Brazil; MECB, Museu Entomológico Ceslau Biezanko, Departamento de Fitossanidade, Universidade Federal de Pelotas, Pelotas, Rio Grande do Sul, Brazil; MNHN, Muséum National d'Histoire Naturelle, Paris, France; MNRJ, Museu Nacional, Universidade Federal do Rio de Janeiro, Rio de Janeiro, Rio de Janeiro, Brazil, recently destroyed by fire (2-IX-2018); MZSP, Museu de Zoologia, Universidade de São Paulo, São Paulo, Brazil; NHMUK, The Natural History Museum, London, England; OM, Coleção Olaf Mielke, Curitiba, Paraná, Brazil; USNM, National Museum of Natural History, Smithsonian Institution, Washington, DC, USA; ZUEC, Museu de Zoologia da Universidade Estadual de Campinas, Unicamp, Campinas, São Paulo, Brazil; ZUEC-AVLF, André V. L. Freitas Collection, Universidade Estadual de Campinas, Campinas, São Paulo, Brazil. (see below). Besides the two recently collected individuals, specimens of the new species described here were found only in the DZUP, IOC, and OM. The Lamas collection of Neotropical butterfly type specimen photographs at the MUSM (also available online in Warren *et al* 2017), representing most currently relevant names and recognized species of *Actinote* (Lamas 2004), was examined.

To infer the phylogenetic position of the new species within *Actinote*, total genomic DNA was extracted, using the DNeasy Tissue Kit (Qiagen) and following the manufacturer's protocol, from legs of six individuals: one of the new species here described, three *A. zikani* and two *A. eberti* (the collection data and GenBank (Benson *et al* 2005) accession codes are shown in Table 1). DNA extractions were stored in TE buffer at − 20°C. The barcode region, which is the 5′ extremity of the mitochondrial DNA (mtDNA) gene cytochrome oxidase subunit I (COI, ca. 658 bp), was amplified using PCR reaction with the primers LCO 5′ (GGTCAACAAATCAT AAAGATATTGG) and NANCY (5′ CCTGGTAAAATTAAAATATA AACTTC) (Folmer *et al* 1994; Caterino & Sperling 1999). Sequences were examined and aligned using Geneious 10.0 (Kearse *et al* 2012). The final matrix comprised 30 specimens of *Actinote*, including the six above-mentioned individuals combined with several sequences of *Actinote* from Silva-Brandão *et al* (2008) (Table 1). A total of four genes was selected: the entire mitochondrial gene cytochrome oxidase I (COI—1508 bp), cytochrome oxidase II (COII—678 bp) and the nuclear genes elongation factor-1a (EF-1a—1240 bp) and wingless (wgl—403 bp). W-IQ-TREE (Trifinopoulos *et al* 2016) was used to perform maximum likelihood analyses (ML). Substitution models were determined for each partition (as in Rota *et al* 2018), using TIGER (Cummins & McInerney 2011) for sorting sites based on their relative evolutionary rates, and then using their algorithm RatePartitions for dividing the sites among partitions (d value = 1.35), applying ModelFinder (Kalyaanamoorthy *et al* 2017). IQ-TREE started the tree reconstruction under the best model scheme founded. Support for nodes was evaluated with 1000 ultrafast bootstrap (UFBoot2) approximations (Hoang *et al* 2017). The mean genetic distances of the barcode region among individuals were obtained using the software Mega 6.0 (Tamura *et al* 2013), with Kimura-2-Parameters substitution model (K2P, Kimura 1980).

Results

Actinote keithbrowni Freitas, Francini & Mielke, **sp. nov**. (Figs 1, 2, 3, 4, and 6)

Actinote eberti: Francini *et al* 2004: 3, fig. 1A

Actinote zikani ssp.: Francini *et al* 2005: 135, fig. 1, 136

Diagnosis

The general wing color and pattern of *A. keithbrowni* **sp. nov**. resemble those of *A. eberti*, exhibiting light beige areas over a gray ground color, distinguishing the new species from all other described species of *Actinote*. *Actinote keithbrowni* **sp. nov**. can be distinguished from *A. eberti* by the following characters (see Fig 2): (1) the forewing subapical light band is broader and continuous in *A. keithbrowni* **sp. nov**., while being narrower and not continuous in *A. eberti*; (2) the light stripe in the forewing discal cell is broad in *A. keithbrowni* **sp. nov**., while it is narrow in *A. eberti*; (3) on dorsal forewing, a small half-moon-shaped light beige patch is present at the tornus in *A. eberti*, while being absent in *A. keithbrowni* **sp. nov**.; (4) the hindwing black margin is not as broad as in *A. eberti*; (5) the central light-beige area in the hindwings is more homogeneous and with well-defined limits in *A. keithbrowni* **sp. nov**., while in *A. eberti* a series of broader and darker intervenal lines results in more poorly defined limits.

Table 1 Actinote specimens studied, with voucher code, sampling site data, and GenBank accession numbers for the four genes sequenced.

Code	Species	Country	Locality	GenBank accession no.		
				COI-COII	EF-1α	Wingless
ac-77	*Actinote alalia*	Brazil	Curitiba, Paraná	EU275617	EU275617	EU275701
ac-14	*Actinote anteas*	Colombia	El Carmen, Antioquia	EU275613	EU275613	EU275697
NW137-24	*Actinote bonita*	Brazil	Itatiaia, Rio de Janeiro	EU275549	EU275549	–
ac-4	*Actinote brylla*	Brazil	Vale do Rio Quilombo, Santos, São Paulo	EU275576	EU275576	EU275656
ac-86	*Actinote canutia*	Brazil	Itirapina, São Paulo	EU275577	EU275577	EU275657
ac-88	*Actinote carycina*	Brazil	Campos do Jordão, São Paulo	EU275550	EU275550	EU275659
ac-10	*Actinote conspicua*	Brazil	Campos do Jordão, São Paulo	EU275580	EU275580	EU275661
ac-12	*Actinote dalmeidai*	Brazil	Paranapiacaba, Santo André, São Paulo	EU275582	EU275582	EU275663
ac-7	*Actinote discrepans*	Brazil	Campos do Jordão, São Paulo	EU275583	EU275583	EU275664
BPU15	*Actinote eberti*	Brazil	São Bento do Sapucaí, São Paulo	MN781111	–	–
BLU-0969	*Actinote eberti*	Brazil	Campos do Jordão, São Paulo	MN781114	–	–
ac-65	*Actinote genitrix*	Brazil	FLONA, Passa Quatro, Minas Gerais	EU275584	EU275584	EU275665
NW155-1	*Actinote guatemalena*	Mexico	Chiapas, Bonam Park	EU275585	EU275585	EU275667
AC-233	*Actinote keithbrowni*	Brazil	Itatiaia, Rio de Janeiro	MN781110	–	–
ac-95	*Actinote kennethi*	Ecuador	Napo	EU275619	EU275619	EU275703
ac-35	*Actinote mamita*	Brazil	Santa Genebra, Campinas, São Paulo	EU275586	EU275586	EU275668
ac-36	*Actinote mantiqueira*	Brazil	Pico do Itapeva, Pindamonhangaba, São Paulo	EU275575	EU275575	EU275655
ac-84	*Actinote melanisans*	Brazil	São Bernardo do Campo, São Paulo	EU275587	EU275587	EU275670
ac-93	*Actinote morio*	Brazil	Peti, Minas Gerais	EU275589	EU275589	EU275672
ac-23	*Actinote parapheles*	Brazil	Vale do Rio Quilombo, Santos, São Paulo	EU275591	EU275591	EU275674
ac-71	*Actinote pellenea epiphaea*	Peru	Abancay Anpay, Apurimac	EU275595	EU275595	EU275678
ac-78	*Actinote pratensis*	Brazil	Águas da Prata, São Paulo	EU275602	EU275602	EU275685
ac-72	*Actinote pyrrha crucis*	Brazil	Vicosa do Ceará, Ceará	EU275603	EU275603	EU275686
ac-80	*Actinote quadra*	Brazil	Campos do Jordão, São Paulo	EU275607	EU275607	EU275690
ac-8	*Actinote rhodope*	Brazil	Campos do Jordão, São Paulo	EU275608	EU275608	EU275691
ac-48	*Actinote surima perisa*	Argentina	Yala, Jujuy	EU275609	EU275609	EU275692
BLU-0963	*Actinote zikani*	Brazil	Paranapiacaba, Santo André, São Paulo	MN781112	–	–
BLU-0966	*Actinote zikani*	Brazil	Paranapiacaba, Santo André, São Paulo	MN781113	–	–
BLU-1043	*Actinote zikani*	Brazil	Paranapiacaba, Santo André, São Paulo	MN781115	–	–
ac-83	*Actinote zikani*	Brazil	Paranapiacaba, Santo André, São Paulo	EU275616	EU275616	EU275700
NW90-14	*Altinote stratonice*	Ecuador	La Bonita	AY218233	AY218233	AY218252

Description

Male (Figs 1A, 2A, and 6A): Antenna black, 11–13 mm in length ($n = 3$), extending to mid-costa, with 34 antennomeres, 9 of which form a well-defined club. Forewing narrow and elongated, length 30–33 mm ($n = 3$); hindwing rounded, about two-thirds length of forewing, length 21–23 mm ($n = 3$). Body dark brown. Forewing dorsal ground color light gray, with light beige patches as follows: a broad subapical band, extending from costal margin to CuA1; a small squared patch in distal part of discal cell; a large translucent patch in space 2A-CuA2, extending halfway to wing margin; a broad stripe extending to half-length of discal cell. Hindwing dorsal background light gray with a large light beige central area delimiting a broad marginal band, several broad light-gray stripes extending in intervenal spaces including one stripe in discal cell. Ventral wings with a pattern very similar to dorsal pattern.

Male genitalia (Fig 3): Valval length about four times width of median portion; curved, broad at base, narrowing toward apex; apex pointed. In dorsal view, basal portion of uncus wide, abruptly narrowing, ending in a point. Tegumen long and broad, trapezoidal. Gnathos absent. In ventral view, saccus shaped as an isosceles triangle, about half-length of genital capsule. Aedeagus about length of genital capsule, ending in a sharp point, slightly curved ventrally in lateral view. Fultura inferior broad, diamond shaped; length equal to width.

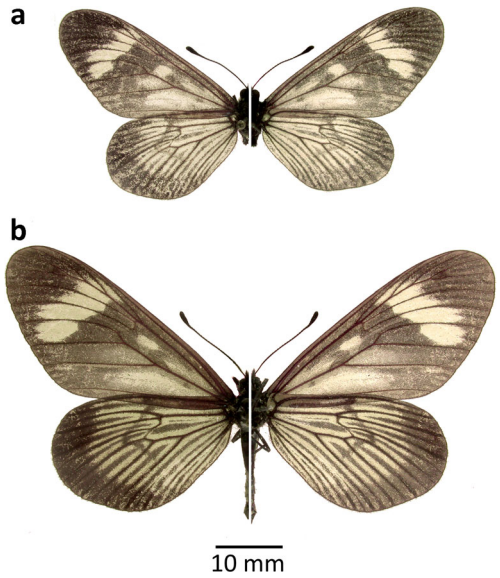

Fig 1 (A) Holotype male and (B) paratype female (DZ 5.982) of *Actinote keithbrowni* **sp. nov**. (left = dorsal; right = ventral).

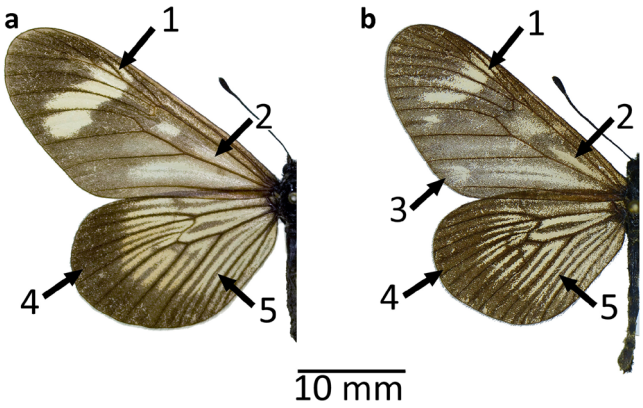

Fig 2 Principal differences in wing pattern between *Actinote keithbrowni* **sp. nov**. and *Actinote eberti*. (A) Male paratype of *A. keithbrowni* **sp. nov**. (DZ 6.013) (dorsal); (B) Male holotype of *A. eberti* (dorsal). Numbers refer to diagnostic characters explained in the text.

Female (Fig 1B): Antenna black, 12–14 mm in length ($n = 5$), extending to mid-costa, with 33 antennomeres, 10 in club. Forewing narrow and elongated, length 33–38 mm ($n = 5$). Hindwing rounded, not translucent, length 24–27 mm ($n = 5$). Color pattern of female wings similar to that of males.

Systematic position

Based on molecular data, *A. keithbrowni* **sp. nov**. is the sister species of *A. zikani*, with a mean genetic distance of 0.57% between them (Fig 4). This close relationship is highly supported and reinforced by the morphology of male genitalia, which is very similar to that of *A. zikani*.

Taxonomy and variation

As mentioned before, individuals of *A. keithbrowni* **sp. nov**. are very similar in wing pattern to *A. eberti*, and both species were found in mixed series in DZUP and IOC. Intraspecific variation is low among the few examined individuals.

Holotype (Fig 1A). Male from Itatiaia, Rio de Janeiro, Brazil. Deposited at the DZUP. Labels on the holotype (eight labels separated by transverse bars): / HOLOTYPUS / Holotypus *Actinote keithbrowni* Freitas, Francini & Mielke, det. 2019/ Serra do Itatiaia (R.[io de] J.[aneiro] 800 m, 23 3. 67 (Ebert) / Coleção H. Ebert / *Actinote eberti* Francini *et al* 2004 Paluch *det*. 2004 / Genitália Preparada M. Paluch 2003 / 421 / DZ 6.574 /.

Paratypes (All from Rio de Janeiro, Brazil). Itatiaia, Penedo, 400 m, 1 male, 10.XI.1990, 22°26′S 44°30″W RB Francini, AVL Freitas, CF Klitzke & KS Brown Jr leg. (ZUEC LEP 11046) (ZUEC); Parque Nacional do Itatiaia, [cachoeira da] Maromba, 1 female, 7.II.2011, 1130 m, 22°25′43″S 44°37′10″W, A. V. L. Freitas leg. (DNA voucher AC 233) (ZUEC-AVLF);

Fig 3 Male genitalia of *Actinote keithbrowni* **sp. nov**., lateral view (top), dorsal view (middle), ventral view (bottom). ae, aedeagus; sa, saccus; un, uncus; va, valva.

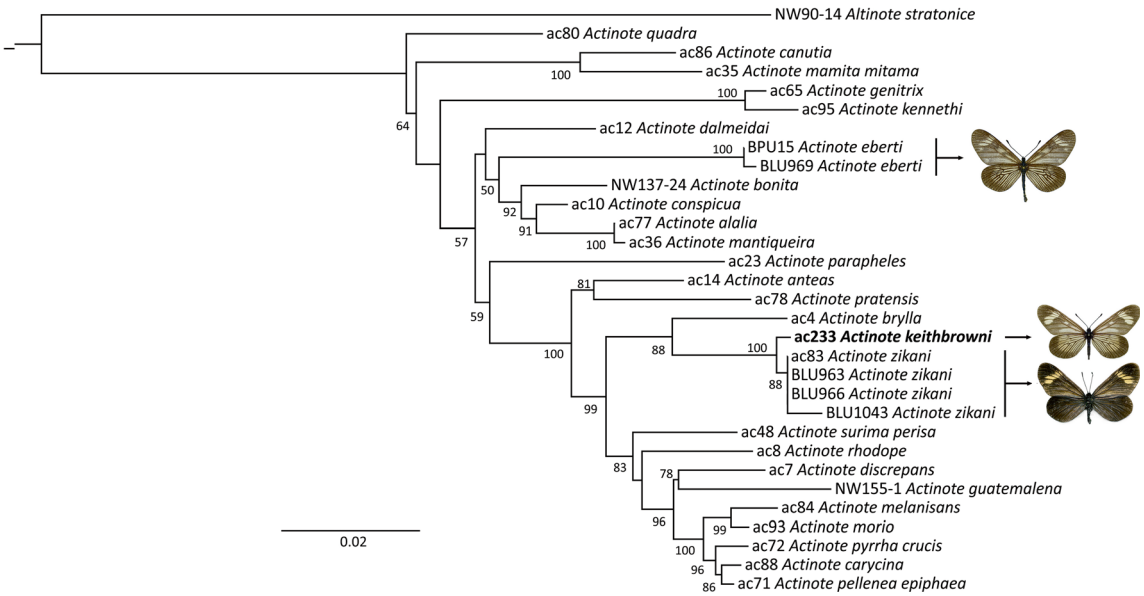

Fig 4 Phylogenetic relationships of *Actinote* based on four genes and obtained by a maximum likelihood analysis. Numbers on branches refer to bootstrap values (data from present study and from Silva-Brandão *et al* 2008).

Itatiaia, Parque Nacional do Itatiaia, 1 male (with an attached female abdomen, glued on), 23.III.1967, 800 m, H. Ebert leg. (DZ 6.013) (Fig 2A); 1 female, 26.III.1967, 800 m, H. Ebert leg. (DZ 5.982) (Fig 1B); Itatiaia, Parque Nacional do Itatiaia, 1 female, 29.V.1946, 700 m, J. F. Zikán leg. (N27.148) (IOC); 1 female, 21.VII.1961, 1 female, 18.VII.1963, 900 m, O. H. H. Mielke leg. (3857, 5337) (OM).

Habitat and natural history

Based on the few available data, *A. keithbrowni* **sp. nov**. is apparently associated with wet montane and submontane forests from 400 to 1100 m (Fig 5). Based on collecting records, adults are possibly multivoltine, with records from February, March, May, July, and November. With only eight known individuals the species can be considered rare, possibly occurring at low densities. For example, after the capture of a female individual in February 2011 (one of the paratypes, see above), on a bridge near the Maromba falls in Itatiaia National Park, no additional individuals were observed in the five following days, even with hours of collecting effort at the same spot and in other similar habitats in the surroundings. With its unusual wing pattern, *A. keithbrowni* **sp. nov**. is likely a co-mimic of *A. eberti*, forming what we here call the "light-gray mimicry complex," a pattern also occurring in individuals that lack typical orange/yellow wing patches of the sympatric species *Actinote carycina* Jordan, 1913, *Actinote melanisans* Oberthür, 1917 and *Actinote rhodope* R.F. d'Almeida, 1923 (see Francini 1989) (Fig 6). The host plant and immature stages are unknown.

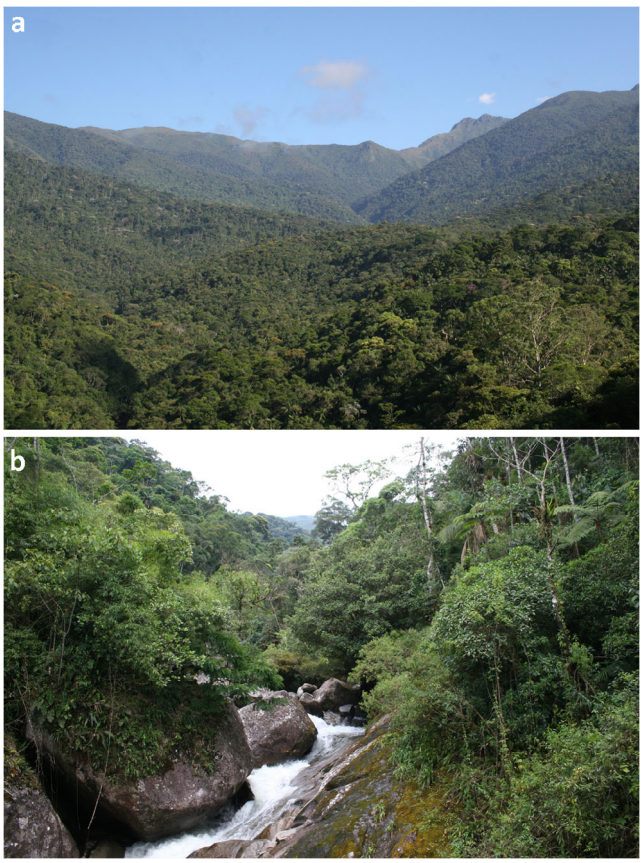

Fig 5 Habitat of *Actinote keithbrowni* **sp. nov**. in the region of the Itatiaia massif, Rio de Janeiro. (A) General view of the forests, (B) close view of the habitat near the Maromba falls, where a female specimen was collected.

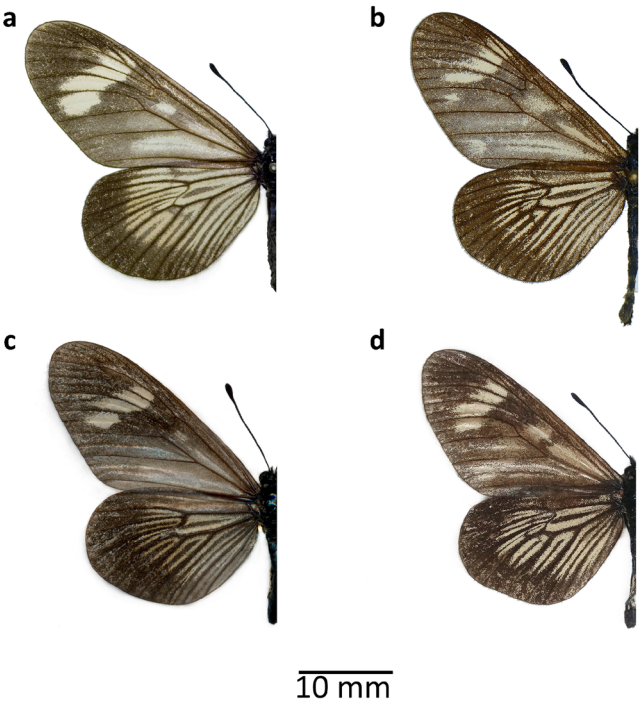

Fig 6 Males of several species of *Actinote* belonging to the "light-gray mimicry complex." (A) *Actinote keithbrowni* **sp. nov.**, (B) *Actinote eberti*, (C) rare light gray phenotype of *Actinote rhodope*, (D) rare light gray phenotype of *Actinote carycina*.

Geographic distribution

The species is only known from the region of the Itatiaia massif, in the Serra da Mantiqueira mountain range, and except for one specimen from the neighborhood of Penedo, all other individuals were collected inside Itatiaia National Park, Rio de Janeiro.

Etymology. This species is dedicated to Dr. Keith S. Brown Jr., a North American scientist who came to live in Brazil in 1964 and subsequently stayed in that country, profoundly influencing several generations of butterfly researchers during his scientific life. Keith Brown was also the organizer of the field trip in November 1990 which resulted in the collection of the individual that would be the trigger for the recognition of this new species (see "Introduction").

Discussion

The description of a new species of *Actinote* is not remarkable in itself, since species in this genus can be notoriously difficult to identify and museum collections often have unidentified specimens (see Freitas *et al* 2018 and references therein). In the present case, *A. keithbrowni* **sp. nov**. was confused with the distantly related *A. eberti*, with which it shares a similar color pattern (Francini *et al* 2004, Paluch 2006), but not with *A. zikani*, its sister species (see Fig 4).

A noteworthy aspect concerning the present study is the apparent rarity of *A. keithbrowni* **sp. nov**.: in total, only eight individuals of this species are known despite years of collecting effort in the Itatiaia region since the 1940s (Zikán & Zikán 1968, KSB, OHHM, AVLF and AHBR, unpublished data). Considering that its co-mimic, *A. eberti*, is also extremely scarce (less than 20 individuals known in all museum collections), and this wing color pattern appears as a very occasional variation of some other species of *Actinote* (see above), the "light-gray mimicry complex" appears to be very rare in southeastern Brazil.

The rarity of both *A. keithbrowni* **sp. nov**. and *A. eberti* deserves attention. Since they are gregarious in all life stages, most species of *Actinote* can be abundant or at least locally common (D'Almeida 1935, Francini 1989, 1992, Brown 1992, Brown & Francini 1990, Francini & Freitas 2010). This is also true for some threatened species, such as *A. zikani* (Francini *et al* 2005, 2011) and *A. quadra* (Schaus, 1902) (Freitas *et al* 2009), which are locally abundant and thus usually easily detected when present. It is possible that the apparent rarity of *A. keithbrowni* **sp. nov**. is related to an asynchrony between flying adults and collecting expeditions which undoubtedly contributed to the delay in rediscovering the critically endangered *A. zikani* (Francini *et al* 2005), and kept two conspicuous species of *Actinote* undetected for decades in an intensively sampled region of Central Brazil (e.g., Freitas *et al* 2019). Another possibility is that populations of *A. keithbrowni* **sp. nov**., and also of its co-mimic *A. eberti*, undergo extreme fluctuations in the number of mature individuals through time, as also reported for *A. zikani* (Francini *et al* 2005). Finally, it is possible that both species (*A. keithbrowni* **sp. nov.** and *A. eberti*) might require particular microclimatic conditions near the host plants where the immature stages can complete their development (as also suggested for *A. zikani*, Francini *et al* 2011), and these ideal conditions do not occur every year. In this case, unless researchers are in the field during years of high abundance (as suggested for *A. zikani*, see Francini *et al* 2005), the few observed adults could be neglected or mistakenly considered light variations of other, more common species.

Lastly, the clade formed by *A. keithbrowni* **sp. nov**. and *A. zikani* includes the two species of *Actinote* from southeastern Brazil with the most restricted known geographical distributions. In fact, both species are known from only two localities each. If both species really are genuinely narrowly distributed, given the combination of (1) restricted area of occupancy, (2) few known populations, (3) small total population size, and (4) extreme fluctuations in number of mature individuals and recognized population threats, the newly described *A. keithbrowni* **sp. nov**. could be a candidate for recognition as a threatened species (IUCN Standards and

Petitions Committee 2019). However, before a formal recommendation can be made, a proper evaluation should be conducted by collecting additional specimens and performing intensive field studies, particularly targeting the discovery of the host plant and immature stage biology.

Acknowledgments We thank Clécio F. Klitzke and Keith S. Brown Jr. for helping in field work. To Marcio Felix for helping with material from the IOC. Tamara M. C. Aguiar for help spreading specimens. RBF thanks to Universidade Católica de Santos for logistic support. Keith R. Willmott carefully read the last version of the manuscript. The authors also thank the ICMBio for the permits (#10438-1 and #10802-24) and Léo Nascimento (Parque Nacional do Itatiaia) for providing logistic support during field work. This publication is part of the "RedeLep - Rede Nacional de Pesquisa e Conservação de Lepidópteros" - SISBIOTA-Brasil/CNPq (563332/2010-7). Butterfly species are registered in the SISGEN (A0E7804).

Nomenclature **Publication:** http://zoobank.org/urn:lsid:zoobank.org:pub: A2DFF815-29E0-4B94-B286-9BF6C750CF70

Nomenclatural act: http://zoobank.org/urn:lsid:zoobank.org:act:999AC802-E425-454E-B7BF-A6A17CBD9B10

Author's Contribution All authors revised critically the content, approved the version to be published, and agree to be accountable for all aspects of the work in ensuring that questions related to theaccuracy or integrity of any part of this research are appropriately investigated and resolved.

Funding Information AVLF acknowledges support from Fundação de Amparo à Pesquisa do Estado de São Paulo – (FAPESP) (Biota-Fapesp grants 2011/50225-3, 2013/50297-0) and from the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) (303834/2015-3). LMM acknowledges FAPESP (PhD scholarship - 2015/24663-4). AHBR thanks Coordenação de Aperfeiçoamento de Pessoal de Nível Superior - Brazil (CAPES) - Finance Code 001. OHHM acknowledges support from the – CNPq (304639/2014-2).

References

Benson DA, Karsch-Mizrachi I, Lipman DJ, Ostell J, Wheeler DL (2005) GenBank. Nucleic Acids Res 33(DATABASE ISS):34–38

Brown KS Jr (1992) Borboletas da Serra do Japi: diversidade, habitats, recursos alimentares e variação temporal. In: Morellato LPCO (ed) História natural da Serra do Japi. Unicamp, Fapesp, Campinas, SP, pp 142–187

Brown KS Jr, Francini RB (1990) Evolutionary strategies of chemical defense in aposematic butterflies: cyanogenesis in Asteraceae-feeding American Acraeinae. Chemoecology 1(2):52–56

Caterino MS, Sperling FAH (1999) Papilio phylogeny based on mitochondrial cytochrome oxidase I and II genes. Mol Phylogenet Evol 11(1): 122–137

Comstock JH, Needham JG (1898) The wings of insects. Am Nat 32(373): 43–48

Cummins CA, McInerney JO (2011) A method for inferring the rate of evolution of homologous characters that can potentially improve phylogenetic inference, resolve deep divergence and correct systematic biases. Syst Biol 60(6):833–844

D'Almeida RF (1925) Quelques rectifications sur les Actinote de la partie orientale deL'Amérique du Sud (Lép. Rhopalocères). Ann Soc Entomol Fr 94(3):333–354

D'Almeida RF (1935) Les *Actinote* de la partie orientale de L'Amérique du Sud. An Acad Bras Cienc 7(1):69–88 (31 March), (2):89-112 + 13 plates (30 June)

Folmer O, Black M, Hoeh W, Lutz R, Vrijenhoek R (1994) DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. Mol Mar Biol Biotechnol 3(5):294–299

Francini RB (1989) Biologia e ecologia das borboletas *Actinote* (Lepidoptera: Nymphalidae, Acraeinae) do sudeste do Brasil. Ms Thesis. Universidade Estadual de Campinas, Campinas

Francini RB (1992) Ecologia das taxocenoses de *Actinote* (Lepidoptera: Nymphalidae) em Asteraceae (Angiosperma, Magnoliatae) no sudeste do Brasil: subsídios para conservação. PhD Thesis. Unicamp, Campinas

Francini RB, Freitas AVL (2010) Aggregated oviposition in *Actinote pellenea pellenea* Hübner (Lepidoptera: Nymphalidae). J Res Lepid 42:74–78

Francini RB, Penz CM (2006) An illustrated key to male *Actinote* from southeastern Brazil (Lepidoptera, Nymphalidae). Biota Neotrop 6(1): 46. http://www.biotaneotropica.org.br/v6n1/pt/abstract?identification-key+bn00606012006. Accessed 25 Nov 2019

Francini RB, Freitas AVL, Penz CM (2004) Two new species of *Actinote* (Lepidoptera, Nymphalidae) from southeastern Brasil. Zootaxa 719:1–10

Francini RB, Freitas AVL, Brown KS Jr (2005) Rediscovery of *Actinote zikani* (D'Almeida) (Nymphalidae, Heliconiinae, Acraeini): natural history, population biology and conservation of an endangered butterfly in SE Brazil. J Lepid Soc 59(3):134–142

Francini RB, Barbosa EP, Freitas AVL (2011) Immature stages of *Actinote zikani* (Nymphalidae: Heliconiinae), a critically endangered butterfly from southeastern Brazil. TropLepid Res 21(1):20–26

Freitas AVL, Francini RB, Souza TS (2009) Immature stages and natural history of the threatened butterfly *Actinote quadra* (Nymphalidae: Heliconiinae: Acraeini). Trop Lepid Res 19:83–88

Freitas AVL, Francini RB, Paluch M, Barbosa EP (2018) A new species of *Actinote* Hübner (Nymphalidae: Heliconiinae: Acraeini) from Southeast Brazil. Rev Bras Entomol 62:135–147

Freitas AVL, Gueratto PE, Nascimento AR (2019) New records of high altitude *Actinote* (Nymphalidae: Heliconiinae) from Central Brazil extend the known distribution by 650 km. J Lepid Soc 73(3):208–210

Hoang DT, Vinh LS, Chernomor O, Minh BQ, von Haeseler A (2017) UFBoot2: improving the ultrafast bootstrap approximation. Mol Biol Evol 35(2):518–522

IUCN Standards and Petitions Committee (2019) Guidelines for using the IUCN red list categories and criteria. Version 14. Prepared by the Standards and Petitions Committee. Downloadable from: http://www.iucnredlist.org/documents/RedListGuidelines.pdf. Accessed 25 Nov 2019

Kalyaanamoorthy S, Minh BQ, Wong TKF, von Haeseler A, Jermiin LS (2017) ModelFinder: fast model selection for accurate phylogenetic estimates. Nat Methods 14:587

Kearse M, Moir R, Wilson A, Stones-Havas S, Cheung M et al (2012) Geneious basic: an integrated and extendable desktop software platform for the organization and analysis of sequence data. Bioinformatics 28(12):1647–1649

Klots AB (1970) Lepidoptera. In: Tuxen SL (ed) Taxonomist's glossary of genitalia in insects. Munksgaard, Copenhagen, pp 115–130

Lamas G (2004) Checklist: part 4A. Hesperioidea-Papilionoidea. In: Heppner JB (ed) Atlas of Neotropical Lepidoptera, Vol 5A. Association for Tropical Lepidoptera/Scientific Publishers, Gainesville, xxxvi +, p 439

Neild AFE (2008) The butterflies of Venezuela, part 2: Nymphalidae II (Acraeinae, Libytheinae, Nymphalinae, Ithomiinae, Morphinae). Meridian Publications, Greenwich, London, p 272

Paluch M (2006) Revisão das espécies de *Actinote* Hübner, [1819] (Lepidoptera, Nymphalidae, Heliconiinae, Acraeini). PhD Thesis. Universidade Federal do Paraná, Curitiba

Penz CM, Francini RB (1996) New species of *Actinote* Hübner (Nymphalidae: Acraeinae) from southeastern Brazil. J Lepid Soc 50(4):309–320

Rota J, Malm T, Chazot N, Peña C, Wahlberg N (2018) A simple method for data partitioning based on relative evolutionary rates. PeerJ 6: e5498

Silva-Brandão KL, Wahlberg N, Francini RB, Azeredo-Espin AML, Brown KS Jr, Paluch M, Lees DC, Freitas AVL (2008) Phylogenetic relationships of butterflies of the tribe Acraeini (Lepidoptera, Nymphalidae, Heliconiinae) and the evolution of host plant use. Mol Phylogenet Evol 46:515–531

Tamura K, Stecher G, Peterson D, Filipski A, Kumar S (2013) MEGA6: molecular evolutionary genetics analysis version 6.0. Mol Biol Evol 30(12):2725–2729

Trifinopoulos J, Nguyen LT, von Haeseler A, Minh BQ (2016) W-IQ-TREE: a fast online phylogenetic tool for maximum likelihood analysis. Nucleic Acids Res 44(W1):W232–W235

Wahlberg N, Leneveu J, Kodandaramaiah U, Peña C, Nylin S, Freitas AVL, Brower AVZ (2009) Nymphalid butterflies diversify following neardemise at the Cretaceous/Tertiary boundary. Proc R Soc Lond B 276:4295–4302

Warren AD, Davis KJ, Stangeland EM, Pelham JP, Willmott KR, Grishin NV (2017) Illustrated Lists of American Butterflies (North and South America) 10-XII-2017. http://butterfliesofamerica.com/L/Neotropical.htm. Accessed 25 Nov 2019

Willmott KR, Freitas AVL, Hall JPW, Silva-Brandão KL, Paluch M (2009) A new species of *Actinote* Hübner from the Eastern Andes of Ecuador (Lepidoptera: Nymphalidae: Heliconiinae). Proc Entomol Soc Wash 111:47–56

Wootton RJ (1979) Function, homology and terminology in insect wings. Syst Entomol 4:81–93

Zikán JF, Zikán W (1968) Inseto-fauna do Itatiaia e da Mantiqueira. III Lepidoptera. Pesq Agropec Bras (Agronomia) 3:45–109