# Plucking with the plectrum: phylogeny of the New World buthid scorpion subfamily Centruroidinae Kraus, 1955 (Scorpiones: Buthidae) reveals evolution of three pecten-sternite stridulation organs 

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#### Abstract

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#### Abstract

All New World buthid scorpions except one South American genus, Ananteris Thorell, 1891, comprise a monophyletic group. The monophyly of two subfamilies, Centruroidinae Kraus, 1955 (= Rhopalurusinae Bücherl, 1971) and Tityinae Bücherl, 1971, proposed to accommodate a subset of these genera, has never been tested. The genera accommodated within Centruroidinae are diverse and poorly defined. Prior to the research presented here, Rhopalurus Thorell, 1876 had a disjunct distribution in the Greater Antilles, the Guiana Shield of northern South America, and northeastern Brazil, where Physoctonus Mello-Leitão, 1934 and Troglorhopalurus Lourenço et al., 2004 also occur. The generic distinction between Rhopalurus and Centruroides Marx, 1890, the most speciose genus of Centruroidinae, distributed from the midwestern United States to northern South America, and throughout the Caribbean, was also unclear. Previous studies suggested Centruroides was paraphyletic with respect to Rhopalurus and vice versa. The study presented here, the first rigorous test of the monophyly of Centruroidinae and its component genera, is based on 90 morphological characters and 4,260 aligned base-pairs of DNA sequence from three mitochondrial and two nuclear DNA loci for 102 terminal taxa, representing 24 species in seven ingroup genera, and nine species in three outgroup genera. Molecular and morphological data, analyzed separately and simultaneously, yielded congruent results. Centruroidinae was monophyletic whereas Tityinae was paraphyletic. Centruroides was monophyletic whereas Rhopalurus was paraphyletic, comprising several monophyletic groups congruent with its disjunct distribution. The results of this analysis justify the redefinition of Rhopalurus and Troglorhopalurus, the revalidation of Heteroctenus Pocock, 1893, and the recently created genera Ischnotelson Esposito et al., 2017 and Jaguajir Esposito et al., 2017. The phylogeny indicates that three distinct types of pecten-sternite stridulation organ evolved in Heteroctenus, Jaguajir and Rhopalurus.


Key words. New World, savannah, Arachnida, Rhopalurus, systematics.

## 1. Introduction

The family Buthidae C.L. Koch, 1837, comprising 89 genera and approximately 1,110 described species of extant scorpions, is distributed worldwide and includes most of the world's medically important scorpions. Although the intrafamilial phylogenetic relationships of Buthidae remain largely untested, there is general agreement that
all New World buthids except one genus occurring in South America, Ananteris Thorell, 1891 (Microananteris Lourenço, 2003 was justifiably synonymized with Ananteris by Botero-Trujillo \& Noriega 2011), comprise a monophyletic group, referred to hereafter as the "New World buthids" (Coddington et al. 2004). This diverse


Fig. 1. Male pedipalp chela, retrolateral aspect, and movable finger, dorsal aspect of species in the New World buthid scorpion subfamily Centruroidinae Kraus, 1955, illustrating shapes and denticle rows. A: Physoctonus debilis (C.L. Koch, 1840). B: Physoctonus striatus Esposito et al., 2017. C, D: Jaguajir rochae (Borelli, 1910). E: Ischnotelson guanambiensis (Lenarducci et al., 2005). F: Tityus serrulatus Lutz \& Mello, 1922.
group, originally named Centrurini Kraepelin, 1891, is endemic to the New World and comprises approximately 398 described species in 14 genera: Alayotityus Armas, 1973; Centruroides Marx, 1890; Chaneke Francke et al., 2014; Heteroctenus Pocock, 1893; Ischnotelson Esposito et al., 2017; Jaguajir Esposito et al., 2017; Mesotityus Gonzalez-Sponga, 1981; Microtityus Kjellesvig-Waer-
ing, 1966; Physoctonus Mello-Leitão, 1934; Rhopalurus Thorell, 1876; Tityopsis Armas, 1974; Tityus C.L. Koch, 1836; Troglorhopalurus Lourenço et al., 2004; Zabius Thorell, 1893.

The monophyly of New World buthids, excluding Ananteris, is supported by evidence from multiple independent sources. Comparative morphological studies of


Fig. 2. Male metasoma, dorsal aspect, of species in the New World buthid scorpion subfamily Centruroidinae Kraus, 1955, illustrating posterior widening. A: Rhopalurus ochoai Esposito et al., 2017. B: Ischnotelson guanambiensis (Lenarducci et al., 2005). C: Jaguajir pintoi (Mello-Leitão, 1932).
pedipalp trichobothrial patterns (Fet et al. 2005), book lungs (Kamenz \& Prendini 2008), and ovariuterine structure (Volschenk et al. 2008) revealed potential synapomorphies, toxicological studies demonstrated the presence of a unique toxin class ( $\beta$ toxins), absent in other buthids (Froy et al. 1999), and phylogenetic analyses of transcriptomes (Sharma et al. 2015) and nuclear and/or mitochondrial DNA sequences recovered its monophyly (Fet et al. 2003a; Coddington et al. 2004; Borges \& Graham 2016; Ojanguren-Affilastro et al. 2017).

Although the monophyly of New World buthids has never been contested, there is less agreement about the names applied to the clade. Bücherl (1971) proposed two subfamilies to accommodate the four genera of New World buthids recognized at the time. Rhopalurinae, comprising Centruroides and Rhopalurus, was defined by the presence of pro- and retrolateral accessory (or supernumerary) denticles in the median denticle rows of the pedipalp chela fingers (Fig. 1B,D), whereas Tityinae Bücherl, 1971, comprising Tityus and Zabius, was defined by their absence (Fig. 1F). A great deal of confusion has surrounded the nomenclature for the New World buthid clade containing the genera Centruroides and Rhopalurus. Kraepelin (1891) first proposed Centrurini, subsequently emended to Centrurinae (Kraepe-
lin 1899), on Centrurus Ehrenberg, 1829. Kraepelin (1894) later synonymized Centrurus with Heterometrus Ehrenberg, 1828 of family Scorpionidae Latreille, 1802. Centruroides Marx, 1891 was meanwhile proposed, not as a replacement name for Centrurus, but for Buthus exilicauda Wood, 1863, a North American species, and another species that was named but not described (Fet \& Lowe 2000: 98). The name Centrurus was incorrectly used for many years to denote species of Centruroides because of an erroneous understanding and placement of Centrurus. Centruroidinae was considered a nomen nudum because it was first proposed after 1930 and Roewer (1943) omitted a description from the designation, hence Fet \& Lowe (2000: 57) suggested Rhopalurinae should be used instead. Fet et al. (2003b) emended Rhopalurinae to Rhopalurusinae to avoid homonymy. Esposito et al. (2017), following Fet \& Lowe (2000) and Fet et al. (2003b), used Rhopalurusinae. Soon after, Armas (2017) synonymized Rhopalurusinae with Centruroidinae and considered Kraus (1955) the author of the latter, based on a brief description published in a footnote. Following arguments set forth in more detail in Appendix 1, the valid subfamily name is Centruroidinae Kraus, 1955, and the names Rhopalurinae and Rhopalurusinae, are junior synonyms thereof. On the other hand, Centruri-


Fig. 3. Sternite III, ventral aspect, illustrating pars stridens of pecten-sternite stridulation organs of species in the New World buthid scorpion subfamily Centruroidinae Kraus, 1955. A: Ischnotelson guanambiensis (Lenarducci et al., 2005), đ (MZSP). B: Jaguajir agamemnon (C.L. Koch, 1839), $q$ (MZSP). C: Heteroctenus junceus (Herbst, 1800), $q$ (AMNH). D: Rhopalurus ochoai Esposito et al., 2017, holotype ठ (AMNH).
nae is synonymous with Scorpionidae not Buthidae, as it is based on Centrurus, an available junior synonym of Heterometrus (Braunwalder \& Fet 1998; Fet 2000; Fet \& Lowe 2000). Centrurus is not a senior synonym of Centruroides or a nomen nudum as Francke (1985) suggested (see also ICZN 1986: 144, 145).

Additional genera, described after the creation of these subfamilies, were not formally accommodated within them. No subfamilial classification of Buthidae is in widespread use (Fet \& Lowe 2000) as the monophyly of buthid subfamilies has never been rigorously tested. Four studies, each using a different data source and taxon sample, and in each case representing a very small sample of the taxonomic diversity within the family, recovered different results (Fet et al. 2003a; Sharma et al. 2015; Borges \& Graham 2016; Ojanguren-Affilastro et al. 2017). It has long been suspected that the group of taxa herein referred to Centruroidinae may be monophyletic, however, due to the presence of accessory denticles in the median denticle rows of the pedipalp chela fingers, unique to these taxa among Buthidae (Sissom 1990).

Prior to the research presented here, the genera accommodated within Centruroidinae were also poorly defined. The 'club-tailed' scorpions of the genus Rhopalurus and the related genera, Physoctonus and Troglorhopalurus, were a morphologically heterogeneous group of mostly large-bodied, often strikingly colored scorpions, usually with a broad metasoma that becomes noticeably wider posteriorly and an incrassate pedipalp chela in the adult male (Figs. 1, 2). Rhopalurus comprised 23 species and two subspecies whereas Physoctonus and Troglorhopalurus were monotypic (Table 1). The unique ability to stridulate audibly by scraping nodules and/or ridges on the dorsal surfaces of the pectinal teeth against granules on the ventral surfaces of mesosomal sternite III (Figs. $3-5$ ), a remarkable behavior that presumably functions to deter would-be predators (Рососк 1904; Lourenço \& Cloudsley-Thompson 1995; Lourenço 2007; Prendini et al. 2009), was considered synapomorphic for Rhopalurus by Lourenço (1986) although this had not been tested quantitatively.

Physoctonus, long regarded a junior synonym of Rhopalurus (Francke 1977), was recently resurrected (Lou-


Fig. 4. Dextral pecten, ventral aspect ( $\mathrm{A}, \mathrm{C}, \mathrm{E}, \mathrm{G}$ ), and pectinal teeth, dorsal aspect ( $\mathrm{B}, \mathrm{D}, \mathrm{F}$ ), illustrating plectrum of pecten-sternite stridulation organs of species in the New World buthid scorpion subfamily Centruroidinae Kraus, 1955. A: Heteroctenus junceus (Herbst, 1800), ठ̃ (AMNH). B: Heteroctenus bonettii (Armas, 1999), ¢ (AMNH). C: Rhopalurus laticauda Thorell, 1876, ō (MZSP). D: Jaguajir rochae (Borelli, 1910), (AMNH). E: Rhopalurus laticauda Thorell, 1876, § (AMCC [LP 2845]). F: Troglorhopalurus lacrau (Lourenço \& Pinto-da-Rocha, 1997), $q$ (AMCC [LP 3260]). G: Physoctonus debilis (C.L. Koch, 1840), $q$ (MZSP).
renço 2007), whereas Heteroctenus, the name once applied to the Antillean species of Rhopalurus, remained in synonymy. As defined at the outset of this research, Rhopalurus was distributed in the Greater Antilles (Cuba and Hispaniola), the Guiana Shield of northern South America (Colombia, the Guianas, and Venezuela) and northeastern Brazil, where Physoctonus and Troglorhopalurus also occur (Fig. 6; Fet et al. 2000; Teruel 2006; Lourenço 2008; Prendini et al. 2009; Teruel \& Roncallo 2008; Santiago-Blay 2009), but the monophyly of its disjunct components had not been tested quantitatively, either.

The generic distinction between Rhopalurus and Centruroides was also unclear (Esposito et al. 2017). Centruroides is the most speciose genus of Centruroidinae, comprising 91 described species and three subspecies, distributed from the midwestern United States to northern South America (Colombia, Venezuela, Ecuador, and possibly Peru) and throughout the Caribbean (Fig. 6; Hoffmann 1932; Gantenbein et al. 2001; Sissom \&

Lourenço 1987), Historically, several species had been transferred between Centruroides and Rhopalurus, and the generic definitions revised multiple times (Рососк 1890; Werner 1939; Meise 1934; Mello-Leitão 1945; Lourenço 1979). Centruroides were separated from Rhopalurus by the following combination of characters: pedipalp chela fixed finger trichobothrium $d b$ aligned with or proximal to trichobothrium et, fifth metasomal segment elongated in adult males, and the absence of a pecten-sternite stridulation organ. However, trichobothrial positions and the length of the fifth metasomal segment are interspecifically variable within both genera, and the absence of a stridulation organ is probably plesiomorphic. A phylogenetic analysis of Cuban scorpions based on a single mitochondrial gene locus ( 16 S rDNA) recovered Centruroides paraphyletic with respect to Rhopalurus (Fet et al. 2003a) whereas ovariuterine data suggested Rhopalurus was paraphyletic with respect to Centruroides (Volschenk et al. 2008). A better understanding of the systematic limits and diagnosis of Centruroides is


Fig. 5. Pectinal teeth, dorsal aspect, fine structure, illustrating plectrum of pecten-sternite stridulation organs of species in the New World buthid scorpion subfamily Centruroidinae Kraus, 1955. A: Jaguajir agamemnon (C.L. Koch, 1839), (MZSP). B: Heteroctenus princeps (Karsch, 1879), ð̃ (AMNH). C. Rhopalurus laticauda Thorell, 1876, (MZSP). D: Troglorhopalurus lacrau (Lourenço \& Pinto-da-Rocha, 1997), $q$ (MZSP). E: Ischnotelson guanambiensis (Lenarducci et al., 2005), đ̋ (MZSP). F: Physoctonus debilis (C.L. Koch, 1840), $q$ (MZSP).
of considerable medical importance because the genus includes the only dangerously venomous scorpions in North America, among them eight species responsible for lethal envenomations in humans (Dehesa-Davila \& Possani 1994; Chávez-Haro \& Ortiz 2015).

The taxonomy of species previously assigned to Rhopalurus was in a similar state of disarray when this research began. Mostly large and colorful, and often with the ability to stridulate audibly, these charismatic scorpions have attracted considerable attention. In the past decade alone, several publications proposed taxonomic changes and described new species (Lenarducci et al. 2005; Teruel 2006; Teruel \& Armas 2006, 2012; Lourenço 2007, 2008, 2014; Teruel \& Roncallo 2008, 2013; Teruel \& Tietz 2008; Prendini et al. 2009; Santi-ago-Blay 2009; Flórez 2012), often increasing, rather than decreasing, the taxonomic confusion. For example, Rhopalurus caribensis Teruel \& Roncallo, 2008, Rhopalurus crassicauda Caporiacco, 1947 and Rhopalurus pintoi Mello-Leitão, 1932 were each synonymized and then resurrected. The validity of $R$. crassicauda, its two subspecies, and Rhopalurus virkki Santiago-Blay, 2009 was questioned by several authors (Prendini et al. 2009; Teruel \& Armas 2012).

The study presented here is the first rigorous test of the monophyly of Centruroidinae and its component taxa, based on phylogenetic analysis of 90 morphologi-
cal characters and 4,260 aligned base-pairs of DNA sequence from three mitochondrial and two nuclear DNA loci for 102 terminal taxa, representing 24 species in seven ingroup genera, and nine species in three outgroup genera. The integration of morphological and genomic data provides the benefits of simultaneously testing alternative sources of evidence for the monophyly of Centruroidinae and its component genera, and recognizing diagnostic morphological synapomorphies for their identification. A revised classification of Centruroidinae (Table 2), as Rhopalurusinae, was presented by Esposito et al. (2017). The present study also investigates, for the first time, the evolution of the pecten-sternite stridulation organs of these scorpions, revealing that three distinct types evolved in Heteroctenus, Jaguajir and Rhopalurus.

## 2. Material and methods

### 2.1. Taxon sampling

The classification of Centruroidinae employed here follows Esposito et al. (2017) (Table 2). In order to test the monophyly of the subfamily and its component genera,


Fig. 6. Approximate distributions of genera in the New World buthid scorpion subfamily Centruroidinae Kraus, 1955: A: Centruroides Marx, 1890 and Heteroctenus Pocock, 1893. B: Rhopalurus Thorell, 1876 and Troglorhopalurus Lourenço et al., 2004. C: Ischnotelson Esposito et al., 2017 and Physoctonus Mello-Leitão, 1934. D: Jaguajir Esposito et al., 2017.
as well as the validity of species previously assigned to Physoctonus, Rhopalurus, and Troglorhopalurus, samples were obtained from across the known distributions of as many current and previously recognized species and subspecies as possible, with an emphasis on obtaining material from the proximity of type localities to accurately assign names to populations (Appendix 2). The sample included topotypes of seven infrageneric taxa listed in synonymy by Esposito et al. (2017): Centrurus stenochirus Penther, 1913; Rhopalurus acromelas Lutz \& Mello, 1922; Rhopalurus amazonicus Lourenço, 1986; Rhopalurus laticauda sachsii Karsch, 1879; Rhopalurus piceus Lourenço \& Pinto-da-Rocha, 1997; Rhopalurus virkkii Santiago-Blay, 2009; Rhopalurus aridicola Teruel \& Armas, 2012. Six exemplar species (Prendini 2001b) of Centruroides were selected to represent the taxonomic and geographical diversity of this speciose genus (Esposito 2011).

Six infrageneric taxa, five of which were synonymized by Esposito et al. (2017), were omitted from the ingroup because fresh material was unavailable for DNA extraction. Four of these taxa are restricted to Cuba and could
not be obtained for study: Heteroctenus gibarae (Teruel, 2006) and its junior synonym, Rhopalurus granulimanus Teruel, 2006; Rhopalurus melloleitaoi Teruel \& Armas, 2006, a junior synonym of Heteroctenus junceus (Herbst, 1800). Rhopalurus brejo Lourenço, 2014, known only from the holotype, is a junior synonym of Troglorhopalurus lacrau (Lourenço \& Pinto-da-Rocha, 1997). Rhopalurus crassicauda paruensis Lourenço, 2008 and Rhopalurus pintoi kourouensis Lourenço, 2008 are junior synonyms of Rhopalurus laticauda Thorell, 1876 and Jaguajir pintoi (Mello-Leitão, 1932), respectively.

Exemplar species from two genera of Tityinae were included as outgroups: six species of Tityus, selected to represent four of its five subgenera (Lourenço 2006), and two of the three described species of Zabius. Trees were rooted on the cosmotropical buthid, Isometrus maculatus (DeGeer, 1778). The final taxon sample therefore comprised 24 ingroup species and nine outgroup species, considered satisfactory for testing the monophyly of Centruroidinae and polarizing the morphological characters of its component genera, the aims of this study.

Table 1. Previous classification of the buthid scorpion species assigned to genera Physoctonus Mello-Leitão, 1934, Rhopalurus Thorell, 1876 and Troglorhopalurus Lourenço et al., 2004 with countries of distribution (Prendini et al. 2009; Lourenço 2014).

| Physoctonus debilis (C.L. Koch, 1840) | Brazil |
| :--- | :--- |
| Rhopalurus abudi Armas \& Marcano Fondeur, 1987 | Dominican Republic |
| Rhopalurus acromelas Lutz \& Mello, 1922 | Brazil |
| Rhopalurus agamemnon (C.L. Koch, 1839) | Brazil |
| Rhopalurus amazonicus Lourenço, 1986 | Brazil |
| Rhopalurus aridicola Teruel \& Armas, 2012 | Cuba |
| Rhopalurus bonettii Armas \& Marcano Fondeur, 1987 | Dominican Republic |
| Rhopalurus brejo Lourenço, 2014 | Brazil |
| Rhopalurus caribensis Teruel \& Roncallo, 2008 | Colombia |
| Rhopalurus crassicauda Caporiacco, 1947 | Brazil, Guyana |
| Rhopalurus crassicauda paruensis Lourenço, 2008 | Brazil |
| Rhopalurus garridoi Armas, 1974 | Cuba |
| Rhopalurus gibarae Teruel, 2006 | Cuba |
| Rhopalurus granulimanus Teruel, 2006 | Cuba |
| Rhopalurus guanambiensis Lenarducci et al., 2005 | Brazil |
| Rhopalurus junceus (Herbst, 1800) | Cuba |
| Rhopalurus lacrau Lourenço \& Pinto-da-Rocha 1997 | Brazil |
| Rhopalurus laticauda Thorell, 1876 | Colombia, Venezuela |
| Rhopalurus melloleitaoi Teruel \& Armas, 2006 | Cuba |
| Rhopalurus pintoi Mello-Leitão, 1932 | Brazil, Guyana |
| Rhopalurus pintoi kourouensis Lourenço, 2008 | French Guiana |
| Rhopalurus princeps (Karsch, 1879) | Dominican Republic, Haiti |
| Rhopalurus rochae Borelli, 1910 | Brazil |
| Rhopalurus virkkii Santiago-Blay, 2009 | USA (Mona Is., Puerto Rico) |
| Troglorhopalurus translucidus Lourenço et al., 2004 | Brazil |

Whereas DNA sequences were generated for multiple conspecific individuals of thirteen ingroup species, resulting in a final sample of 102 terminal taxa (Appendix 2), morphological characters were scored for only one terminal taxon per species (i.e., 33 terminal taxa) and extrapolated to all conspecific individuals in the simultaneous analyses with molecular data, because none of the morphological characters were assessed to be intraspecifically polymorphic (Prendini 2001b).

### 2.2. Morphological data

Twenty-seven morphological characters were adopted from published matrices on various scorpion taxa (Lamoral 1978, 1980; Jeram 1994, 1998; Prendini 2000, 2001a, 2004; Soleglad \& Sissom 2001; Soleglad \& Fet 2001, 2003; Volschenk et al. 2008), 33 from unpublished matrices (Sтоскwell 1989; E.S. Volschenk \& L. Prendini unpublished data), and 30 new characters were added for a total of 90 characters, comprising $38(43 \%)$ characters from the prosoma, 25 (33\%) from the mesosoma, and 21 ( $23 \%$ ) from the metasoma. $54 \%$ of the characters were derived from carination and surface macrosculpture, $21 \%$ from shape and morphometrics, $13 \%$ from macrosetae and trichobothria, $9 \%$ from internal and external anatomy, and $3 \%$ from coloration (Appendix 3).

Nomenclature follows Hjelle (1990) and Sissom (1990), except for carapace, tergite and metasomal

Table 2. Revised classification (Esposito et al. 2017) of the buthid scorpion species previously assigned to genera Physoctonus MelloLeitão, 1934, Rhopalurus Thorell, 1876 and Troglorhopalurus Lourenço et al., 2004 with countries of distribution.

| Heteroctenus abudi (Armas \& Marcano Fondeur, 1987) <br> (= Rhopalurus virkkii Santiago-Blay, 2009) | Dominican Republic, <br> USA (Mona Is., <br> Puerto Rico) |
| :--- | :--- |
| Heteroctenus bonettii (Armas \& Marcano Fondeur, 1987) | Dominican Republic |
| Heteroctenus garridoi (Armas, 1974) | Cuba |
| Heteroctenus gibarae (Teruel, 2006) <br> (= Rhopalurus granulimanus Teruel, 2006) | Cuba |
| Heteroctenus junceus (Herbst, 1800) <br> (= Rhopalurus melloleeitaoi Teruel \& Armas, 2006, <br> Rhopalurus aridicola Teruel \& Armas, 2012) | Cuba |
| Heteroctenus princeps (Karsch, 1879) | Dominican Republic, <br> Haiti |
| Ischnotelson guanambiensis (Lenarducci et al., 2005) | Brazil |
| Ischnotelson peruassu Esposito et al., 2017 | Brazil |
| Jaguajir agamemnon (C.L. Koch, 1839) <br> (= Rhopalurus acromelas Lutz \& Mello, 1922) | Brazil |
| Jaguajir pintoi (Mello-Leitão, 1932) <br> (= Rhopalurus pintoi kourouensis Lourenço, 2008) | Brazil, French Guiana, <br> Guyana |
| Jaguajir rochae (Borelli, 1910) | Brazil |
| Physoctonus debilis (C.L. Koch, 1840) | Brazil |
| Physoctonus striatus Esposito et al., 2017 | Brazil |
| Rhopalurus caribensis Teruel \& Roncallo, 2008 | Colombia |
| Rhopalurus laticauda Thorell, 1876 <br> (= Rhopalurus crassicauda Caporiacco, 1947, <br> Rhopalurus amazonicus Lourenço, 1986, <br> Rhopalurus crassicauda paruensis Lourenço, 2008) | Brazil, Colombia, <br> Venezuela |
| Rhopalurus ochoai Esposito et al., 2017 | Venezuela |
| Troglorhopalurus lacrau (Lourenço \& Pinto-da-Rocha, 1997) <br> (= Rhopalurus brejo Lourenço, 2014) | Brazil |
| Troglorhopalurus trans/ucidus Lourenço et al., 2004 | Brazil |

carination (VAChON 1952), pedipalp carination (PRENdini 2001a), pedipalp trichobothria (Vachon 1974), ovariuterine anatomy (Volschenk et al. 2008), and book lung ultrastructure (Kamenz \& Prendini 2008). Measurements follow Stahnke (1970), Lamoral (1979), and Prendini (2001a). Morphological examination of specimens (Appendix 3) was conducted using a Nikon SMZ1500 dissection stereomicroscope. Specimens were measured using Mitutoyo digital calipers and an ocular micrometer. The morphological matrix (Table 3) was assembled and scored in Mesquite v2.74 (Maddison \& Maddison 2010).

### 2.3. Molecular data

Field-collected specimens were injected with and preserved in $95 \%$ ethanol, and stored at $-20^{\circ} \mathrm{C}$. Genomic DNA was extracted from muscle tissue dissected from the fourth leg using a Qiagen DNEasy Blood and Tissue extraction kit according the manufacturers protocols.

Extracted DNA was amplified for five gene loci, selected based on their ability to provide resolution at various taxonomic levels (Arnedo et al. 2002; Giribet et al. 2001; Harrison et al. 1987; Hayashi 1996; Hillis \& Dixon 1991; Wahlberg \& Zimmermann 2000), in overlap-
Table 3. Distribution of 90 morphological characters (Appendix 2) among ingroup and outgroup taxa for phylogenetic analysis of the New World buthid subfamily Centruroidinae Kraus, 1955. Character states scored $0-5$ or inapplicable (-). Asterisk denotes material from populations referable to Rhopalurus amazonicus Lourenço, 1986 [=Rhopalurus laticauda Thorell, 1876]. Material examined is listed in Appendix 1.

ping fragments using universal eukaryote and scorpion specific primers (Table 4): a mitochondrial protein-coding gene, Cytochrome $c$ Oxidase I (COI); mitochondrial structural genes, 12 S rDNA (12S) and 16S rDNA (16S); and nuclear structural genes, 18 S rDNA (18S), 28 S rDNA (28S). The Polymerase Chain Reaction was performed on an Epicenter thermocycler (Eppendorf) using

GoTaq polymerase (Promega). Reactions were verified on a $1.2 \%$ agarose gel stained with Sybr safe DNA gel stain (Invitrogen), and subsequently purified using the Ampure DNA (Agencourt) purification system on a Biomek NX robot (Beckman-Coulter).

Cycle sequencing was conducted using Big Dye v1.1 and automated Sanger sequencing of single-stranded

Table 4. Primers used to amplify DNA sequences of two nuclear and three mitochondrial gene markers for phylogenetic analysis of the New World buthid subfamily Centruroidinae Kraus, 1955.

| Primer Name | Primer Sequence (5' to $3^{\prime}$ ) | Citation |
| :---: | :---: | :---: |
| 18S rDNA |  |  |
| 18Sa2.0 | ATGGTtGCAAAGCTGAAAC | Wheeler et al. (1993) |
| 18Sbi | GAGTCTCGTtCGTtATCGGA | Wheeler et al. (1993) |
| 18S1F | tacctgatthatcctgccagtag | Giribet et al. (1996) |
| 18S3F | GTTCGATTCCGGAGAGGGA | GIribet et al. (1996) |
| 18S5R | CTTGGCAAATGCTtTCGC | GIribet et al. (1996) |
| 18S9R | GAtCCttccacagattcacctac | Giribet et al. (1996) |
| 28 S rDNA |  |  |
| 28Sa | GACCCGTCTTGAAGCACG | Nunn et al. (1996) |
| 28Sb | TCGGAAGGAACCAGCTAC | Nunn et al. (1996) |
| 28Sbout | CCCACAGCGCCAGTTCTGCTTACC | Prendini et al. (2005) |
| 12 S rDNA |  |  |
| 12Sai | AAACTAGGATTAGATACCCTATtAT | Kocher et al. (1989) |
| 12 Sbi | AAGAGCGACGGGCGATGTGT | Kocher et al. (1989) |
| 16S rDNA |  |  |
| 16Sbr | CTCCGGTTTGAACTCAGATCA | Simon et al. (1994) |
| 16Sar | CGCCTGTtTATCAAAAACAT | Simon et al. (1994) |
| Cytochrome c Oxidase I |  |  |
| HCO | TAAACTTCAGGGTGACCAAAAAATCA | Folmer et al. (1994) |
| HCOoutout | GTAAATATATGRTGDGCTC | Prendini et al. (2005) |
| LCO | GGTCAACAAATCATAAAGATATTGG | Folmer et al. (1994) |
| Nancy (C1-N-2191) | CCCGGTAAAATTAAAATATAAACTTC | Harrison et al. (1987) |
| C1-J-1718 | GGNGGATTTGGAAATTGRTTRGTTCC | Harrison et al. (1987) |
| C1-N-2776 | GGATAATCAGAATANCGNCGAGG | Harrison et al. (1987) |
| CruzR | CATACCCAAAGARCCAAAAGG | Valdez-Cruz et al. (2004) |
| LE1R | TCCATTCCCACAGTAAACATATG | Esposito (2011) |
| HCOEXTa | GAAGTtTATATtTtAATTTTACCTGG | Simon et al. (1994) |
| HCOEXTb | CCTATTGAWARAACATARTGAAAATG | Simon et al. (1994) |

DNA performed on an Applied Biosystems Inc. Prism ${ }^{\text {TM }}$ $3730 \times$. Paired-strand reads were aligned using Sequencher ${ }^{\mathrm{TM}}$ and edited by hand. A total 506 DNA sequences were generated (Table 5). The sequences of 98 individuals were complete for all 5 gene loci.

### 2.4. Phylogenetic analysis

Morphological characters were equally weighted a priori and analyzed with parsimony in TNT v1.1 (Goloboff et al. 2003) using the New Technology Search option with 10,000 random addition replicates. Implied character weighting (Goloboff 1993) was employed to evaluate the robustness of the topology to character weighting. Six values for the concavity constant, $k$, were investigated. Bootstrap measures of node support were calculated in TNT.

The leaf stability index (Thorley \& Page 2000) was calculated from the resulting trees in Phyutility v2.2 (Smith \& Dunn 2008) to identify rogue taxa (SanderSON \& Schaffer 2002) which may have an impact on the topology (Thompson \& Schaffer 2010) or measures of support (Pattengale et al. 2010).

Multiple sequence alignments for individual gene partitions were performed in MAFFT (КАтон et al. 2005) using the G-INS-i strategy, recommended by the authors for less than 200 sequences with global homology, and
the PAM1/K $=2$ matrix parameter, recommended by the authors for aligning sequences of closely related taxa. The resulting alignments were manually edited in Geneious v5.1 (Biomatters, Ltd.).

Aligned sequence data from the five gene loci (18S, $28 \mathrm{~S}, 12 \mathrm{~S}, 16 \mathrm{~S}$, COI) were concatenated to produce a molecular data matrix comprising 87 terminals and 4,250 characters, 3,104 of which were invariant, 167 variable but uninformative, and 979 variable and informative. The nucleotide composition was $25 \%$ A, $18.5 \%$ C, $25 \%$ G and $31.5 \% \mathrm{~T}$.

The concatenated DNA sequence alignments were analyzed simultaneously in TNT v1.1, using equal weights in the New Technology Search option comprising 10,000 random addition replicates of Tree Drift (Goloboff 1999) and Ratchet (Nixon 1999) to identify the most parsimonious tree. The concatenated dataset, partitioned by gene and codon position, were analyzed under Bayesian (MrBayes v3.2.1) (Huelsenbeck \& Ronquist 2001; Ronquist \& Huelsenbeck 2003) and likelihood (RaxML) (Stamatakis 2006) criteria. MrModeltest2 v2.3 (Nylander 2004), employing Akaike Information Criteria (Aкаiкe 1973), was used to determine the best fitting model of DNA substitution for each gene and codon position (COI) (Table 6). When the best fitting model included both the $\Gamma$ and invariable sites (I) parameters, the next best fitting model was selected to avoid issues resulting from non-

Table 5. Genbank accession codes for vouchers, deposited at the American Museum of Natural History, New York (AMNH), and the Museu de Zoologia da Universidade São Paulo, Brazil (MZSP), and tissue samples, deposited in the Ambrose Monell Collection for Molecular and Microbial Research (AMCC) at the AMNH, from which DNA was extracted and sequenced for phylogenetic analysis of the New World buthid subfamily Centruroidinae Kraus, 1955. Type localities (TL) of taxa currently recognized or in synonymy [ ] denoted by 'Y'. Provenance data are provided in Appendix 1.

| Species | TL | Voucher | AMCC | 18S | 28 S | 12S | 16S | COI |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| Isometrus maculatus |  | AMNH | LP 1798 | KY982016.1 | KY982111.1 | KY981825.1 | KY981921.1 | KY982207.1 |
| Tityus atriventer |  | AMNH | LP 9033 | KY982074.1 | KY982169.1 | KY981883.1 | KY981978.1 | KY982264.1 |
| Tityus bahiensis |  | AMNH | LP 5641 | KY982075.1 | KY982170.1 | KY981884.1 | KY981979.1 | KY982265.1 |
| Tityus clathratus |  | AMNH | LP 1567 | KY982076.1 | KY982171.1 | KY981885.1 | KY981980.1 | KY982266.1 |
| Tityus discrepans |  | AMNH | LP 1547 | KY982077.1 | KY982172.1 | KY981886.1 | KY981981.1 | KY982267.1 |
| Tityus kuryi |  | AMNH | LP 7659 | KY982078.1 | KY982173.1 | KY981887.1 | KY981982.1 | KY982268.1 |
| Tityus metuendus |  | AMNH | LP 1546 | KY982079.1 | KY982174.1 | KY981888.1 | KY981983.1 | KY982269.1 |
| Tityus smithii |  | AMNH | LP 9046 | KY982081.1 | KY982176.1 | KY981890.1 | KY981985.1 | KY982271.1 |
| Tityus riverai |  | AMNH | LP 10202 | KY982080.1 | KY982175.1 | KY981889.1 | KY981984.1 | KY982270.1 |
| Zabius birabeni |  | AMNH | LP 4251 | KY982082.1 | KY982177.1 | KY981891.1 | KY981986.1 | KY982272.1 |
| Zabius fuscus |  | AMNH | LP 5642 | KY982083.1 | KY982178.1 | KY981892.1 | KY981987.1 | KY982273.1 |
| Centruroides exilicauda |  | AMNH | LP 1692 | KY981988.1 | KY982084.1 | KY981797.1 | KY981893.1 | KY982179.1 |
| Centruroides gracilis |  | AMNH | LP 2013 | KY981989.1 | KY982085.1 | KY981798.1 | KY981894.1 | KY982180.1 |
| Centruroides infamatus |  | AMNH | LP 1822 | KY981990.1 | KY982086.1 | KY981799.1 | KY981895.1 | KY982181.1 |
| Centruroides margaritatus |  | AMNH | LP 1986 | KY981991.1 | KY982087.1 | KY981800.1 | KY981896.1 | KY982182.1 |
| Centruroides rileyi |  | AMNH | LP 6445 | KY981992.1 | KY982088.1 | KY981801.1 | KY981897.1 | KY982183.1 |
| Centruroides schmidti |  | AMNH | LP 9172 | KY981993.1 | KY982089.1 | KY981802.1 | KY981898.1 | KY982184.1 |
| Centruroides vittatus |  | AMNH | LP 2286 | KY981994.1 | KY982090.1 | KY981803.1 | KY981899.1 | KY982185.1 |
| Heteroctenus abudi |  | AMNH | LP 3268 | KY981997.1 | KY982093.1 | KY981806.1 | KY981902.1 | KY982188.1 |
| [= Rhopalurus virkkii] | [Y] | AMNH | LP 10234 | KY981995.1 | KY982091.1 | KY981804.1 | KY981900.1 | KY982186.1 |
|  | [Y] | AMNH | LP 10235 | KY981996.1 | KY982092.1 | KY981805.1 | KY981901.1 | KY982187.1 |
| Heteroctenus bonettii | Y | AMNH | LP 2471 | KY981998.1 | KY982094.1 | KY981807.1 | KY981903.1 | KY982189.1 |
|  | Y | AMNH | LP 3267 | KY981999.1 | KY982095.1 | KY981808.1 | KY981904.1 | KY982190.1 |
| Heteroctenus garridoi | Y | AMNH | LP 10225 | KY982000.1 | KY982096.1 | KY981809.1 | KY981905.1 | KY982191.1 |
| Heteroctenus junceus |  | AMNH | LP 1517 | KY982007.1 | KY982103.1 | KY981816.1 | KY981912.1 | KY982198.1 |
|  |  | AMNH | LP 1565 | KY982008.1 | KY982104.1 | KY981817.1 | KY981913.1 | KY982199.1 |
| [= Rhopalurus aridicola] | [Y] | AMNH | LP 12613 | KY982001.1 | KY982097.1 | KY981810.1 | KY981906.1 | KY982192.1 |
|  | [Y] | AMNH | LP 12618 | KY982002.1 | KY982098.1 | KY981811.1 | KY981907.1 | KY982193.1 |
| Heteroctenus princeps |  | AMNH | LP 12622 | KY982003.1 | KY982099.1 | KY981812.1 | KY981908.1 | KY982194.1 |
|  |  | AMNH | LP 12624 | KY982004.1 | KY982100.1 | KY981813.1 | KY981909.1 | KY982195.1 |
|  |  | AMNH | LP 12627 | KY982005.1 | KY982101.1 | KY981814.1 | KY981910.1 | KY982196.1 |
|  |  | AMNH | LP 1516 | KY982010.1 | KY982106.1 | KY981819.1 | KY981915.1 | KY982201.1 |
|  |  | AMNH | LP 1566 | KY982011.1 | KY982107.1 | KY981820.1 | KY981916.1 | KY982202.1 |
|  |  | AMNH | LP 3262 | KY982012.1 | KY982108.1 | KY981821.1 | KY981917.1 | KY982203.1 |
|  |  | AMNH | LP 3264 | KY982013.1 | - | KY981822.1 | KY981918.1 | KY982204.1 |
|  |  | AMNH | LP 12478 | KY982009.1 | KY982105.1 | KY981818.1 | KY981914.1 | KY982200.1 |
| Ischnotelson guanambiensis | Y | MZSP 30864 | LP 9669 | KY982014.1 | KY982109.1 | KY981823.1 | KY981919.1 | KY982205.1 |
|  |  | MZSP 30865 | LP 9670 | KY982015.1 | KY982110.1 | KY981824.1 | KY981920.1 | KY982206.1 |
| Ischnotelson peruassu | Y | MZSP 31138 | LP 9937 | KY982017.1 | KY982112.1 | KY981826.1 | KY981922.1 | KY982208.1 |
| Jaguajir agamemnon |  | MZSP 30883 | LP 9692 | KY982018.1 | KY982113.1 | KY981827.1 | KY981923.1 | KY982209.1 |
|  |  | MZSP 30884 | LP 9693 | KY982019.1 | KY982114.1 | KY981828.1 | KY981924.1 | KY982210.1 |
|  |  | MZSP 30885 | LP 9694 | KY982020.1 | KY982115.1 | KY981829.1 | KY981925.1 | KY982211.1 |
|  |  | MZSP 30886 | LP 9695 | KY982021.1 | KY982116.1 | KY981830.1 | KY981926.1 | KY982212.1 |
|  |  | MZSP 30887 | LP 9696 | KY982022.1 | KY982117.1 | KY981831.1 | KY981927.1 | KY982213.1 |
| [ = Rhopalurus acromelas] | [Y] | MZSP 31170 | LP 9929 | KY982023.1 | KY982118.1 | KY981832.1 | KY981928.1 | KY982214.1 |
|  |  | MZSP 31133 | LP 9932 | KY982024.1 | KY982119.1 | KY981833.1 | KY981929.1 | KY982215.1 |
|  |  | MZSP 31157 | LP 9933 | KY982025.1 | KY982120.1 | KY981834.1 | KY981930.1 | KY982216.1 |
|  |  | MZSP 31161 | LP 9942 | KY982026.1 | KY982121.1 | KY981835.1 | KY981931.1 | KY982217.1 |
|  |  | MZSP 31167 | LP 9949 | KY982027.1 | KY982122.1 | KY981836.1 | KY981932.1 | KY982218.1 |
|  |  | MZSP 31181 | LP 9958 | KY982028.1 | KY982123.1 | KY981837.1 | KY981933.1 | KY982219.1 |
| Jaguajir pintoi |  |  |  |  |  |  |  |  |
| [= Rhopa/urus crassicauda] | [Y] | AMNH | LP 8278 | KY982029.1 | KY982124.1 | KY981838.1 | KY981934.1 | KY982220.1 |
| [= Rhopalurus piceus] | [Y] | MZSP 30863 | LP 9671 | KY982030.1 | KY982125.1 | KY981839.1 | KY981935.1 | KY982221.1 |
|  | [Y] | MZSP 30862 | LP 9672 | KY982031.1 | KY982126.1 | KY981840.1 | KY981936.1 | KY982222.1 |
|  |  | MZSP 31176 | LP 9928 | KY982032.1 | KY982127.1 | KY981841.1 | KY981937. | KY982223.1 |

Table 5 continued.

| Species | TL | Voucher | AMCC | 18S | 28S | 12S | 16S | COI |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| Jaguajir rochae |  | AMNH | LP 1775 | KY982033.1 | KY982128.1 | KY981842.1 | KY981938.1 | KY982224.1 |
|  |  | AMNH | LP 7638 | KY982034.1 | KY982129.1 | KY981843.1 | KY981939.1 | KY982225.1 |
|  |  | AMNH | LP 7639 | KY982035.1 | KY982130.1 | KY981844.1 | KY981940.1 | - |
|  |  | MZSP 30879 | LP 9682 | KY982036.1 | KY982131.1 | KY981845.1 | KY981941.1 | KY982226.1 |
|  |  | MZSP 30880 | LP 9683 | KY982037.1 | KY982132.1 | KY981846.1 | KY981942.1 | KY982227.1 |
|  |  | MZSP 30881 | LP 9684 | KY982038.1 | KY982133.1 | KY981847.1 | KY981943.1 | KY982228.1 |
|  |  | MZSP 30882 | LP 9685 | KY982039.1 | KY982134.1 | KY981848.1 | KY981944.1 | KY982229.1 |
|  |  | MZSP 31127 | LP 9926 | KY982040.1 | KY982135.1 | KY981849.1 | KY981945.1 | KY982230.1 |
|  |  | MZSP 31151 | LP 9941 | KY982041.1 | KY982136.1 | KY981850.1 | KY981946.1 | KY982231.1 |
|  |  | MZSP 31146 | LP 9943 | KY982042.1 | KY982137.1 | KY981851.1 | KY981947.1 | KY982232.1 |
|  |  | MZSP 31148 | LP 9946 | KY982043.1 | KY982138.1 | KY981852.1 | KY981948.1 | KY982233.1 |
|  |  | MZSP 31124 | LP 9947 | KY982044.1 | KY982139.1 | KY981853.1 | KY981949.1 | KY982234.1 |
|  |  | MZSP 31143 | LP 9951 | KY982045.1 | KY982140.1 | KY981854.1 | KY981950.1 | KY982235.1 |
|  |  | MZSP 31123 | LP 9953 | KY982046.1 | KY982141.1 | KY981855.1 | KY981951.1 | KY982236.1 |
| [ = Centrurus stenochirus] | [Y] | MZSP 31122 | LP 9963 | KY982047.1 | KY982142.1 | KY981856.1 | - | KY982237.1 |
| Physoctonus debilis |  | MZSP 30866 | LP 9678 | KY982048.1 | KY982143.1 | KY981857.1 | KY981952.1 | KY982238.1 |
|  |  | MZSP 30867 | LP 9679 | KY982049.1 | KY982144.1 | KY981858.1 | KY981953.1 | KY982239.1 |
|  |  | MZSP 30868 | LP 9680 | KY982050.1 | KY982145.1 | KY981859.1 | KY981954.1 | KY982240.1 |
| Physoctonus striatus | Y | MZSP 30869 | LP 9681 | KY982051.1 | KY982146.1 | KY981860.1 | KY981955.1 | KY982241.1 |
| Rhopalurus caribensis | Y | AMNH | LP 9341 | KY982053.1 | KY982148.1 | KY981862.1 | KY981957.1 | KY982243.1 |
|  |  | AMNH | LP 13167 | KY982052.1 | KY982147.1 | KY981861.1 | KY981956.1 | KY982242.1 |
| Rhopalurus laticauda |  | AMNH | LP 2462 | KY982057.1 | KY982152.1 | KY981866.1 | KY981961.1 | KY9822471. |
| [= Rhopalurus laticauda sachsii] | [Y] | AMNH | LP 2845 | KY982058.1 | KY982153.1 | KY981867.1 | KY981962.1 | KY982248.1 |
|  |  | AMNH | LP 4221 | KY982059.1 | KY982154.1 | KY981868.1 | KY981963.1 | KY982249.1 |
|  |  | AMNH | LP 9200 | KY982060.1 | KY982155.1 | KY981869.1 | KY981964.1 | KY982250.1 |
|  |  | AMNH | LP 9237 | KY982061.1 | KY982156.1 | KY981870.1 | KY981965.1 | KY982251.1 |
|  |  | AMNH | LP 9253 | KY982062.1 | KY982157.1 | KY981871.1 | KY981966.1 | KY982252.1 |
|  |  | AMNH | LP 9256 | KY982063.1 | KY982158.1 | KY981872.1 | KY981967.1 | KY982253.1 |
|  |  | MZSP 30876 | LP 9675 | KY982064.1 | KY982159.1 | KY981873.1 | KY981968.1 | KY982254.1 |
|  |  | MZSP 30877 | LP 9676 | KY982065.1 | KY982160.1 | KY981874.1 | KY981969.1 | KY982255.1 |
|  |  | MZSP 30878 | LP 9677 | KY982066.1 | KY982161.1 | KY981875.1 | KY981970.1 | KY982256.1 |
| [= Rhopalurus amazonicus] | [Y] | MZSP 30870 | LP 9686 | KY982067.1 | KY982162.1 | KY981876.1 | KY981971.1 | KY982257.1 |
|  |  | MZSP 30871 | LP 9687 | KY982068.1 | KY982163.1 | KY981877.1 | KY981972.1 | KY982258.1 |
|  | [Y] | MZSP 30872 | LP 9688 | KY982069.1 | KY982164.1 | KY981878.1 | KY981973.1 | KY982259.1 |
|  | [Y] | MZSP 30873 | LP 9689 | KY982070.1 | KY982165.1 | KY981879.1 | KY981974.1 | KY982260.1 |
|  | [Y] | MZSP 30874 | LP 9690 | KY982071.1 | KY982166.1 | KY981880.1 | KY981975.1 | KY982261.1 |
|  | [Y] | MZSP 30875 | LP 9691 | KY982072.1 | KY982167.1 | KY981881.1 | KY981976.1 | KY982262.1 |
|  |  | MZSP 31172 | LP 9927 | KY982073.1 | KY982168.1 | KY981882.1 | KY981977.1 | KY982263.1 |
|  |  | AMNH | LP 10046 | KY982054.1 | KY982149.1 | KY981863.1 | KY981958.1 | KY982244.1 |
|  |  | AMNH | LP 10047 | KY982055.1 | KY982150.1 | KY981864.1 | KY981959.1 | KY982245.1 |
|  |  | AMNH | LP 10048 | KY982056.1 | KY982151.1 | KY981865.1 | KY981960.1 | KY982246.1 |
| Rhopalurus ochoai | Y | AMNH | LP 5504 | MF508621.1 | MF508628.1 | - | MF402014.1 | MF508635.1 |
|  |  | AMNH | LP 5505 | MF508622.1 | MF508629.1 | - | MF402015.1 | MF508636.1 |
|  |  | AMNH | LP 9199 | MF508623.1 | MF508630.1 | - | MF402016.1 | MF508637.1 |
|  |  | AMNH | LP 9207 | MF508624.1 | MF508631.1 | - | MF402017.1 | MF508638.1 |
| Troglorhopalurus lacrau |  | AMNH | LP 7637 | MF508626.1 | MF508633.1 | MF508616.1 | MF508619.1 | MF508640.1 |
|  | Y | AMNH | LP 10211 | MF508625.1 | MF508632.1 | MF508615.1 | MF508618.1 | MF508639.1 |
| Troglorhopalurus translucidus | Y | MZSP 30888 | LP 9668 | MF508627.1 | MF508634.4 | MF508617.1 | MF508620.1 | MF508641.1 |

independence of the $\Gamma$ and I parameters. The Bayesian analysis was performed on the CiPRES supercomputing cluster (Miller et al. 2009) in four independent runs for 60 million generations, sampling every 1000 generations. Burn-in times were determined by eye using $\ln$-likelihood in Tracer v1.5 (Rambaut \& Drummond 2007) and convergence assessed by the standard deviations of split frequencies in AWTY (Nylander et al. 2008). A maximum
clade credibility tree was computed from the post-burn-in trees with TreeAnnotator v1.6.1 (Rambaut \& Drummond 2007). The concatenated dataset was also analyzed in raxmIHPC v7.0.4 (Stamatakis 2006). Each partition was analyzed under the GTR $+\Gamma$ model (Yang 1994).

The morphological character matrix and concatenated DNA sequence alignments were analyzed simultaneously with parsimony and Bayesian Inference. Par-
simony analysis was conducted using TNT with equal weighting. The Bayesian analysis was performed in MrBayes v.3.2.1 on the CiPRES supercomputing cluster in two independent runs for 50 million generations, sampling every 1000 generations. Burn-in times were determined by eye using $\ln$-likelihood in Tracer v1.5 (Rambaut \& Drummond 2007) and convergence assessed by the standard deviations of split frequencies in AWTY (Nylander et al. 2008). A maximum clade credibility tree was computed from the post-burn-in trees in TreeAnnotator v1.6.1 (Rambaut \& Drummond 2007).

### 2.5. Stridulation

The pecten-sternite stridulation organ was examined and standardized images prepared of the pectines and sternite III of seventeen species of Centruroidinae, using material in the collections of the University of São Paulo (USP) and the American Museum of Natural History: Heteroctenus abudi (Armas \& Marcano Fondeur, 1987); Heteroctenus bonettii (Armas, 1999); Heteroctenus garridoi (Armas, 1974); H. junceus; Heteroctenus princeps (Karsch, 1879); Ischnotelson guanambiensis

Table 6. Best-fitting models of nucleotide substitution calculated for five loci in phylogenetic analysis of the New World buthid subfamily Centruroidinae Kraus, 1955. The best model was determined for each codon position for Cytochrome $c$ Oxidase subunit I (COI). Partitions indicated with an asterisk are the second-best model following the exclusion of models that included both the $\Gamma$ and I parameters.

| Locus: Partition | AIC Model |
| :--- | :--- |
| 18 S rDNA | $\mathrm{GTR}+\mathrm{I}$ |
| 28 S rDNA | $\mathrm{GTR}+\mathrm{I}$ |
| 12 S rDNA | $\mathrm{GTR}+\mathrm{G}^{*}$ |
| 16 S rDNA | $\mathrm{HKY}+\mathrm{G}^{*}$ |
| COI: $1^{\text {st }}$ position | $\mathrm{GTR}+\mathrm{G}^{*}$ |
| COI: $2^{\text {nd }}$ position | $\mathrm{GTR}+\mathrm{G}^{*}$ |
| COI: $3^{\text {rd }}$ position | $\mathrm{GTR}+\mathrm{G}^{*}$ |

(Lenarducci et al., 2005); Ischnotelson peruassu Esposito et al., 2017; Jaguajir agamemnon (C.L. Koch, 1839); J. pintoi; Jaguajir rochae (Borelli, 1910); R. caribensis; R. laticauda; Rhopalurus ochoai Esposito et al., 2017; T. lacrau; Troglorhopalurus translucidus Lourenço et al., 2004; Physoctonus debilis (C.L. Koch, 1840); Physoctonus striatus Esposito et al., 2017.

Table 7. Length (steps), consistency indices (CI) and retention indices (RI) of 90 morphological characters on the most parsimonious tree obtained by equal weights and implied weighting $(k=3-6)$ for phylogenetic analysis of the New World buthid subfamily Centruroidinae Kraus, 1955.

| Char. | Steps | Cl | RI | Char. | Steps | CI | RI | Char. | Steps | CI | RI |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 1 | 2 | 0.50 | 0.50 | 31 | 6 | 0.50 | 0.82 | 61 | 6 | 0.17 | 0.44 |
| 2 | 4 | 0.25 | 0.70 | 32 | 4 | 0.50 | 0.85 | 62 | 5 | 0.40 | 0.25 |
| 3 | 1 | 1.00 | 1.00 | 33 | 5 | 0.40 | 0.82 | 63 | 10 | 0.10 | 0.31 |
| 4 | 3 | 0.33 | 0.50 | 34 | 1 | 1.00 | 1.00 | 64 | 4 | 0.25 | 0.77 |
| 5 | 2 | 0.50 | 0.67 | 35 | 5 | 0.20 | 0.67 | 65 | 6 | 0.33 | 0.60 |
| 6 | 3 | 0.33 | 0.71 | 36 | 7 | 0.29 | 0.17 | 66 | 6 | 0.33 | 0.67 |
| 7 | 1 | 1.00 | 1.00 | 37 | 6 | 0.17 | 0.67 | 67 | 7 | 0.14 | 0.14 |
| 8 | 6 | 0.17 | 0.00 | 38 | 7 | 0.29 | 0.17 | 68 | 1 | 1.00 | 0.00 |
| 9 | 1 | 1.00 | 1.00 | 39 | 3 | 0.33 | 0.67 | 69 | 7 | 0.14 | 0.25 |
| 10 | 1 | 1.00 | 1.00 | 40 | 3 | 0.67 | 0.92 | 70 | 3 | 0.33 | 0.33 |
| 11 | 1 | 1.00 | 1.00 | 41 | 5 | 0.40 | 0.63 | 71 | 3 | 0.33 | 0.50 |
| 12 | 3 | 0.33 | 0.33 | 42 | 5 | 0.20 | 0.60 | 72 | 2 | 0.50 | 0.50 |
| 13 | 4 | 0.25 | 0.40 | 43 | 3 | 0.67 | 0.83 | 73 | 1 | 1.00 | 0.00 |
| 14 | 4 | 0.25 | 0.40 | 44 | 11 | 0.45 | 0.45 | 74 | 3 | 0.33 | 0.33 |
| 15 | 8 | 0.13 | 0.53 | 45 | 2 | 0.50 | 0.50 | 75 | 7 | 0.29 | 0.44 |
| 16 | 6 | 0.17 | 0.58 | 46 | 3 | 0.33 | 0.50 | 76 | 7 | 0.29 | 0.74 |
| 17 | 2 | 0.50 | 0.00 | 47 | 7 | 0.14 | 0.40 | 77 | 2 | 0.50 | 0.83 |
| 18 | 2 | 0.50 | 0.50 | 48 | 9 | 0.22 | 0.53 | 78 | 6 | 0.17 | 0.50 |
| 19 | 8 | 0.25 | 0.50 | 49 | 7 | 0.29 | 0.69 | 79 | 7 | 0.29 | 0.62 |
| 20 | 3 | 0.67 | 0.90 | 50 | 3 | 0.67 | 0.92 | 80 | 5 | 0.40 | 0.77 |
| 21 | 9 | 0.33 | 0.65 | 51 | 2 | 1.00 | 1.00 | 81 | 4 | 0.25 | 0.40 |
| 22 | 5 | 0.20 | 0.50 | 52 | 1 | 1.00 | 1.00 | 82 | 9 | 0.11 | 0.47 |
| 23 | 5 | 0.20 | 0.00 | 53 | 6 | 0.17 | 0.55 | 83 | 9 | 0.22 | 0.46 |
| 24 | 7 | 0.29 | 0.69 | 54 | 4 | 0.25 | 0.67 | 84 | 2 | 0.50 | 0.75 |
| 25 | 7 | 0.29 | 0.69 | 55 | 4 | 0.25 | 0.50 | 85 | 6 | 0.17 | 0.29 |
| 26 | 5 | 0.20 | 0.73 | 56 | 2 | 0.50 | 0.00 | 86 | 2 | 1.00 | 1.00 |
| 27 | 1 | 1.00 | 1.00 | 57 | 4 | 0.25 | 0.25 | 87 | 2 | 0.50 | 0.50 |
| 28 | 4 | 0.25 | 0.57 | 58 | 2 | 0.50 | 0.00 | 88 | 1 | 1.00 | 1.00 |
| 29 | 1 | 1.00 | 1.00 | 59 | 1 | 1.00 | 0.00 | 89 | 1 | 1.00 | 0.00 |
| 30 | 1 | 1.00 | 1.00 | 60 | 5 | 0.20 | 0.43 | 90 | 4 | 0.25 | 0.00 |



Fig. 7. Phylogeny of the New World buthid scorpion subfamily Centruroidinae Kraus, 1955 obtained by separate analysis of 90 morphological characters with parsimony. Strict consensus of four most parsimonious trees (MPTs) obtained by analysis under equal weighting. Synapomorphies optimized with accelerated transformation. Black circles indicate uniquely derived apomorphic states, white circles indicate parallel derivations of apomorphic states, numbers above indicate characters, and numbers below indicate states. Refer to Appendix 2 for character descriptions.

Material fixed in 70\% ethanol was cleaned using a sonicator, and subsequently dehydrated with acetone. One pecten per species was dissected, fixed on a stub, and dried in an oven for approximately 8 hours. The material was covered with gold using a Sputter Coater Balzer SCD 50, and images of the pectines prepared using a ZEISS DSM 940 scanning electronic microscope at USP. Ultraviolet fluorescence images of sternite III were also prepared, using a Microptics ML-1000 digital imaging system (Prendini 2003a; Volschenk 2006).

Stridulation behavior was observed and recorded in ten species: H. abudi; I. guanambiensis; J. agamemnon; J. pintoi; J. rochae; T. translucidus; T. lacrau; P. debilis; P. striatus; R. crassicauda.

## 3. Results

### 3.1. Separate morphological analyses

Parsimony analysis of the morphological character matrix with equal weighting obtained five most parsimonious trees (MPTs) with a length of 380 steps, Consistency Index (CI) of 0.32 , and Retention Index (RI) of 0.62 , in two islands of tree topologies (Table 7). Tityus and Za bius consistently formed a monophyletic group, with Za bius monophyletic, and Tityus paraphyletic with respect to Zabius. Centruroidinae was consistently monophyletic and comprised seven primary clades corresponding to


Fig. 8. Phylogeny of the New World buthid scorpion subfamily Centruroidinae Kraus, 1955 obtained by separate analysis of 90 morphological characters with parsimony. Single MPT with alternative topology obtained by analysis under equal weighting and implied weighting with mild concavity ( $k=3-6$ ). Synapomorphies optimized with accelerated transformation. Black circles indicate uniquely derived apomorphic states, white circles indicate parallel derivations of apomorphic states, numbers above indicate characters, and numbers below indicate states. Refer to Appendix 2 for character descriptions.
genera Centruroides, Heteroctenus, Ischnotelson, Jaguajir, Physoctonus, Rhopalurus and Troglorhopalurus. Four of the MPTs were almost identical, exhibiting only minor rearrangements among the species of Centruroides and Rhopalurus, as indicated by the strict consensus (Fig. 7). These topologies placed Centruroides sister to the remaining six centruroidine genera, all previously accommodated within Rhopalurus: (Centruroides (Troglorhopalurus (Physoctonus (Ischnotelson (Rhopalurus (Heteroctenus + Jaguajir)))))). The fifth MPT recovered by the equal weighting analysis differed from the other MPTs by reversing the positions of Physoctonus and Troglorhopalurus, and placing Centruroides in a clade with Heteroctenus, sister to a clade comprising Ischno-
telson, Jaguajir and Rhopalurus (Fig. 8): (Physoctonus (Troglorhopalurus ((Ischnotelson (Jaguajir + Rhopalurus)) (Centruroides + Heteroctenus))))).

Analyses with implied weighting under $k=1-6$ obtained a single MPT in each case. As in the equal weighting analysis, Tityus and Zabius consistently formed a clade, with Zabius monophyletic, and Tityus paraphyletic with respect to Zabius. Centruroidinae was consistently monophyletic and comprised seven primary clades corresponding to the abovementioned genera. However, two alternative topologies were recovered, depending on the strength of weighting against homoplasy (the concavity constant, $k$ ). The topology obtained by analyses with mild concavity ( $k=3-6$ ) was identical to the fifth MPT ob-


Fig. 9. Phylogeny of the New World buthid scorpion subfamily Centruroidinae Kraus, 1955 obtained by separate analysis of 90 morphological characters with parsimony. Single most parsimonious tree obtained by analysis under implied weighting with strong concavity ( $k$ $=1-2$ ). Synapomorphies optimized with accelerated transformation. Black circles indicate uniquely derived apomorphic states, white circles indicate parallel derivations of apomorphic states, numbers above indicate characters, and numbers below indicate states. Refer to Appendix 2 for character descriptions.
tained by the analysis with equal weighting (Fig. 8). The topology obtained by the analyses with strong concavity ( $k=1$ or 2 ) recovered Tityus monophyletic and placed Centruroides sister to a clade comprising Heteroctenus, Ischnotelson, Jaguajir and Rhopalurus, to the exclusion of Physoctonus and Troglorhopalurus (Fig. 9): (Physoctonus (Troglorhopalurus (Centruroides (Heteroctenus (Ischnotelson (Jaguajir + Rhopalurus)))))). Heteroctenus was paraphyletic with respect to Jaguajir.

Leaf stability indices of the topology obtained by separate analyses of the morphological character matrix with equal weighting and implied weighting under $k=3-6$ were greater than 0.98 for the outgroup taxa (Tityus and

Zabius) indicating that the monophyly of Centruroidinae is stable and most rearrangements occur among its component genera. Leaf stability indices for the seven genera were also high, varying between 0.80 and 0.88 , indicating that the monophyly of these genera is well supported, despite some uncertainty regarding their relative positions.

### 3.2. Separate molecular analyses

Parsimony analysis of the concatenated molecular dataset produced 14 MPTs of 6,688 steps, CI of 0.33 and RI of 0.75 . The only topological disagreement concerned


Fig. 10. Phylogeny of the New World buthid scorpion subfamily Centruroidinae Kraus, 1955 obtained by separate analysis of concatenated DNA sequences from three mitochondrial (12S rDNA, 16 S rDNA, Cytochrome $c$ Oxidase I) and two nuclear ( 18 S rDNA, 28 S rDNA) gene loci with Bayesian Inference. Maximum clade credibility tree with posterior probabilities at nodes before backslash. Nearly identical topology recovered with likelihood, bootstrap support at nodes after backslash.


Fig. 11. Phylogeny of the New World buthid scorpion subfamily Centruroidinae Kraus, 1955 obtained by simultaneous analysis of 90 morphological characters and concatenated DNA sequences from three mitochondrial ( 12 S rDNA, 16 S rDNA, Cytochrome $c$ Oxidase I) and two nuclear ( 18 S rDNA, 28 S rDNA) gene loci with parsimony under equal weighting. Strict consensus of 39 most parsimonious trees with bootstrap support values at nodes after backslash. Identical topology recovered with Bayesian Inference, maximum clade credibility tree, its posterior probabilities at nodes before backslash.
the relationships among conspecific terminals within $H$. princeps, P. debilis, and the three species of Rhopalurus. As in the separate analyses of the morphological character matrix, Zabius and the seven centruroidine genera were each consistently monophyletic. Additionally, Tity$u s$ was monophyletic, as in the morphological analyses with implied weighting under $k=1$ and 2 (Fig. 9). Unlike the morphological analyses, however, Centruroidinae was rendered paraphyletic by the placement of Ischnotelson sister to Tityus, to the exclusion of Zabius, placed sister to the remaining genera of Centruroidinae. Among the remaining centruroidine genera, Centruroides was
monophyletic with Heteroctenus, as in the morphological analyses with equal weighting and implied weighting under $k=3-6$ (Fig. 8): (Troglorhopalurus ((Centruroides + Heteroctenus) (Jaguajir (Physoctonus + Rhopalurus)))).

Similarly, Bayesian Inference recovered a paraphyletic Centruroidinae with the placement of Ischnotelson sister to Tityus, to the exclusion of Zabius, placed sister to the remaining genera of Centruroidinae (Fig. 10). Among the remaining centruroidine genera, Centruroides was monophyletic with Heteroctenus, as in the morphological analyses with equal weighting and implied weighting under $k=3-6$ (Fig. 8), but placed sister to a clade com-
prising Physoctonus and Rhopalurus, to the exclusion of Jaguajir and Troglorhopalurus: (Troglorhopalurus (Jaguajir ((Centruroides + Heteroctenus) (Physoctonus + Rhopalurus)))).

Likelihood analysis of the molecular dataset again recovered the monophyly of all genera, including Tityus, and the clades comprising Centruroides and Heteroctenus, and Physoctonus and Rhopalurus. Unlike the parsimony analysis, however, Centruroidinae was monophyletic, with the following relationships among its component genera: (Ischnotelson ((Physoctonus + Rhopalurus) (Troglorhopalurus (Jaguajir (Centruroides + Heteroctenus))))).

### 3.3. Simultaneous analyses

Simultaneous analysis of the morphological character matrix and the concatenated molecular dataset with equal weights parsimony retrieved 39 MPTs of 7,097 steps, CI 0.30 and RI 0.75 (Fig. 11). Tityus, Zabius and the seven centruroidine genera were each consistently monophyletic with high support. As in the parsimony analyses of the molecular dataset, however, Centruroidinae was rendered paraphyletic by the placement of Zabius sister to Tityus, to the exclusion of Ischnotelson (Fig. 11). Alternative hypotheses for the relative positions of these three groups resulted in a basal polytomy between Za bius, a weakly supported clade comprising Ischnotelson and Tityus, and a clade comprising the remaining genera of Centruroidinae. Relationships among the remaining centruroidine genera were better supported and mostly congruent with the topologies recovered by the separate analyses of the morphological and molecular data. Jaguajir was placed sister to a clade comprising Physoctonus and Rhopalurus, which together was sister to a clade comprising Centruroides and Heteroctenus, to the exclusion of Troglorhopalurus: (Troglorhopalurus $(($ Centruroides + Heteroctenus) (Jaguajir (Physoctonus + Rhopalurus)))).

The maximum clade credibility tree obtained from the simultaneous analysis with MrBayes was well supported (Fig. 11). Tityus, Zabius, and the seven centruroidine genera were each monophyletic with posterior probabilities $(\mathrm{PP})=1$. Tityus and Zabius formed the monophyletic sister group ( $\mathrm{PP}=0.96$ ) of a monophyletic Centruroidinae ( $\mathrm{PP}=$ 0.72 ). Relationships among the genera of Centruroidinae resembled those obtained by the simultaneous analysis with parsimony, except for the placement of Ischnotelson. Jaguajir was placed sister to a clade comprising Physoctonus and Rhopalurus ( $\mathrm{PP}=1$ ), in turn placed sister to a clade comprising Centruroides and Heteroctenus ( $\mathrm{PP}=$ 1), to the exclusion of Troglorhopalurus.

### 3.4. Preferred hypothesis

The tree topologies obtained from separate and simultaneous analyses using various analytical and sampling methods were mostly congruent. The preferred hypothe-


Fig. 12. Characters associated with pecten-sternite stridulation (Table 3, Appendix 2) in the New World buthid scorpion subfamily Centruroidinae Kraus, 1955. PEP = proximally expanded pectines. White squares $=$ stridulatory state absent (plesiomorphic state). Bicolored squares $=$ intermediate state. Black squares $=$ stridulatory state present (apomorphic state). Two squares $=$ both character states present
sis for the relationships among the genera of Centruroidinae is the topology recovered by simultaneous analysis of the molecular and morphological data, likelihood analysis of the molecular dataset, and one of the topologies recovered in the parsimony analysis morphology dataset (Fig. 11). This topology was mostly congruent with the consensus of the MPTs obtained from the separate morphological analyses with equal weights and implied weights under mild concavity ( $k=3-6$ ).

### 3.5. Stridulation

The morphology of the pecten-sternite stridulation organ varies among the species of Centruroidinae (Table 8, Figs. 3-5, 12). A stridulation organ was considered present in species that possess granular depressions on sternite III (pars stridens) and regular (i.e., continuous and approximately parallel) striations on the dorsal surfaces of the pectinal teeth (plectrum). Stridulation does not occur in Alayotityus, Ischnotelson, Isometrus, Mesotityus, Physoctonus, Tityus, and Troglorhopalurus, due to the absence of one or both structures.

The pars stridens is synapomorphic for the Centruroidinae, but exhibits varying levels of development among the genera, e.g., the granules are small in most genera, but large in Jaguajir. Its absence in some Centruroides, Physoctonus and Troglorhopalurus is considered an independent secondary loss in each case. A dorsal ridge on the pectinal teeth arose independently in Jaguajir and some Heteroctenus.

The plectrum arose within the centruroidine clade that excludes Ischnotelson. The striations reverted to ir-

Table 8. Morphological characteristics of the pecten-sternite stridulation organ of the species of the New World buthid genera Heteroctenus Pocock, 1893, Ischnotelson Esposito et al., 2017, Jaguajir Esposito et al., 2017, Physoctonus Mello-Leitão, 1934, Rhopalurus Thorell, 1876, and Troglorhopalurus Lourenço et al., 2004. Characters that define the presence of a stridulation organ indicated in boldface. Numbers in parentheses refer to character numbers in the morphological matrix (Table 3, Appendix 2).

|  |  | Pars stridens <br> Sternite III Lateral Depressions (51) | Proximally Expanded <br> Pectines | Plectrum |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  | Pectinal Teeth Striations (40) |  | Pectinal Teeth Dorsal Keel (39) | Pectinal Teeth Nodules (41) |
| Type IV-A | J. agamemnon |  | Present, large granules | $>2 \times$ medial | Present, parallel | Present | Absent |
|  | J. pintoi | Present, large granules | $>2 \times$ medial | Present, parallel | Present | Absent |
|  | J. rochae | Present, large granules | $>2 \times$ medial | Present, parallel | Present | Absent |
| Type IV-B | H. abudi | Present, small granules | $1.5 \times$ medial | Present, parallel | Present | Present |
|  | H. bonettii | Present, small granules | $>2 \times$ medial | Present, parallel | Present | Present |
|  | H. junceus | Present, small granules | $>2 \times$ medial | Present, parallel | Present | Present |
|  | H. princeps | Present, small granules | $1.5 \times$ medial | Present, parallel | Present | Present |
|  | H. garridoi | Present, small granules | $>2 \times$ medial | Present, parallel | Absent | Present |
| Type IV-C | R. laticauda | Present, small granules | $1.5 \times$ medial | Present, parallel | Absent | Absent |
|  | R. caribensis | Present, small granules | $1.5 \times$ medial | Present, parallel | Absent | Absent |
|  | R. crassicauda | Present, small granules | $1.5 \times$ medial | Present, parallel | Absent | Absent |
| Absent | T. lacrau | Absent, irregular | $1.5 \times$ medial | Present, irregular | Absent | Absent |
|  | T. trans/ucidus | Absent, irregular | $1.5 \times$ medial | Present, irregular | Absent | Absent |
|  | 1. guanambiensis | Present, small granules | Absent | Absent | Absent | Present |
|  | 1. peruassu | Present, small granules | Absent | Absent | Absent | Present |
|  | P. debilis | Absent | $1.5 \times$ medial | Absent | Absent | Absent |
|  | P. striatus | Absent | $1.5 \times$ medial | Absent | Absent | Absent |

regular in Centruroides and were lost in Physoctonus.
Pronounced proximal expansion of the pectinal lamellae, observed in the clade containing Centruroides, Heteroctenus, and Jaguajir, was secondarily reduced in some Heteroctenus (e.g., H. princeps) and lost in Centruroides.

## 4. Discussion

### 4.1. Centruroidinae and Centruroides monophyletic

Centruroides, Physoctonus, Troglorhopalurus and the species previously assigned to Rhopalurus (Esposito et al. 2017) were monophyletic and well-supported in all except two topologies, confirming the monophyly of Centruroidinae. Among Buthidae, the presence of prolateral accessory denticles in the median denticle rows of the pedipalp chela fingers is uniquely synapomorphic for the subfamily (Fig. 1). Additional synapomorphies include a transverse row of median tubercles on the chelicerae, dorsobasal setation on the cheliceral fixed finger, and a bifurcated prolateral pedal spur on leg $I$, reduced in Physoctonus.

Centruroides was also consistently monophyletic and well-supported in the analyses presented, contradicting the findings of Teruel et al. (2006), in which Cuban species of Centruroides were paraphyletic with respect to Cuban species of Rhopalurus, currently placed in Heteroctenus (Esposito et al. 2017). Morphological synapomorphies of Centruroides include convergence
of the prodorsal and proventral carinae of the pedipalp patella, trichobothrium $d b$ of the fixed finger of the pedipalp chela aligned with or distal to trichobothrium et, trichobothrium est of the fixed finger situated between trichobothria $d b$ and et or proximal to et, and metasomal segment V elongate, particularly in adult males (length $>$ $2.5 \times$ width).

### 4.2. Rhopalurus s.l. paraphyletic

The group of species assigned to Rhopalurus by previous authors (Fet \& Lowe 2000; Teruel 2006; Teruel \& Armas 2006, 2012; Lourenço 2007, 2008, 2014; Teruel \& Roncallo 2008, 2013; Teruel \& Tietz 2008; Prendini et al. 2009; Santiago-Blay 2009; Flórez 2012) was consistently paraphyletic in the analyses presented here, contradicting the suggestion by Lourenço (1986) that these species are united by the presence of a pectensternite stridulation organ (Figs. 3-5). The components of Rhopalurus sensu lato consistently formed six wellsupported monophyletic groups, the species composition of which comes as little surprise, given their disjunct distributions (Fig. 6).

Heteroctenus, removed from synonymy with Rhopalurus by Esposito et al. (2017), comprises all former species of Rhopalurus occurring in the Greater Antilles, and represented in the analyses by the type species, $H$. junceus, and five species transferred from Rhopalurus by Esposito et al. (2017): H. abudi and its junior synonym, R. virkkii; H. bonettii; H. garridoi; H. gibarae; H. princeps.

Two genera, recently created by Esposito et al. (2017), accommodate two distinct groups of species, all
except two of which were transferred from Rhopalurus by Esposito et al. (2017). Ischnotelson comprises two species characterized by a very narrow telson, I. guanambiensis and I. peruassu, from the caatinga and cerrado of northeastern Brazil. Jaguajir comprises three large-bodied species from northern Brazil: J. pintoi from savanna formations on the Guiana Shield, and J. agamemnon and $J$. rochae from the caatinga and cerrado of northeastern Brazil.

Physoctonus comprises two small epigean species from the arid caatinga of northeastern Brazil, P. debilis, transferred to Rhopalurus by Francke (1977) and reinstated by Lourenço (2007), and P. striatus.

Rhopalurus comprises three compact species with moderately pale coloration, from savanna formations on the Guiana Shield of northern South America, the type species, R. laticauda, R. caribensis, and R. ochoai.

Troglorhopalurus comprises two species from caves in northeastern Brazil, the troglobite T. translucidus and the troglophile T. lacrau, transferred from Rhopalurus by Esposito et al. (2017).

### 4.3. Heteroctenus revalidated

Рососк (1893) created Heteroctenus to accommodate three species that were, at the time, placed in Centrurus (later transferred to Centruroides), designated the Cuban species, H. junceus, as type species and noted in the description that Heteroctenus was closely allied with Centrurus. Рососк (1902) synonymized Heteroctenus with Rhopalurus. However, in the analyses presented here, the characters on which Heteroctenus was originally defined, i.e., proximally expanded pectinal lamellae, enlarged pectinal plate, and pronounced median carina on mesosomal sternite III, were found to be synapomorphic for the Caribbean species of Rhopalurus and form the justification, in combination with additional morphological and molecular evidence, for revalidating Heteroctenus and transferring the Caribbean species to it. Additional morphological synapomorphies for the genus include: dorsal surfaces of proximal pectinal teeth with regular striations and multiple nodules, lateral margins of sternite III with smooth carina, and telson without subaculear tubercle.

The analyses presented here also clarified the status of $R$. virkkii, described from Isla Mona, an islet between Hispaniola and Puerto Rico (Santiago-Blay 2009). Teruel \& Armas (2012) suggested R. virkkii might be synonymous with $R$. abudi, described from Isla Saona, Dominican Republic, off the southwestern coast of Hispaniola, and later reported from mainland Hispaniola (Prendin et al. 2009), but were unable to examine material from Isla Mona. Based on evidence presented here, R. virkkii is merely a pale color form of $H$. abudi, with little genetic divergence from the mainland population thereof, justifying the synonymy by Esposito et al. (2017).

The validity of four Cuban taxa, referable to Heteroctenus, merits further discussion. Esposito et al. (2017)
synonymized $R$. aridicola and $R$. melloleitaoi with $H$. junceus, based on the absence of convincing morphological differences or evidence of geographical isolation from the latter and, in the case of $R$. aridicola, on the low genetic divergence between topotypes thereof and samples conspecific with $H$. junceus, presented here. Rhopalurus granulimanus was synonymized with H. gibarae based on the absence of convincing morphological differences, the limited sample size, and the observation that the type localities of the two taxa are less than 25 km apart (Esposito et al. 2017). Approximately nine days after publication of these synonyms, Armas (2017) published a rebuttal in an online journal that claims to be peerreviewed, revalidating the three taxa synonymized by Esposito et al. (2017), and formally transferring each to Heteroctonus. The arguments presented by Armas (2017) are unconvincing, however, for the following reasons

In justifying the revalidation of $H$. aridicola, Armas (2017) presented four arguments, to which we respond in turn. (1) Heteroctenus aridicola differs from H. junceus based on the presence of (i) stronger metasomal carinae; (ii) a more attenuated metasoma in the male; and (iii) a very small proximal gap between the pedipalp chela fingers of the male (Armas 2017). In our experience, a wide range of variation in granulation/carination, metasomal width, and size of the proximal gap between the pedipalp chela fingers of the male is evident across the distribution of $H$. junceus, and in other widespread species of Heteroctenus, and the variation described for H. aridicola falls well within this range. As such, these characters are unreliable for species diagnosis, especially when comparing small samples. (2) Heteroctenus aridicola and $H$. junceus are sympatric and syntopic (Armas 2017). The reasoning behind this argument is circular. These concepts, by definition, assume the presence of more than one species and therefore cannot be used to justify the existence of more than one species a priori. (3) In the laboratory, H. aridicola and H. junceus are capable of interbreeding but immatures resulting from those breedings died prior to reaching adulthood, suggesting postzygotic reproductive isolation (Armas 2017). Failure to reach adulthood in captivity is not proof of reproductive isolation. The immatures from those breedings may have died for other reasons. Furthermore, data presented herein includes specimens matching the description of $H$. aridicola and collected from within the range defined for that species, yet which cannot be distinguished genetically from specimens collected elsewhere across the range of $H$. junceus, falsifying the hypothesis of reproductive isolation. (4) Heteroctenus aridicola is endemic to the xerophytic coastal area between Punta Negra and Punta de Maisí, whereas specimens from Santa Rosa and Baracoa appear to be accidental introductions (Armas 2017). This argument appears to contradict the argument based on sympatry (2) and, as stated, a sample from Baracoa was genetically indistinguishable from other samples of $H$. junceus, suggesting panmixis.

Concerning the synonymy of $H$. melloleitaoi, Armas (2017) presented three arguments, to which we respond
in turn. (1) Heteroctenus melloleitaoi was described from five localities in the Niquero Municipality of Granma Province not "a single locality" (Armas 2017). Nevertheless, the distance between the furthermost localities of $H$. melloleitaoi is less than 15 km , and all five localities occur within the Parque Nacional Desembarco del Granma, throughout which H. junceus is also distributed (Esposito et al. 2017). (2) Heteroctenus melloleitaoi was collected syntopically with $H$. junceus not "in close proximity to many known locality records of $H$. junceus" (Armas 2017). As stated above, the reasoning behind this argument is circular. This concept, by definition, assumes the presence of more than one species and therefore cannot be used to justify the existence of more than one species a priori. (3) Heteroctenus melloleitaoi differs from H. junceus in the metasoma and pedipalps being more attenuated, mainly in the females, and the significantly higher pectinal tooth count (Armas 2017). As previously stated, variation in meristics and other characters is observed across the distribution of $H$. junceus and other widespread species of Heteroctenus. The pectinal tooth count allegedly diagnostic for $H$. melloleitaoi does not differ statistically from that of $H$. junceus.

With respect to Armas' (2017) criticism that the type material was not examined, it should be noted that the decisions of Esposito et al. (2017) were based on data presented in the published diagnoses and accompanying illustrations of these taxa, which are presumed to be sufficient to document the variation (indeed, L.F. de Armas and colleagues regularly publish taxonomic decisions based solely on literature and/or photographs of specimens unavailable for loan to Cuba). Based on the available evidence, the following synonyms are therefore upheld: Rhopalurus granulimanus Teruel, $2006=$ Heteroctenus gibarae (Teruel, 2006); Rhopalurus melloletaoi Teruel \& Armas, 2006 and Rhopalurus aridicola Teruel \& Armas, 2012 = Heteroctenus junceus (Herbst, 1800). The validity of $H$. gibarae, as distinct from $H$. garridoi, will be reassessed when material becomes available for study.

### 4.4. Additional genera from Brazil

The species formerly assigned to Rhopalurus from northern and northeastern Brazil formed two clearly defined, monophyletic groups in the analyses presented here, justifying the creation of two genera by Esposito et al. (2017). Ischnotelson accommodates two unusual Brazilian species, $R$. guanambiensis and a second, allopatric species, which share a uniquely narrow telson in addition to fused lateral ocular, central lateral, and posterior central submedian carinae of the carapace. Jaguajir accommodates three morphologically diverse, large-bodied species, R. agamemnon, R. pintoi, R. rochae, united by the possession of fused lateral ocular and anterior central submedian carinae on the carapace.

Although unequivocally monophyletic, the phylogenetic positions of Ischnotelson and, to a lesser extent,

Jaguajir were unstable in the analyses presented here. Whereas the two genera formed a monophyletic group with Rhopalurus in the separate morphological analyses, these genera were not monophyletic with one another or with Rhopalurus in the separate parsimony and likelihood analyses of the molecular dataset, which placed Ischnotelson sister to Tityus, rendering Centruroidinae paraphyletic, or in the combined analyses, which placed Ischnotelson sister to a group comprising all other Centruroidinae. The placement of Ischnotelson sister to all other centruroidine genera is the most plausible reconstruction, based on the presence of accessory denticles in the median denticle rows of the pedipalp chela fingers, an incrassate pedipalp chela manus in the adult male, and a posterior widening of the metasoma. Jaguajir was consistently placed sister to Rhopalurus or (Heteroctenus + Centruroides), except for one topology resulting from the morphological analysis, in which it rendered Heteroctenus paraphyletic. Its consistent placement sister to (Heteroctenus + Centruroides) in topologies recovered by the separate and combined analyses is the most plausible hypothesis.

The analyses presented here also clarified the status of several infrageneric taxa assigned to Jaguajir by Esposito et al. (2017), the validity of which was previously confused. Lourenço (1982, 1984, 1986a,b, 1992, 1997) relegated R. pintoi to a subspecies of R. laticau$d a$ and synonymized $R$. crassicauda therewith but later (Lourenço \& Pinto-da-Rocha 1997) described another species, $R$. piceus, from the vicinity of the type locality of R. pintoi. KovaR̆ík (1998) listed R. pintoi at the rank of species but Fet \& Lowe (2000) continued to list it as a subspecies of $R$. laticauda in accordance with Lourenço (1982). Lourenço (2002) formally reinstated R. pintoi and removed $R$. crassicauda from synonymy. Teruel (2006) suggested $R$. pintoi might be a senior synonym of R. piceus. Teruel \& Tietz (2008) formally synonymized R. piceus, erroneously declaring $R$. pintoi to be a nomen nudum, and questioned whether $R$. crassicauda is distinct from R. laticauda. Lourenço (2008) suggested R. piceus may yet prove to be valid and rejected the suggestion that $R$. crassicauda is a junior synonym of $R$. laticauda, instead proposing it might be a subspecies thereof, and creating a new subspecies, R. crassicauda paruensis, along with a new subspecies of $R$. pintoi. Prendini et al. (2009), however, agreed with the synonymy of $R$. piceus with R. pintoi by Teruel \& Tietz (2008), and the suggestion that $R$. crassicauda is probably a junior synonym of R. laticauda. The evidence and analyses presented here supported the validity of $J$. pintoi as distinct from R. laticauda, upheld the synonymy of $R$. piceus therewith, and justified the synonymy of $R$. piceus and R. pintoi kourouensis by Esposito et al. (2017). Rhopalurus crassicauda, on the other hand, was determined to be conspecific with R. laticauda and synonymized by Esposito et al. (2017). Additionally, R. acromelas was demonstrated to be conspecific with J. agamemnon, justifying its synonymy, and that of its previous synonyms, Rhopalurus melleipalpus Lutz \& Mello, 1922, Rhopalurus iglesiasi Werner, 1927, Rhopalurus lambdophorus Mello-Leitão, 1932, Rho-
palurus dorsomaculatus Prado, 1938, and Rhopalurus goiasensis Prado, 1940, by Esposito et al. (2017).

### 4.5. Physoctonus validated

Physoctonus debilis was originally placed in the nonbuthid genus Vaejovis C.L. Koch, 1836 but was transferred to Rhopalurus by Borelli (1910) where it remained until Lourenço (2002) resurrected Physoctonus. Physoctonus, created to accommodate Physoctonus physurus Mello-Leitão, 1934, was synonymized with Rhopalurus when Francke (1977) synonymized P. physurus with Rhopalurus debilis.

Physoctonus debilis and a second species described by Esposito et al. (2017) were consistently monophyletic in the analyses presented here, justifying Lourenço's (2002) decision to reinstate the genus. Physoctonus is supported by several morphological synapomorphies: pedipalp femur with retrolateral accessory carina; pectinal proximal dorsal fulcrae asetose; telson slightly ovate (length ca. $1.5 \times$ width), metasomal segment V without ventrosubmedian carina; sternite III surface planar, i.e., without anterior elevation.

The phylogenetic position of Physoctonus within Centruroidinae remains somewhat uncertain. Physoctonus was consistently placed sister to Rhopalurus in the separate analyses of the molecular data and the simultaneous analyses of the morphological and molecular data, an unexpected relationship, given the allopatric distributions of these taxa. In contrast, separate analyses of the morphological data consistently placed Physoctonus sister to a monophyletic group comprising all centruroidine genera except Troglorhopalurus.

### 4.6. Rhopalurus redefined

As redefined by Esposito et al. (2017), Rhopalurus comprises only three species, $R$. caribensis, $R$. laticauda, and R. ochoai, united by the following morphological synapomorphies: fused central lateral and posterior central submedian carinae of the carapace, and the presence of a pecten-sternite stridulation organ (proximal pectinal teeth, dorsal surfaces without nodules but with regular striations, sternite III, ventromedian carina elevated anteriorly, ventrosubmedian surfaces forming paired depressions, finely and irregularly granular, lateral margins forming smooth, raised carina). Rhopalurus are savanna specialists, endemic and allopatrically distributed in savanna formations on the Guiana Shield of northern South America.

The status of $R$. caribensis, occurring in the Llanos of the Magdalena, Colombia, and separated from the nearest populations of $R$. laticauda by the Cordillera de Mérida (Andes), has been the subject of controversy. Lourenço (2008) suggested R. caribensis is a morph of R. laticau$d a$ rather than a distinct species. Florez (2012) adopted this suggestion and synonymized $R$. caribensis with $R$. laticauda based in part on images of Tityus alleged to be
R. caribensis. Teruel \& Roncallo (2013) subsequently revalidated $R$. caribensis. The analyses presented here supported the validity of R. caribensis based on genetic divergence from $R$. laticauda. Although morphologically similar, the two species can be consistently diagnosed by the color pattern on the ventral surface of the metasoma. Whereas $R$. caribensis displays three distinct, narrow stripes of pigmentation along the ventral surface (a ventromedian stripe flanked on either side by a ventrosubmedian stripe), these stripes are fused into a single, broad band of pigmentation (more pronounced in populations from the southeast of the distribution, formerly referable to R. amazonicus) in R. laticauda.

Unlike $R$. caribensis, no evidence was found to support the continued recognition of R. amazonicus, justifying its synonymy with $R$. laticauda by Esposito et al. (2017). According to Lourenço (2008), R. amazonicus inhabits savanna "islands" surrounded by rainforest, but the genetic results presented here indicate substantial gene flow between these populations. The noticeably darker and more contrasting coloration of populations referable to $R$. amazonicus, compared with typical populations of $R$. laticauda to the north and west, was found to represent a difference in the intensity, rather than the pattern, of pigmentation. For example, these populations exhibit the single, broad band of pigmentation along the ventral surface of the metasoma, only more pronounced than observed in typical populations of $R$. laticauda.

The validity of $R$. crassicauda, another species repeatedly confused with $R$. laticauda, has been subject to considerable speculation (Lourenço 1982, 2002, 2008; Teruel \& Tietz 2008; Prendini et al. 2009). When Lourenço (1982) relegated $R$. pintoi to a subspecies of $R$. laticauda, R. crassicauda was synonymized therewith. Ten years later, when Lourenço (2002) reinstated R. pintoi, R. crassicauda was removed from synonymy. Teruel \& Tietz (2008) questioned whether $R$. crassicauda is distinct from $R$. laticauda but refrained from a formal synonymy in the absence of material for examination. Lourenço (2008) rejected the suggestion that $R$. crassicauda is a junior synonym of $R$. laticauda, proposing instead that it might be a subspecies thereof, and creating a new subspecies, $R$. crassicauda paruensis. Prendini et al. (2009), however, agreed with Teruel \& Tietz (2008) that R. crassicauda is probably a junior synonym of $R$. laticauda, and emphasized the need to clarify the distinction between R. laticauda, R. crassicauda and its subspecies. Based on the evidence presented here, R. crassicauda is indeed conspecific with $R$. laticauda and its subspecies, justifying its synonymy by Esposito et al. (2017).

One possible explanation for the lack of clear diagnostic characters among the species of Rhopalurus is the relatively short time period since the last glacial maxima, during which time the northern savannas of South America expanded and reconnected with one another (Lourenço 2008). Despite the limited genetic differentiation within Rhopalurus, a distinct group of populations, representing a previously unrecognized species, R. ochoai, distributed
around Lake Maracaibo, east of the Cordillera de Perijá, and north and west of the Cordillera de Mérida, was identified and described by Esposito et al. (2017).

### 4.7. Troglorhopalurus redefined

The monotypic genus Troglorhopalurus was created to accommodate Troglorhopalurus translucidus Lourenço et al., 2004, based on a single, troglomorphic specimen from a Brazilian cave. In comparing Troglorhopalurus with Rhopalurus, Lourenço et al. (2004: 1153, 1156) noted that "all modifications presented by the new troglobitic scorpion are the result of adaptation to a cave dwelling life," prompting Prendini et al. (2009) to suggest that Troglorhopalurus might be a junior synonym of Rhopalurus. A troglophile species, Rhopalurus lacrau Lourenço \& Pinto-da-Rocha, 1997, had been described from caves belonging to the same subterranean formation in Brazil and, in the description of Troglorhopalurus, Lourenço et al. (2004) suggested the relationship between these taxa should be investigated using molecular data. Accordingly, the consistent placement of $R$. lacrau sister to T. translucidus in the analyses presented here comes as little surprise, and justifies the transfer of $R$. lacrau (and its junior synonym, Rhopalurus brejo Lourenço, 2014) to Troglorhopalurus by Esposito et al. (2017).

Several morphological synapomorphies support Troglorhopalurus, as redefined by Esposito et al. (2017): pectinal peg sensillae elongate and acuminate; metasomal segment V elongate (length $>2.5 \times$ width); telson slightly ovate (length ca. $1.5 \times$ width); telson vesicle width approximately equal to metasomal segment V width. The metasomal and telson characters resemble characters observed in Centruroides, and are generally associated with elongation of the metasoma. However, these characters apparently evolved convergently in Troglorhopalurus and are presumed to be an adaptation to life in caves (Prendini et al. 2009).

### 4.8. Evolution of stridulation

Stridulation, defined as the emission of sound by rubbing together different parts of the body - typically a structure, appendix or projection, i.e., the plectrum, against a modified surface, i.e., the pars stridens - is used for intraspecific (e.g., mating behavior) and/or interspecific (e.g., defense) communication (Dumortier 1964a). Stridulation in scorpions is usually associated with defense behavior (Dumortier 1964b; Alexander 1958; Acosta \& Maury 1990; McCormick \& Polis 1990; Lourenço \& Cloudsley-Thompson 1995; Prendini 2001a; Prendini et al. 2003). Dumortier (1964a) recognized four different types of stridulation organs in scorpions, according to the structures of which they are comprised (Dumortier 1964a): type I, pedipalp-leg (scorpionids of the genera Heterometrus, Pandinus Thorell, 1876 and relatives); type II, chelicera-cephalothorax (scorpionids of the ge-
nus Opistophthalmus C.L. Koch, 1837); type III, meta-soma-aculeus (buthids of the genus Parabuthus Pocock, 1890); and type IV, pecten-sternite (Centruroidinae). Acosta \& Maury (1990) described a fifth type, in which sound is produced by the friction of tergites III-VI, in the bothriurid genus Timogenes Simon, 1880; a similar stridulatory apparatus was reported in another bothriurid, Brachistosternus ehrenbergii (Gervais, 1841) by Осноа \& Ojanguren-Affilastro (2007).

Type IV stridulation, caused by friction of nodules and striations on the dorsal surface of the pectinal teeth (plectrum) with granulation in depressions on the ventral surface of sternite III (pars stridens), was first recorded among Centruroidinae by W.J. Burchell during a visit to Brazil in 1828. Burchell observed the behavior in a species later determined by Рососк (1904) to be Rhopalurus borelli, a junior synonym of Jaguajir agamemnon (Lourenço \& Cloudsley-Thompson 1995). The pectensternite stridulation organ was historically regarded as a diagnostic character of Rhopalurus (Lourenço 1982; Sissom 1990; Lourenço \& Cloudsley-Thompson 1995; Lourenço et al. 2000; Fet et al. 2000) but is now known to exist in several species of Heteroctenus, Jaguajir, and Rhopalurus (Esposito et al. 2017).

Prior to the present study, the evolution of the pectensternite stridulation organ had not been investigated in a phylogenetic context (Prendini et al. 2009). As demonstrated by the analyses presented here, the organ is actually a complex of several characters associated with the pars stridens and the plectrum, which evolved independently in Centruroidinae. Three distinct types of pectensternite stridulation organ, first noted by Рососк (1904), may be recognized.

Type IV-A occurs in Jaguajir, the only genus in which audible stridulation accompanied by movement of the pectines has been observed (H.Y. Yamaguti, pers. obs.). This is the most pronounced stridulation organ. The pars stridens is characterized by deep lateral depressions on sternite III which are flat and coarsely granular across the surface (Fig. 3B). The pectinal lamellae are expanded proximally (Fig. 4A) such that the proximal width is more than twice the medial width. This membranous widening creates a tympanum-like structure, which might amplify the intensity of stridulation (Рососк 1904; Lourenço \& Cloudsley-Thompson 1995; Prendini et al. 2009). The pectinal teeth each possess a broad ridge on the dorsal surface (Fig. 4D), where deep, parallel striations are concentrated (Fig. 5A), creating a sinuous shape.

Type IV-B occurs in Heteroctenus and there are records of audible stridulation in species of the genus (Pocock 1904; Lourenço et al. 2000). This stridulatory apparatus is also well developed. The pars stridens is characterized by fine granulation on sternite III (Рососк 1904; Fig. 3C). Some species, such as H. princeps, possess lateral depressions on sternite III with irregular granulation. The pectines are expanded proximally (Fig. 4A) to varying degrees among the different species (Table 6). The striations on the dorsal surface of the pectinal teeth resemble those of Type A (Fig. 4B). Nodules on the stri-
ated surfaces of the pectinal teeth of Heteroctenus species may serve to increase the striated area (Fig. 5B), perhaps enhancing the audibility of stridulation (Lourenço et al. 2000). However, as the nodules are also found in I. guanambiensis, which does not possess a stridulation organ, their presence may not be directly related to stridulation.

Type IV-C occurs in Rhopalurus. This is the least developed type of stridulation organ (Pocock 1904; Teruel 2006; Teruel \& Roncallo 2008). The pars stridens is characterized by fine granulation on sternite III with shallower sternite depressions than observed in Types IV-A and IV-B (Fig. 3D). The pectinal lamellae are only slightly expanded proximally, such that the proximal width is about $1.5 \times$ the medial width (Figs. 4C,E). The dorsal surfaces of the pectinal teeth are flat (without a ridge) and the striations, although well defined, are less developed than in Types IV-A and IV-B (Fig. 5C). There are no records of pectinal movement or sound production in Rhopalurus species, despite the presence of a stridulation organ.

## 5. Conclusions

This study presents the first rigorous test of the monophyly of Centruroidinae and its component taxa, based on phylogenetic analysis of morphological characters and DNA sequence data from multiple gene loci. The benefits of integrating and simultaneously analysing diverse sources of data are evident in the insights gained concerning the monophyly, diagnostic characters and distributions of these scorpions from the species level on up. The need for improved understanding of the classification of Centruroidinae cannot be understated. Their unique stridulation ability, disjunct biogeographical distributions, and the medical potential of their venoms offer promise for future evolutionary, biogeographical and biomedical research. Their distribution in some of the most environmentally sensitive areas of the world, e.g., savanna surrounding the Amazon rainforest, make them prime candidates for studies on the impact of recent and future climate change.

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## 8. Appendix 1

Arguments justifying why Centruroidinae Kraus, 1955, not Centrurinae Koch, 1837, should be in Buthidae C.L. Koch, 1837, whereas Centrurinae Koch, 1837 should be in Scorpionidae Latreille, 1802.

Centrurus Ehrenberg, 1829: 350, originally published with a brief description, is an available name (Article 12.1) regardless of the absence of included species.

The first and only included available species was Centrurus galbineus Koch, 1838: 110-112, pl. CXXXIX, fig. 320, fixing it as the type species of Centrurus by subsequent monotypy. Косн (1838) described and illustrated the species, which is evidently an immature Heterometrus, and indicated that the locality was unknown. Fet \& Lowe (2000: 434) reported the type lost.

Kraepelin (1894: 34) synonymized C. galbineus with Heterometrus longimanus (Herbst, 1800), hence Centrurus became a junior synonym of Heterometrus Ehrenberg, 1828, fixing its current placement in Scorpioninae Latreille, 1802 (Article 61).

Косн (1837: 38) published and characterized "Centrurides" for two genera, Centrurus and Vaejovis C.L. Koch, 1836. Centrurides is an unlatinized, vernacular, family-group name formed from Centrurus, which is the type genus (Article 12.2.4). Without latinization, the name is unavailable (Article 11.7.2).

Centrurides was latinized as Centrurini and generally accepted as valid by subsequent authors (e.g., see Peters 1861: 512; Thorell 1876: 7; Thorell \& Lindström 1885: 25; Kraepelin 1891: 149), hence Centrurini Koch, 1837 was then available (Article 11.7.2). KraepeLin (1899) later changed the suffix from that of a tribe to that of a subfamily, Centrurinae.

Centrurini or Centrurinae, when cited in Buthidae C.L. Koch, 1837, are misidentifications, not synonymous with Buthidae (Fet \& Lowe 2000: 55, 56).

The correct placement of Centrurini or Centrurinae is in Scorpionidae, where Centrurinae should be cited as a synonym of the nominotypical subfamily Scorpioninae (Fet 2000: 427).

The type species of Centrurus is a junior synonym of Heterometrus longimanus (Herbst, 1800) (= Scorpio longimanus) (Kraepelin 1894: 34; Fet 2000: 434), hence Centrurus is a junior synonym of Heterometrus Ehrenberg, 1828 (Fet 2000: 431).

Centrurus is not a senior synonym of Centruroides or a nomen nudum as Francke (1985) suggested (see also ICZN 1986: 144, 145).

Given the preferred classification of Centrurus as a synonym of Heterometrus in the Scorpioninae, the fami-ly-group name based on Centrurus will necessarily have to be applied (as valid or as a synonym) in the Scorpioninae (Article 61).

The Principle of Typification renders it impossible for Centrurinae or Centrurini to be assigned to any family other than the one that harbors Centrurus galbineus.

Marx (1890: 211) established Centruroides by including one available species, Buthus exilicauda Wood, 1863 (Article 12.2.5). Centruroides was proposed as a genus in its own right, not as a replacement name for Centrurus. However, due to a misunderstanding of Centrurus, the name was often used to refer to species that belonged in Centruroides and the latter was erroneously considered a junior synonym of the former. That misunderstanding appears to have precipitated the idea that Centrurinae was the correct name of the buthid subfamily.

Early usages of Centruroidinae were invalid because the taxon was undescribed and the name accordingly unavailable (Roewer 1943: 218; Jaume 1954: 1087). Centruroidinae became available when Kraus (1955: 101) published a brief description in a footnote. Centruroidinae Kraus, 1955 is the correct name, not Centrurinae, Rhopalurinae or Rhopalurusinae.

Article 40.1 states that "When the name of a type genus of a nominal family-group taxon is considered to be a junior synonym of the name of another nominal genus, the family-group name is not to be replaced on that account alone." It might therefore be argued that Article 40.1 does not make a distinction regarding whether or not the junior synonym belongs to the same family, and therefore requires Centrurinae to be the correct name (O.F. Francke, pers. comm.). This argument overlooks The Principal of Typification (Article 61), however, which makes clear that the type of a name determines its placement in a classification and the type of a name cannot be separated from the name.

Article 40.2 states that "If, however, a family-group name was replaced before 1961 because of the synonymy of the type genus, the substitute name is to be maintained if it is in prevailing usage." The following arguments have been offered to justify that the concept of Centrurus and Centrurinae was always in Buthidae, not Scorpionidae, and Centruroidinae is not in prevailing usage (O.F. Francke, pers. comm.):

1. Between 1876 and 1900, eighteen species where described in Centrurus by Thorell (1876; $n=4$ ), Karsch (1879a,b; $n=2$ ), Kraepelin (1891, 1898; $n=2$ ), Рососк (1898; $n=9$ ) and BANKS ( $1900 ; n=1$ ). All were described in Buthidae, and today are in Centruroides and remain in Buthidae. The concept back then of Centrurus was of a buthid not a scorpionid. Kraepelin (1891) proposed subfamily Centrurini within Buthidae and Kraepelin (1899) used the proper ending for a subfamily, Centrurinae, five years after synonymizing Centrurus galbineus (KraepeLin 1894); the subfamily concept therefore remained within Buthidae.
2. The species known today as Centruroides gracilis (Latreille, 1804) was published as the combination Centrurus gracilis by nineteen different authors, always within Buthidae. Similarly, the species known today as Centruroides margaritatus (Gervais, 1841) was pub-
lished as Centrurus margaritatus twenty times, always within Buthidae.
3. Centruroidinae was used by only four authors between 1943 and 1998 (Fet \& Lowe 2000) whereas Centrurinae was used 23 times.

The argument that the concept of Centrurus and Centrurinae has always been in Buthidae neglects the identity and placement of the type species of Centrurus and
type genus of Centrurinae, which are in Scorpionidae. Centruroides was not proposed as a replacement name for Centrurus, but was proposed for Buthus exilicauda, a North American species, and for an undescribed species that was named but not described. The name Centrurus was incorrectly used for many years to denote species of Centruroides because of an erroneous understanding and placement of Centrurus.

## 9. Appendix 2

Tissue samples from which DNA was extracted and sequenced, and associated voucher specimens and additional material examined for morphological character matrix used in phylogenetic analysis of the New World buthid subfamily Centruroidinae Kraus, 1955, deposited in the following collections: American Museum of Natural History (AMNH), New York, U.S.A., incorporating the Alexis Harington (AH) Collection; Museum National d'Histoire Naturelle (MNHN), Paris, France; Museu de Zoologia da Universidade de São Paulo (MZSP), Brazil; Natal Museum (NM), Pietermaritzburg, South Africa; Museum of Natural History, Oxford University, U.K. (OUMNH); South African Museum (SAM), Cape Town; Natur-Museum Senckenberg, Frankfurt (SMF), Germany; Universidade Federal de Minas Gerais (UFMG), Belo Horizonte, Brazil; Zoologisches Museum der HumboldtUniversität, Berlin (ZMB), Germany; Zoologisches Museum der Universität Hamburg (ZMH), Germany. Samples, stored in the Ambrose Monell Cryocollection (AMCC) of the American Museum of Natural History, New York, and vouchers, in the AMNH Collection of Arachnida and Myriapoda, share the same number.

Centruroides exilicauda (Wood, 1863): MEXICO: Baja California Sur: Município Los Cabos: Cabo San Lucas, 15 mi . E, $22^{\circ} 53.383^{\prime} \mathrm{N} 109^{\circ} 54.933^{\prime} \mathrm{W}$, 1.vi.1999, M.E. Soleglad, 1 ô (AMCC [LP 1692]); San Bartolo, 6 km SW, $23^{\circ} 41.816^{\prime} \mathrm{N}$ $109^{\circ} 50.800^{\prime} \mathrm{W}, 100 \mathrm{~m}, 13 . v i i .2008$, H. Montaño and E. González, mesic vegetation, 1 q (AMCC [LP 8840]).
Centruroides gracilis (Latreille, 1804): MEXICO: Hidalgo: Município Tamazunchale: Tamazunchale, 24 km SE, $21^{\circ} 11^{\prime} 14^{\prime \prime} \mathrm{N}$ $98^{\circ} 54^{\circ} 16^{\prime \prime} \mathrm{W}$, 3.viii.2002, L. Prendini and E. González, under stones on grassy hillside (deforested), $1 \delta^{\lambda}, 2$ juv. (AMCC [LP 2013]). San Luis Potosí: Antena de Microondas de Ciudad Valles, $21^{\circ} 58^{\prime} 55^{\prime \prime} \mathrm{N} 99^{\circ} 08^{\prime} 58^{\prime \prime} \mathrm{W}, 340 \mathrm{~m}, 2$. viii.2002, L. Prendini, J. Soriano and E. González, forest (disturbed), collected at night with UV light, 4 §, 3 中, 3 juv. (AMCC [LP 2051]).
Centruroides infamatus (C.L. Koch, 1844): MEXICO: Michoacan: Município Tandamangapio: Los Tabanos, $19.9749^{\circ} \mathrm{N}$ $102.84226^{\circ}$ W, 223 m, 31.v.2006, O.F. Francke, H. Montaño, A. Valdez and A. Ballesteros, 2 \& (AMCC [LP 6420]). Município Tinqambato: Tinqambato, $4 \mathrm{~km} \mathrm{~W}, 23 . i i i .2000$, E. González, under stones, $1 \sigma^{\top}$ (AMCC [LP 1822]).
Centruroides margaritatus (Gervais, 1841): NICARAGUA: Granada: Domitila, $55 \mathrm{~m}, 27 . \mathrm{v}-1 . \mathrm{vi} .2002$, J.M. Maes, 1 ex. [legs] (AMCC [LP 1986]); Domitila Research Station (Domitila Reserva Silvestre Privada), $11^{\circ} 42.54^{\prime} \mathrm{N} 85^{\circ} 57.21^{\prime} \mathrm{W}$, 8.vi.2005,
C.S. Chaboo, 1 中 (AMCC [LP 4326]), 10.vi.2005, Chaboo and Shepard, 1 ठ (AMCC [LP 4325]).
Centruroides rileyi Sissom, 1995: MEXICO: San Luis Potosi: Município Axtlan de Terrazas: Axtlan de Terrazas, $21.42601^{\circ} \mathrm{N}$ $98.87821^{\circ}$ W, 100 m, 28.iv.2006, O.F. Francke, A. Valdez, G. Villegas and R. Paredes, 1 ठ (AMCC [LP 6445]).
Centruroides schmidti Sissom, 1995: HONDURAS: Dept. Francisco Morazán: Município San Antonio de Oriente: E.A.P. Zamorano, Monte Redondo, Acuacultura, $13.999^{\circ} \mathrm{N} 86.989^{\circ} \mathrm{W}, 773 \mathrm{~m}$, 23.ix.2008, C. Viquez, night UV, 8 juv. (AMCC [LP 9172]). Islas del Bahía: Município Roatán: Cayos Cochinos, Cayos Menor, forest trails, $15^{\circ} 57.448^{\prime} \mathrm{N} 86^{\circ} 30.055^{\prime} \mathrm{W}, 101 \mathrm{~m}$, 2.viii. 2012 , K. Sagastume and S. Longhorn, scrub oak forest, 1 \& (OUMNH); Isla Utila, Utila, wet savannah forest, $16^{\circ} 06.369^{\prime} \mathrm{N} 86^{\circ} 54.135^{\prime} \mathrm{W}$, 12 m, 21.vii.2012, K. Sagastume and S. Longhorn, scrub forest/ savannah, 1 \& (OUMNH).
Centruroides vittatus (Say, 1821): U.S.A.: Texas: Hays Co.: Buda, iii.2003, P. Sprouse, in house, 1 §, 1 \& (AMCC [LP 2286]).

Heteroctenus abudi (Armas \& Marcano Fondeur, 1987): DOMINICAN REPUBLIC: La Altagracia Prov: Parque Nacional del Este: Cabo Flaso (entrance zone), $18^{\circ} 22^{\prime 2} 25^{\prime \prime} \mathrm{N} 68^{\circ} 37^{\prime} 01^{\prime \prime} \mathrm{W}$, 67.7 m, 14.vii.2004, E.S. Volschenk and J. Huff, $1 \delta$ (AMNH); track between Ranger Station (at Boca de Yuma) and Punta Faustino, $18^{\circ} 21^{\prime} 17.2^{\prime \prime} \mathrm{N} 68^{\circ} 36^{\prime} 52.3^{\prime \prime} \mathrm{W}, 3.3 \mathrm{~m}, 14 . v i i .2004$, E.S. Volschenk and J. Huff, dense canopy humid forest, hand collected from under stones and blacklighting, especially along an old rock wall along the start of the track, $19 \jmath^{\lambda}, 15 \uparrow, 1$ subad. $\AA^{\lambda}$, 1 subad. +5 juv., $1021^{\text {st }}$ instars (AMNH), 1 juv. (AMCC [LP 3268]); San Rafael de Yuma, $18^{\circ} 21.332^{\prime} \mathrm{N} 68^{\circ} 37.095^{\prime} \mathrm{W}, 46 \mathrm{~m}$, 8.vi.2012, CarBio Team, rock wall, 1 o (AMCC [LP 12463]). U.S.A.: Puerto Rico: Isla Mona, trail \#1 to Punta Capitan from Sardiniera, $18^{\circ} 05.294^{\prime} \mathrm{N} 67^{\circ} 56.289^{\prime} \mathrm{W}, 16 . x .2009$, L. Esposito and H.Y. Yamaguti, blacklighting, primarily scrub forest and cactus, on rocks, sympatric with Centruroides and Cazierius, 10 §, 9 \&, 3 subad., 4 juv. (AMNH), 2 juv. (AMCC [LP 10235]); road to El Faro, $18^{\circ} 03.833^{\prime} \mathrm{N} 67^{\circ} 52.114^{\prime} \mathrm{W}$ to $18^{\circ} 05.126^{\prime} \mathrm{N}$ $67^{\circ} 50.871^{\prime}$ W, 17.x.2009, L. Esposito and H.Y. Yamaguti, blacklighting, population sparse, vegetation primarily desert grassland dominated by organ cactus and tall grasses, drier than east coast, 2 §̃, 4 Q, 1 subad., 1 juv. (AMNH), 1 juv. (AMCC [LP 10234]); main road at intersection to trail \#26 to Playa India, $18^{\circ} 03.806^{\prime} \mathrm{N}$ $67^{\circ} 53.239^{\prime}$ W, 18.x.2009, L. Esposito and H.Y. Yamaguti, $1 ठ^{\top}$ (AMNH).
Heteroctenus bonettii (Armas, 1999): DOMINICAN REPUBLIC: Pedernales Prov.: Parque National Jaragua: Cabo Rojo, $17^{\circ} 53^{\prime} 45.2^{\prime \prime} \mathrm{N}^{7} 71^{\circ} 39^{\prime} 35.8^{\prime \prime} \mathrm{W}, 15 \mathrm{~m}$, 9.vii.2004, E.S. Volschenk and J. Huff, dry cactus and spiny forest on limestone karst, hand collected from under stones and logs, and with blacklights, 5 § , 11 Q, 5 subad., 2 juv. (AMNH), 1 juv. ${ }^{\top}$ (AMCC [LP 3267]); road to Cabo Rojo, 0.6 km S of DR $44,17^{\circ} 58.201^{\prime} \mathrm{N} 71^{\circ} 39.036^{\prime} \mathrm{W}$, 14 m, 7.vii.2010, J. Huff and S. Schoenbrun, karst limestone,
 road to Fondo Paradi， 1.8 km from Highway $44,17^{\circ} 48.692^{\prime} \mathrm{N}$ $71^{\circ} 26.600^{\prime} \mathrm{W}, 302 \mathrm{ft}, 12.1 .2004$ ，J．Huff，found between rocks， 1 \＆（AMCC［LP 2471］）， 1 q（AMCC［LP 3265］）；unmarked track into park between Manuell Goa and Oviedo， $17^{\circ} 48^{\prime} 41.5^{\prime \prime}$ $71^{\circ} 26^{\prime} 35.9^{\prime \prime}$ W， 83.3 m，9．vii．2004，E．S．Volschenk and J．Huff， deciduous forest and thorny scrub，hand collected from under stones and logs and with blacklights， 15 §， 8 \＆， 1 subad．， 1 juv． （AMNH）， 1 juv．［pedipalps］（AMCC［LP 3266］）．
Heteroctenus garridoi（Armas，1974）：CUBA：Guantanamo Prov．：Guantanamo，x．2009，ex G．Molisani， 1 \＆（AMCC［LP 10225］）．U．S．Guantanamo Bay Naval Base：Guantanamo Bay， Graffiti Hill， $19^{\circ} 55^{\prime} 00.48^{\prime \prime} \mathrm{N} 75^{\circ} 06^{\prime} 08.64^{\prime \prime}$ W，7．v．2010，P．Tol－ son，S．Droege and S．Brady，native scrub， 1 §（AMNH）．
Heteroctenus junceus（Herbst，1800）：Antillen？， 1 ठ， 2 \＆（ZMB 7370）．Porto Rico［erroneous］，Stahl， 2 \＆， 1 juv．（ZMB 7280）． Santiago de las Caballeros［Dominican Republic，erroneous］， 1936，P．Thumb， 1 §（ZMH）．CUBA：Gundlach， 1 §， 1 q（ZMB 738）， 2 ㅇ（ZMB 2637）， 1 juv．（ZMB 7343）．vii．2007，C．Ham－ ilton， 1 juv．（AMCC［LP 7009］）．Pinar del Río Prov．：Sierra del Rosario，near Aspuru，xi．1937，H．H．Voelckers， 1 ठ， 1 q， 1 juv．中（ZMH）．Artemisa Prov．：near Baños［probably Santiago del los Baños］，v．1918， 2 ô（AMNH）．Camagüey Prov．：Sierra de Cubitas，Limones－Tuabaquey Ecological Reserve，area around field station， $21^{\circ} 32.887^{\prime} \mathrm{N} 77^{\circ} 46.705^{\prime} \mathrm{W}, 55 \mathrm{~m}, 13-14 . \mathrm{iv} .2012$ ， CarBio Team， 1 ठ（AMCC［LP 12619］）， 1 §（AMCC［LP 12620］）， 1 subad．$q$（AMCC［LP 12621］）， 1 subad．$q$（AMCC ［LP 12622］）， 1 juv．đ̋（AMCC［LP 12623］）．Guantanamo Prov．： Alejandro Humboldt National Park：near El Yunque de Bara－ coa， $20^{\circ} 19.907^{\prime} \mathrm{N} 74^{\circ} 34.151^{\prime} \mathrm{W}$ ， 74 m ，4．iv．2012，CarBio Team， 1 q， 26 juv．（AMCC［LP 12432］）， $20^{\circ} 20.701^{\prime} \mathrm{N} 74^{\circ} 33.985^{\prime} \mathrm{W}$ ， $370 \mathrm{~m}, 5 . \mathrm{iv} 2012$ ，CarBio Team， 1 ô（AMCC［LP 12613］）， $20^{\circ} 19.64^{\prime} \mathrm{N}^{\mathrm{N}} 74^{\circ} 35.59^{\prime} \mathrm{W}, 530 \mathrm{~m}, 6-7 . \mathrm{iv} .2012$ ，CarBio Team， 1 ふ， 2 ㅇ（AMNH）， 1 ठ（AMCC［LP 12614］）， 4 \＆（AMCC［LP 12615－12618］）．Havana Prov．：Havana， 1 q（AMNH），iv．1941， E．Weiss， 1 q， 1 subad．（AMNH）．Holguin Prov．：viii．2000， Heist，captive bred， 1 juv．（AMCC［LP 1928］）；Guardalavaca， 29．iii．1993，W．Altmann，captive bred， $1 \precsim$（AMCC［LP 1565］）； Mayari，Parque Nacional＂Mensura－Piloto＂， 1 km después de la carretera al Hotel＂Mayari＂， 716 m，10．v．2013，F．Cala－Riquelme and A．Deler－Hernández，secondary riverine forest with abun－ dance of pines，collected in the evening and at night on vegeta－ tion and trunks， 1 juv．$\widehat{\jmath}$（AMCC［LP 12896］）．Isla de la Juven－ tud Prov：Isle of Pines， 1 đ（AMNH）．Mayabeque Prov．：Arroyo Bermejo，near Fibacoa［Jibacoa］，15．vi．1967， 1 ¢（ZMB 31021）， vi．1967， 1 juv．（ZMB 31022），31．v．1967，Kleiderschrank， 1 § （ZMB 31020）；Guisa，mountains near，x．1936，P．Thumb， 1 ㅇ， 28 juv．（ZMH），Moa，ix．1937，P．Thumb， 1 §（ZMH）．Ori－ ente Prov：1938，P．Thumb， 4 ¢（ZMH）．Pinar del Río Prov：： Guanahacabiles，Akad．－stat．El Beral，xii．1967，G．Peters， 1 subad．（ZMB 31023）；Sierra de Anafe，23．ii．194，M．Barro， 2 subad．（AMNH）；Viñales National Park，near Dos Hermanas， $22^{\circ} 37.265^{\prime} \mathrm{N} 83^{\circ} 44.3^{\prime} \mathrm{W}, 130 \mathrm{~m}, 18 . \mathrm{iv} .2012$ ，CarBio Team， 2 juv． ふ（AMCC［LP 12624，12625］）， 1 juv．+ （AMCC［LP 12626］）， $22^{\circ} 39.424^{\prime} \mathrm{N} 83^{\circ} 42.097^{\prime} \mathrm{W}, 280 \mathrm{~m}, 20-21$. iv．2012，CarBio Team， 6 juv．đ（AMCC［LP 12627－12632］）， 1 juv．of（AMCC ［LP 12633］）；Vinales Valley，1940，Osorio， 1 q（AMNH）．Sancti Spíritus Prov．：Trinidad，viii．1978，B．Acosta， 1 §（AMNH［AH 4514］）．Santiago de Cuba Prov．：La Socapa， 10 km SW of San－ tiago de Cuba，9．iv．1999，R．Teruel， 1 §（AMNH）， 3 ¢（AMCC ［LP 1509，1517，1518］）；Santiago de Cuba， 1 ठ， 2 juv．（AMNH）．
Heteroctenus princeps（Karsch，1879）：DOMINICAN REPUB－ LIC：Independencia Prov．：Parque Nacional Isla Cabritos：Isla Cabritos， $18^{\circ} 30.019^{\prime} \mathrm{N} 71^{\circ} 43.228^{\prime} \mathrm{W}, 110 \mathrm{ft}, 7.1 .2004$ ，J．Huff， under rock，coral， 6 §， 4 q， 3 subad．， 17 juv．（AMNH）， 3 juv． （AMCC［LP 2470］）， 1 subad．， 2 juv．（AMCC［LP 3260］）；Rang－ er station， $18^{\circ} 33^{\prime} 45^{\prime \prime} \mathrm{N} 71^{\circ} 41^{\prime} 50^{\prime \prime} \mathrm{W}$ ，-19 m ，8．vii．2004，E．S． Volschenk and J．Huff，dry forest，hand collected from under stones and logs，and with blacklights， 3 §， 7 ¢， 6 subad．， 2 juv． （AMNH）， 1 subad．（AMCC［LP 3264］）；behind Ranger Sta－
tion， $18.56287^{\circ} \mathrm{N} 71.69762^{\circ} \mathrm{W}$ ，-23 m ，8．viii． 2005 ，L．Esposito， mixed dry forest with succulents，UV detection， $35^{\circ} \mathrm{C}, 3 \widehat{\delta}, 8$ ， 2 subad．$q, 321^{\text {st }}$ instars（AMNH）， $1 \delta$（AMCC［LP 5102］）；park entrance to Lago Enriquillo， $18^{\circ} 33.772^{\prime} \mathrm{N} 71^{\circ} 41.859^{\prime} \mathrm{W}, 18 \mathrm{~m}$ ， 21．ii．2012，J．Huff and R．C．West， 1 juv．§（AMCC［LP 12102］）． Parque Nacional Sierra de Bahoruco：road between Rabo de Gato and Duverge， $18^{\circ} 19^{\prime} 38^{\prime \prime} \mathrm{N} 71^{\circ} 33^{\prime} 55^{\prime \prime} \mathrm{W}, 447 \mathrm{~m}$ ，7．vii．2004， E．S．Volschenk and J．Huff，arid thorny scrub，hand collected from under stones and in dead and dry agaves， 3 §， 3 ， 4 juv．（AMNH）， 1 \＆（AMCC［LP 3263］）；Puerto Escondido，Si－ erra de Bahoruco， $18^{\circ} 19.762^{\prime} \mathrm{N} 71^{\circ} 33.502^{\prime} \mathrm{W}, 1592 \mathrm{ft}, 6 . \mathrm{i} .2004$ ， J．Huff，under dead agave， 1 §， 3 ค， 1 juv．（AMNH）， 1 juv． （AMCC［LP 3261］）；Puerto Escondido， 6 km NNE， $18^{\circ} 21.084^{\prime} \mathrm{N}$ $71^{\circ} 32.048^{\prime}$ W， 240 m，6．vii．2010，J．Huff and S．Schoenbrun， 1 subad．©（AMCC［LP 10523］）；Road to Puerto Escondido， $18^{\circ} 20.376^{\prime}$ N $71^{\circ} 33.345^{\prime}$ W， 1388 ft ，6．i．2004，J．Huff，under rocks in gravel quarry， 1 \＆（AMNH）， 1 juv．（AMCC［LP 3262］）．La Al－ tagracia Prov：：San Rafael，El Morro Monte Cristi， $19^{\circ} 47.3^{\prime} \mathrm{N}$ $70^{\circ} 43.02^{\prime} \mathrm{W}, 40 \mathrm{~m}, 22 . v i .2012$ ，CarBio Team， 1 \＆（AMCC［LP 12479］）， 1 subad．§（AMCC［LP 12478］）．Pedernales Prov： Manuel Goja，3．9．km N， $17^{\circ} 50^{\prime} 20.81^{\prime \prime} \mathrm{N} 71^{\circ} 27^{\prime} 18.84^{\prime \prime} \mathrm{W}$ ， 9．v．1998，D．Huber， 1 §（AMCC［LP 1566］）；Oviedo to Peder－ nales， $11.5 \mathrm{~km} \mathrm{~N}, 17^{\circ} 56^{\prime} 18.69^{\prime \prime} \mathrm{N} 71^{\circ} 32^{\prime} 37.25^{\prime \prime} \mathrm{W}, 8 . \mathrm{v} .1998$ ，D． Huber， 1 §（AMCC［LP 1516］）．HAITI：Dept．Ouest：Port－au－ Prince， $18^{\circ} 32^{\prime} 21.15^{\prime \prime} \mathrm{N} 72^{\circ} 19^{\prime} 44.39^{\prime \prime} \mathrm{W}$ ，Ehrenberg，holotype $\delta^{\lambda}$ （ZMB 116）．
Ischnotelson guanambiensis（Lenarducci et al．，2005）：BRA－ ZIL：Bahía：Ceraíma：Guanambi，Aeroporto de Guanambi， $14^{\circ} 13^{\prime} 00^{\prime \prime} \mathrm{S} 42^{\circ} 46^{\prime} 60^{\prime \prime}$ W，17．xii．2007，H．Y．Yamaguti et al．， 1 subad．+ （MZSP 30864），［leg］（AMCC［LP 9669］）， 1 subad．§ （MZSP 30865），［leg］（AMCC［LP 9670］）．
Ischnotelson peruassu Esposito et al．，2017：BRAZIL：Mi－ nas Gerais：Januária：Parque Nacional Cavernas do Peruaçu， $15^{\circ} 07.43^{\prime} \mathrm{N} 44^{\circ} 14.467^{\prime} \mathrm{W}, 4-25 . \mathrm{i} .2009$ ，R．S．Recoder and M． Teixeira－Júnior，paratype ơ（MZSP 31138），［leg］（AMCC［LP 9937］）．
Isometrus maculatus（DeGeer，1778）：SRI LANKA：Wellawaya， 24．ii．2000，D．Huber， 1 §， 1 ¢（AMCC［LP 1798］）．
Jaguajir agamemnon（C．L．Koch，1839）：BRAZIL：Mara－ nhão：Balsas， $07^{\circ} 28^{\prime} 44^{\prime \prime}$ S $46^{\circ} 07^{\prime} 09^{\prime \prime} \mathrm{W}$ ，4．vi．2008，H．Y．Yama－ guti et al．，1 \＆（MZSP 31132），［leg］（AMCC［LP 9948］ex）， 1 juv．ふِ（MZSP 31133），［leg］（AMCC［LP 9932］）；Caxias， $04^{\circ} 56^{\prime} 26^{\prime \prime} \mathrm{S}^{\prime} 43^{\circ} 27^{\prime} 59^{\prime \prime} \mathrm{W}, 16 . v i i i .2008$ ，R．Pinto－da－Rocha et al．， 1 juv．§（MZSP 30883），［leg］AMCC［LP 9692］），04ํ56＇50＂S $43^{\circ} 29^{\prime} 45^{\prime \prime} \mathrm{W}, 15 . v i i i .2008$ ，R．Pinto－da－Rocha et al．， 1 juv．${ }^{\text {º }}$ （MZSP 31134），［leg］（AMCC［LP 9936］）；Santa Barbara，on shore of Rio Parnaiba，vi．1962，G．Eiten， 1 ठ（AMNH）．Pernam－ buco：Exu， 10 km N，13．iii．1977，L．J．Vitt，rocky habitat within thorn scrub forest， 1 \＆， 1 subad．$\uparrow$ ， 4 juv．（AMNH），14．iii．1977， L．J．Vitt，rocky habitat in thorn scrub， 1 §， 1 q（AMNH）；Exu， 10 km NE，28．iv．1977，L．J．Vitt， 1 §， 1 t， 2 subad． ， 2 sub－ ad．， 1 juv．（AMNH），25．ix．1977，L．J．Vitt， 1 §̃， 1 ¢（AMNH）； Exu， 15 km NE，14．v．1977，L．J．Vitt，high caatinga，under bark of tree， 1 subad．$\circ$（AMNH）；Exu， 20 km E，30．iii．1977，L．J． Vitt， 1 juv．ô（AMNH）；Fazenda Caterino， 10 km NE Exu， 1．viii．1977，L．J．Vitt， 1 juv．ठ（AMNH），25．ix．1977，L．J．Vitt， 1 ㅇ（AMNH），9．vii．1977，L．J．Vitt， 1 subad．đ̄（AMNH）．Pi－ auí：Barras， $04^{\circ} 19^{\prime} 04^{\prime \prime} \mathrm{S} 42^{\circ} 18^{\prime} 26^{\prime \prime} \mathrm{W}$ ，18．viii．2008，R．Pinto－ da－Rocha et al．， 1 subad．+ （MZSP 30884），［leg］（AMCC［LP 9693］）；Castelo do Piauí， $05^{\circ} 13^{\prime} 43^{\prime \prime} \mathrm{S} 41^{\circ} 41^{\prime} 57^{\prime \prime} \mathrm{W}$ ，13．viii．2008， R．Pinto－da－Rocha et al．， 1 \＆（MZSP 30887），［leg］（AMCC ［LP 9696］）， 1 subad．ふ（MZSP 31157），［leg］（AMCC［LP 9933］）， 1 juv．đ（MZSP 31156），［leg］（AMCC［LP 9939］）；Oei－ ras， $06^{\circ} 58^{\prime} 28^{\prime \prime} \mathrm{S} 42^{\circ} 06^{\prime} 31^{\prime \prime} \mathrm{W}, 2-3 . i v .2008$ ，H．Y．Yamaguti et al．， 1 o（MZSP 31160），［leg］（AMCC［LP 9954］）， 1 of（MZSP 31161），［leg］（AMCC［LP 9942］）；Piracuruca，near Parque Na－ cional Sete Cidades， $04^{\circ} 10^{\prime} 07^{\prime \prime} \mathrm{S} 41^{\circ} 41^{\prime} 56.7^{\prime \prime} \mathrm{W}$ ，16．viii．2008，R． Pinto－da－Rocha and L．S．Carvalho， 1 o（MZSP 31167），［leg］ （AMCC［LP 9949］）；Sítio Ouro Verde，Teresina，0454＇13．9＂S $42^{\circ} 47^{\prime} 27.1^{\prime \prime} \mathrm{W}, 27 . \mathrm{vii} .2008$ ，L．S．Carvalho， 1 subad．§（MZSP
30886), [leg] (AMCC [LP 9695]), 1 juv. § (MZSP 31169), [leg] (AMCC [LP 9960]); Teresina, Campus UFPI, $05^{\circ} 02^{\prime} 43.5^{\prime \prime} \mathrm{S}$ 42 $46^{\prime} 13.4^{\prime \prime} \mathrm{W}, 16 . v i i i .2008$, UFPI, 1 đ (MZSP 31170), [leg] (AMCC [LP 9929]). Tocantins: Mateiros, Jalapão, $10^{\circ} 33.811^{\prime}$ S $46^{\circ} 27.409^{\prime} \mathrm{W}, 17 . i v .2009$, S. Outeda-Jorge and F. Marques, 1 ठ (MZSP 31182), [leg] (AMCC [LP 9938]), 1 subad. \& (MZSP 31180), [leg] (AMCC [LP 9934]), 1 juv. § (MZSP 31181), [leg] (AMCC [LP 9958]); Rio da Conceição, estrada para E.E. Serra Geral, $11^{\circ} 22^{\prime} 26^{\prime \prime}$ S $46^{\circ} 49^{\prime} 11^{\prime \prime}$ W, 7.vi.2008, H.Y. Yamaguti, M.B. da Silva and T.J. Porto, 1 q (MZSP 30885), [leg] (AMCC [LP 9694]), 1 subad. of (MZSP 31193), [leg] (AMCC [LP 9935]).
Jaguajir pintoi (Mello-Leitao, 1932): BRAZIL: Roraima: Normandia, 03 $53^{\prime} 44^{\prime \prime} \mathrm{N} 59^{\circ} 37^{\prime} 40^{\prime \prime} \mathrm{W}$, 14.xi.2008, H.Y. Yamaguti and R. Pinto-da-Rocha, 1 juv. ठ (MZSP 31176), [leg] (AMCC [LP 9928]), 1 juv. § (MZSP 31177), [leg] (AMCC [LP 9944]); Vila Tepequém, Amajari, $03^{\circ} 47^{\prime} 54^{\prime \prime} \mathrm{N} 61^{\circ} 43^{\prime} 08^{\prime \prime} \mathrm{W}$, 11.xi.2008, H.Y. Yamaguti and R. Pinto-da-Rocha, 1 Q (MZSP 30862), [leg] (AMCC [LP 9672]), $03^{\circ} 47^{\prime} 54^{\prime \prime} \mathrm{N} 61^{\circ} 44^{\prime} 57^{\prime \prime} \mathrm{W}, 17 . x i .2008$, H.Y. Yamaguti and R. Pinto-da-Rocha, 1 \& (MZSP 30863), [leg] (AMCC [LP 9671]). GUYANA: Upper Takutu-Upper Essequibo Region: Rupununi region, SW Guyana, near Venezuelan border, iii.2008, 1 juv. ठ (AMCC [LP 8278]), 24.ix.2008, imported L. Arden, 9 \&, 3 juv. (OUMNH 2009-001).

Jaguajir rochae (Borelli, 1910): BRAZIL: Bahía: Barra, Igarité, 5.vi.2008, 1 \& (MZSP 31122), [leg] (AMCC [LP 9963]); Catu, Ibiraba, 2.viii.2000, 1 \& (MZSP 31123), [leg] (AMCC [LP 9953]); Ceraíma, Guanambi, $14^{\circ} 13^{\prime} 00^{\prime \prime} \mathrm{S} 42^{\circ} 46^{\prime} 60^{\prime \prime} \mathrm{W}$, 10-17.xii.2007, H.Y. Yamaguti et al., 1 § (MZSP 30881), [leg] (AMCC [LP 9684]); Guanambí, $7 \mathrm{~km} \mathrm{~S}, 14^{\circ} 17^{\prime 5} 56^{\prime \prime} \mathrm{S}$ $42^{\circ} 47^{\prime} 2.2^{\prime \prime}$ W, $533 \mathrm{~m}, 24 . \mathrm{i} .2007$, C.I. Mattoni, R. Pinto-daRocha and H.Y. Yamaguti, UV sampling, modified savanna, cloudy and raining, 1 juv. (AMCC [LP 7638]); Guanambí, 16 km SE, $14^{\circ} 17^{\prime} 19^{\prime \prime} \mathrm{S} 42^{\circ} 41^{\prime} 31.1^{\prime \prime} \mathrm{W}, 559 \mathrm{~m}, 25 . \mathrm{i} .2007$, C.I. Mattoni, R. Pinto-da-Rocha and H.Y. Yamaguti, UV sampling and under leaf litter, banana plantation and surrounds, 1 juv. (AMCC [LP 7655]); Fazenda du Fabiano, 8 km NE Guanambí, $14^{\circ} 10^{\prime} 17.6^{\prime \prime} \mathrm{S} 42^{\circ} 43^{\prime} 56.4^{\prime \prime} \mathrm{W}, 539 \mathrm{~m}, 24 . i .2007$, C.I. Mattoni, R. Pinto-da-Rocha and H.Y. Yamaguti, under rocks, rocky hill and surrounds, open savanna modified, 1 \& (AMNH), 1 §, 2 juv. (AMCC [LP 7639]); Espadoado de Cima, Jeremoabo, $10^{\circ} 04^{\prime} \mathrm{S}$ $38^{\circ} 23^{\prime}$ W, 27.v.2008, H.Y. Yamaguti, T.J. Porto and M.B. da Silva, 1 subad. + (MZSP 30882), [leg] (AMCC [LP 9685]); Ibotirama, $12^{\circ} 10^{\prime} 34^{\prime \prime} \mathrm{S} 43^{\circ} 11^{\prime} 33^{\prime \prime} \mathrm{W}, 8$. .vi.2008, H.Y. Yamaguti, M.B. da Silva and T.J. Porto, 1 § (MZSP 31125), [leg] (AMCC [LP 9956]), 1 q (MZSP 31124), [leg] (AMCC [LP 9947]); Jeremoabo, $10^{\circ} 04^{\prime}$ S $38^{\circ} 23^{\prime}$ W, 25.v. 2008 , H.Y. Yamaguti, M.B. da Silva and T.J. Porto, 1 § (MZSP 31126), [leg] (AMCC [LP 9945]); Várzea do Poço, 19.x.2007, 1 q (MZSP 31127), [leg] (AMCC [LP 9926]). Minas Gerais: Janaúba, 11.xii.2007, H.Y. Yamaguti, S. Outeda-Jorge and C.A. Souza, 1 juv. $\AA^{\star}$ (MZSP 31136), [leg] (AMCC [LP 9957]); UNIMONTES, campus Janaúba, Janaúba, 11.xii.2007, H.Y. Yamaguti et al., 1 subad. đ (MZSP 30879), [leg] (AMCC [LP 9682]). Paraiba: Soledade, $07^{\circ} 02.118^{\prime} \mathrm{S}$ $36^{\circ} 27.311^{\prime} \mathrm{W}, 575 \mathrm{~m}, 16 . \mathrm{iii} .1999$, A. Kury and A. Giupponi, 3 ठ (AMCC [LP 1581, 1582, 1775]). Pernambuco: Escola Aquicola, Exu, 30.iii.1977, L.J. Vitt, caatinga, 1 ठ (AMNH), 27.vi.1977, L.J. Vitt, $1 ठ^{\top}$ (AMNH); Exu, $07^{\circ} 26^{\prime} 44^{\prime \prime}$ S $39^{\circ} 44^{\prime} 21^{\prime \prime}$ W, 1.vi.2008, H.Y. Yamaguti, M.B. da Silva and T.J. Porto, 1 q (MZSP 31144), [leg] (AMCC [LP 9955]), 1 juv. đ (MZSP 31143), [leg] (AMCC [LP 9951]); Exu, 18 km E, 5.iii.1977, L.J. Vitt, under leaf of granite on boulder, caatinga habitat, $2 \uparrow, 681^{\text {st }}$ instars (AMNH); Exu, 20 km E, 30.iii.1977, L.J. Vitt, 1 §, 1 \&, 2 juv. (AMNH); Exu, 3 km NW, 10.iii.1977, L.J. Vitt, 2 §, 1 q, 3 juv. (AMNH); Exu, 3 km W, 30.v.1977, L.J. Vitt, 2 ô, 4 甲, 4 juv. (AMNH), 1.vi.1977, L.J. Vitt, 1 \& (AMNH); Exu, 5 km E, 8.v.1977, L.J. Vitt, 1 juv. (AMNH); Exu, 5 km N, 6.iv.1977, L.J. Vitt, caatinga, 1 ठ, 1 juv. (AMNH), 18.i.1978, L.J. Vitt and K.E. Streilein, 1 juv. (AMNH); Exu, $6 \mathrm{~km} \mathrm{N}, \mathrm{15.iii.1977}, \mathrm{L.J}. \mathrm{Vitt}$, (cotton), under fallen logs, 1 q, 1 juv. đ (AMNH); Exu, 6 km NE, 16.iii.1977, L.J. Vitt, under rock on larger rock, caatinga
habitat, 1 Q, $491^{\text {st }}$ instars (AMNH); Fazenda Batente, 5 km NE Exu, 29.iii.1977, L.J. Vitt, 1 juv. (AMNH); Fazenda Caterino, 10 km NE Exu, 1.viii.1977, L.J. Vitt, 7 §, 3 q, 3 juv. (AMNH); Fazenda Chelonia, 8 km S Exu, 28.vii.1977, L.J. Vitt, 2 juv. (AMNH); Fazenda Guarani, 3 km N Exu, 14.vii.1977, L.J. Vitt, $1 \sigma^{\top}, 3$ \&, 1 subad., 3 juv. (AMNH); Fazenda Guarani, 5 km N Exu, 29.vii.1977, L.J. Vitt, 1 q, 3 juv. (AMNH), 19.ii.1978, L.J. Vitt, 1 \& (AMNH); Serra Talhada, $07^{\circ} 58^{\prime} 11^{\prime \prime} \mathrm{S} 38^{\circ} 19^{\prime} 16^{\prime \prime} \mathrm{W}$, 30.v.2008, H.Y. Yamaguti, M.B. da Silva and T.J. Porto, 1 q (MZSP 31146), [leg] (AMCC [LP 9943]), 1 q (MZSP 31147), [leg] (AMCC [LP 9930]); Vitória do Santo Antão, 0807’S $35^{\circ} 25^{\prime}$ W, 28.v.2008, H.Y. Yamaguti, M.B. da Silva and T.J. Porto, 1 \& (MZSP 31148), [leg] (AMCC [LP 9946]). Piauí: Castelo do Piauí, $05^{\circ} 13^{\prime} 43^{\prime \prime}$ S $41^{\circ} 41^{\prime} 57 \prime \prime$ W, 13.viii.2008, R. Pinto-daRocha and L.S. Carvalho, 1 § (MZSP 31150), [leg] (AMCC [LP 9961]), 1 中 (MZSP 31151), [leg] (AMCC [LP 9941]). Sergipe: near Genipapo, vii.1982, O.F. Francke, 1 §, 2 q (AMNH); UHE Xingó, Canindé de São Francisco, 22.viii.2007, Arnaldo Jr., 1 q (MZSP 30880), [leg] (AMCC [LP 9683]).
Physoctonus debilis (C.L. Koch, 1840): BRAZIL: Pernambuco: Exu, $18 \mathrm{~km} \mathrm{~N}, 5 . i i i .1977$, L.J. Vitt, under leaf of granite on boulder, caatinga habitat, 1 \& (AMNH); Exu, $5 \mathrm{~km} \mathrm{~N}, 4 . x .1977$, L.J. Vitt, 1 ( (AMNH), 18.i.1978, L.J. Vitt and K.E. Streilein, 1 q (AMNH); Fazenda Batente, 13 km E Exu, 10.xi.1977, L.J. Vitt and K.E. Streilein, 1 q (AMNH); Fazenda Caterino, 10 km NE Exu, 9.vii.1977, L.J. Vitt, 1 q (AMNH), 25.ix.1977, L.J. Vitt, 1 q (AMNH). Piaui: Castelo do Piauí, $05^{\circ} 13^{\prime} 43^{\prime \prime} \mathrm{S} 41^{\circ} 41^{\prime} 57^{\prime \prime} \mathrm{W}$, 13.viii.2008, R. Pinto-da-Rocha et al., 1 q (MZSP 30868), [leg] (AMCC [LP 9680]), 1 \& (MZSP 31158), [leg] (AMCC [LP 9931]); Oeiras, $06^{\circ} 5^{\prime} 28^{\prime \prime} \mathrm{S} 42^{\circ} 06^{\prime} 31^{\prime \prime} \mathrm{W}, 2-3 . v i .2008, ~ H . Y$. Yamaguti et al., $1 \not \subset$ (MZSP 31162), [leg] (AMCC [LP 9940]), 1 subad. ㅇ (MZSP 31164), [leg] (AMCC [LP 9962]), 3.vi.2008, H.Y. Yamaguti et al., 1 \& (MZSP 30866), [leg] (AMCC [LP 9678]); near Parque Nacional Sete Cidades, Brasileira e Piracuruca, $04^{\circ} 10^{\prime} 02^{\prime \prime} \mathrm{S} 41^{\circ} 41^{\prime} 56.7^{\prime \prime} \mathrm{W}, 16 . v i i i .2008$, R. Pinto-daRocha and L.S. Carvalho, 1 q (MZSP 30867), [leg] (AMCC [LP 9679]).
Physoctonus striatus Esposito et al., 2017: BRAZIL: Bahía: Xique-Xique, $10^{\circ} 49^{\prime} 60^{\prime \prime} \mathrm{S} 42^{\circ} 43^{\prime} 60^{\prime \prime} \mathrm{W}, 2 . x .2008$, T.J. Porto, paratype $\widehat{0}$ (MZSP 30869), [leg] (AMCC [LP 9681]), 3.x.2008, T.J. Porto, paratype $\begin{gathered} \\ \text { (MZSP 31128), [leg] (AMCC [LP 9950]). }\end{gathered}$

Rhopalurus caribensis Teruel \& Roncallo, 2008: COLOMBIA: Magdalena Dept.: Município Santa Marta: Bahia de Guairaca, Tayrona Park, 31.x.1985, H.-G. Muller, 1 \& (SMF 37027); Corregimiento de Bonda, Vereda Girocasaca, Finca Guaipi, $11^{\circ} 13^{\prime} 05.5^{\prime \prime} \mathrm{N} 74^{\circ} 06^{\prime} 14.3^{\prime \prime} \mathrm{W}, 173 \mathrm{~m}, 21-24 . v i i i .2014$, J.A. Moreno and W. Galvis, nocturnal, manual collection with UV light, 1 § (AMCC [LP 13167]), 1 subad. § (AMNH); Finca Las Delicias, 80 m, 17.v.2008, J.A. Noriega, 1 § (AMCC [LP 9341]); Pozo Colorado, 11 km W Santa Marta, 18-30.iv.1968, B. Malkin, 1 q, 1 subad., $191^{\text {st }}$ instars (AMNH); Puente de Los Clavos, 15 km E Pueblo Bello, Sierra Nevada de Santa Marta, 1500 m, 13.vi.1968, B. Malkin, 1 subad. đ̃ (AMNH); Santa Marta, 29.vi-31.vii.1966, 2 q (SMF 39120).
Rhopalurus laticauda Thorell, 1876: 2 \& (ZMB 14865). "Mexico", Dr v. Hubl, 1 đ (ZMB 14866). BRAZIL: F. Kummerow, 1 đ̃, 1 \& (ZMB 8226). Acre: Rio Branco, Amazonasgebiet, 1912, E. Ule, 1 juv. + (ZMB 14867). Pará: Alter do Chão, Santarém, $02^{\circ} 31^{\prime} 36^{\prime \prime} \mathrm{S} 54^{\circ} 54^{\prime} 19^{\prime \prime} \mathrm{W}, 28 . x .2008$, R. Pinto-daRocha and H.Y. Yamaguti, 1 § (MZSP 30870), [leg] (AMCC [LP 9686]), 1 subad. $\widehat{0}$, (MZSP 30872), 1 juv. (AMCC [LP 9688]), 1 juv. + (MZSP 30873), [leg] (AMCC [LP 9689]), 1 juv. $q$ (MZSP 30874), [leg] (AMCC [LP 9690]), 1 juv. (MZSP 30875), [leg] (AMCC [LP 9691]); Monte Alegre, 01056'32"S $54^{\circ} 08^{\prime} 13^{\prime \prime}$ W 31.x.2008, H.Y. Yamaguti and R. Pinto-da-Rocha, 1 \& (MZSP 30871), [leg] (AMCC [LP 9687]), 1 \& (MZSP 31141), [leg] (AMCC [LP 9924]). Roraima: Alto Alegre, $02^{\circ} 56^{\prime} 34^{\prime \prime} \mathrm{N} 61^{\circ} 03^{\prime} 09^{\prime \prime} \mathrm{W}, 10 . x i .2008$, H.Y. Yamaguti and R. Pinto-da-Rocha, 1 § (MZSP 31173), [leg] (AMCC [LP 9952]), 1 juv. đ̋ (MZSP 31172), [leg] (AMCC [LP 9927]); Bonfim,
$03^{\circ} 22^{\prime} 45^{\prime \prime} \mathrm{N} 59^{\circ} 49^{\prime} 18^{\prime \prime} \mathrm{W}, 13 . x i .2008$ ，H．Y．Yamaguti and R． Pinto－da－Rocha， 1 đ（MZSP 30878），［leg］（AMCC［LP 9677］）， 1 subad．đ（MZSP 31175），［leg］（AMCC［LP 9925］）；Mt．Rorai－ ma， $2 \widehat{J}^{\top}, 1$ q， 1 subad．（AMNH［29180］）；Mucajaí， $02^{\circ} 27^{\prime} 38^{\prime \prime} \mathrm{N}$ $60^{\circ} 54^{\prime} 24^{\prime \prime} \mathrm{W}$ ，12．xi．2008，H．Y．Yamaguti and R．Pinto－da－Rocha， 1 ふ（MZSP 30877），［leg］（AMCC［LP 9676］）；Normandia， $03^{\circ} 53^{\prime} 44^{\prime \prime} \mathrm{N} 59^{\circ} 37^{\prime} 40^{\prime \prime} \mathrm{W}, 14 . x i .2008$ ，H．Y．Yamaguti and R．Pin－ to－da－Rocha， 1 §（MZSP 31178），［leg］（AMCC［LP 9959］）， 1 q （MZSP 30876），［leg］（AMCC［LP 9675］）．COLOMBIA：Guavi－ are Dept．：San José del Guaviare，xii．1955，Meden， 1 \＆（SMF 39252）．VENEZUELA：Apure：Elorza， 10 km SW，road to San Felipe， $07^{\circ} 03.749^{\prime} \mathrm{N} 69^{\circ} 30.249^{\prime} \mathrm{W}, 89 \mathrm{~m}, 5 . \mathrm{x} .2008$ ，J．A．Ochoa
 （AMNH）， 1 juv．（AMCC［LP 9200］）．Aragua：Cagua， $10^{\circ} 11^{\prime} \mathrm{N}$ $67^{\circ} 27^{\prime}$ W，1903， 1 §， 1 ㅇ（SAM 6512）；Maracay，Fahrenholz， 1 §， 1 个， 1 subad．（SMF 8876／218）， 1 subad．đ（SMF 29208）； Parque Nacional Henri Pittier：Puerto Colombia，Cristo Mirador， $10^{\circ} 30.572^{\prime} \mathrm{N} 67^{\circ} 36.253^{\prime} \mathrm{W}, 1-10 \mathrm{~m}$ ，1．viii．2009，F．Rojas－Run－ jaic，A．Ferrer，L．Prendini and J．A．Ochoa，dry area，hill near to beach， 1 ふ̂， 3 中（AMNH）， 1 juv．（AMCC［LP 10046］）．Boli－ var：A．C．La Ceiba，between Puerto Ordaz and Ciudad Bolívar， $08^{\circ} 14.023^{\prime} \mathrm{N} 62^{\circ} 55.562^{\prime} \mathrm{W}, 102 \mathrm{~m}, 15 . v i i .2009$ ，A．Yepez，M． Blanco and J．A．Ochoa，llanos， $1 \delta^{\AA}$（AMNH）， 1 juv．（AMCC［LP 10047］）；Cedeño ca．Los Pijiguaos， $06^{\circ} 29.878^{\prime} \mathrm{N} 67^{\circ} 02.600^{\prime} \mathrm{W}$ ， 76 m，12．x．2008，J．A．Ochoa and S．E．Bazo Abreu，llanos， 2 ， 1 subad．of（AMNH）， 1 juv．（AMCC［LP 9237］）；Ciudad Bolí－ var，20．ii．1903， 2 q（ZMH），07º37．486＇N $64^{\circ} 05.924^{\prime} \mathrm{W}, 117 \mathrm{~m}$ ， 24．x．2008，J．A．Ochoa and S．E．Bazo Abreu，llanos， 1 §， 3 o， 1 subad．$\circ, 1$ juv．$\widehat{0}, 1$ ठ metasoma（AMNH）， 1 juv．（AMCC ［LP 9256］）；Comunidad Corosal，ca．Pijiguaos， $80 \mathrm{~m}, 25 . x .2008$ ， J．A．Ochoa，forest， 1 §， $4 \uparrow, 2$ subad．$q$（AMNH）， 1 subad． ㅇ（AMCC［LP 9253］）；Gran Sabana， 88 km，xi． 2005 －ii．2006， C．Siederman， 2 §（AMNH）；La Paragua，M．A．de Verde， $1 \circlearrowleft^{\AA}$ （AMNH）；Parque Nacional Canaima：Laguna Canaima，Isla Ana－ toly， $06^{\circ} 15.191^{\prime} \mathrm{N} 62^{\circ} 50.945^{\prime} \mathrm{W}, 395 \mathrm{~m}, 27 . v i i .2009$ ，L．Prendini and J．A．Ochoa，savanna and forest， $3 \circlearrowleft^{\top}, 2 q(\mathrm{AMNH}), 1$ juv． （AMCC［LP 10048］）；Upata，ii．1973，A．Bordes， 1 q（AMNH）． Distrito Federal：Caracas，iii．1999，C．Siederman， 2 ， $201^{\text {st }}$ instars（AMNH）．Guarico：between Calabozo and San Fernan－ do de Apure（about halfway），30．xi．1967，M．A．de Verde， 1 q （AMNH）；Hato Masaguarat， 45 km S Calabozo，7．iv．1978，Y． Lubin， 1 đ（AMNH）．Mérida：Mérida， 2 đ̃， 3 （ P （SF 5712／27）． Miranda：Guatire，15．xii．1975，M．A．Gonzalez－Sponga， 1 §， 1 of（NM 16431），29．iv．2004，R．C．West，under rocks，dry forest， 1 ठ（AMCC［LP 2845］）， 1 \＆（AMNH）；Hacienda Santa Rosa， 3 km N Guatire， $450 \mathrm{~m}, 10 . \mathrm{i} .1973$ ，M．A．Gonzalez－Sponga， 1 §， 1 \＆， 2 juv．（AMNH）．Nueva Esparta：Isla Margarita， N of Peninsula de Macanao， $11^{\circ} 02.618^{\prime} \mathrm{N} 64^{\circ} 21.542^{\prime} \mathrm{E}, 4 . \mathrm{ix} .2005$ ，S． Huber， 1 q（AMCC［LP 4221］）；probably Isla Margarita，2001， C．Siederman， 1 \＆（AMCC［LP 2462］）．
Rhopalurus ochoai Esposito et al．，2017：VENEZUELA：Cara－ bobo：Município Valencia：Valencia，29．xii．1904，F．Kumme－ row，paratype $q$（ZMB 31024），ix．1958，H．Ardelt， 2 o para－ types（ZMH），Valencia，Falcon Distr．，viii．1992，C．Siederman， paratype $\widehat{o}^{\top}$（AMNH）．Trujillo：Município Motatan：San Miguel， ca．Represa Agua Viva， $09^{\circ} 30.225^{\prime} \mathrm{N} 70^{\circ} 34.914^{\prime} \mathrm{W}, 195 \mathrm{~m}$ ， 23．ix．2008，J．A．Ochoa and S．E．Bazo Abreu，dry forest， $1 \delta^{\top}, 1$ q paratypes（AMNH）， 1 subad．+1 juv．+ paratypes（AMCC［LP 9199］）．Município Valera：Valera region，N，x．2005，S．E．Bazo Abreu，paratype $q$（AMCC［LP 5504］），paratype $q$（AMCC［LP 5505］）．Zulia：Município Jesus Enrique Lozada：San Agustín， $10^{\circ} 45.841^{\prime} \mathrm{N} 71^{\circ} 44.108^{\prime}, 44 \mathrm{~m}, 28 . i x .2008$ ，J．A．Ochoa and S．E． Bazo Abreu，dry forest，holotype $\widehat{J}^{\lambda}, 1 \widehat{\jmath}, 2$ q， 1 subad．q para－ types（AMNH）， 1 subad．$q$ paratype（AMCC［LP 9207］）．
Tityus atriventer Pocock，1897：GRENADA：St．Andrew Parish： Balthazar Estate， $12.124^{\circ} \mathrm{N} 61.660^{\circ} \mathrm{W}$ ，18．vii．2008，L．Esposito and D．Schiff，agricultural land，under stones， $1 \&$（AMCC［LP 9033］）．
Tityus bahiensis（Perty，1833）：ARGENTINA：Misiones Prov．： San Ignacio，road to Playas del Sol， $27^{\circ} 16^{\prime} 15^{\prime \prime} \mathrm{S} 55^{\circ} 44^{\prime} 35^{\prime \prime} \mathrm{W}$ ，
$60 \mathrm{~m}, 7 . \mathrm{ii} .2006$ ，C．I．Mattoni and D．Vrech，UV on paranaense forest，in base of tree， 1 \＆（AMCC［LP 5641］）．BRAZIL：São Paulo：São Paulo，13．vi．1893，J．Wiengreen， 1 §， 2 中（ZMH）．
Tityus clathratus C．L．Koch，1844：SURINAM：Paramaribo，
 AND TOBAGO：Trinidad：Gaspar Grande Island，7．vii．1999， L．Prendini，collected at night with UV light， 8 of（AMCC［LP 1567］）．
Tityus discrepans（Karsch，1879）：VENEZUELA：Distrito Fed－ eral：Caracas，xi．1998，M．A．Gonzalez－Sponga， 1 ， 1 juv． （AMCC［LP 1547］）．Miranda：Municipio Los Salias：San An－ tonio de los Altos，Quebrada Aguas Blancas， $10^{\circ} 22.088^{\prime} \mathrm{N}$ $66^{\circ} 58.033^{\prime} \mathrm{W}, 1391 \mathrm{~m}, 22 . v i i i .2009$ ，S．Foghin，F．Rojas－Runjaic and J．A．Ochoa，secondary forest， 1 ठ（AMCC［LP 10102］）．
Tityus kuryi Lourenço，1997：BRAZIL：Bahia：Mucugé：Mucugé， $10 \mathrm{~km} \mathrm{NE}, 12^{\circ} 57^{\prime} 30.7^{\prime \prime} \mathrm{S} 41^{\circ} 1^{\prime} 30.1^{\prime \prime} \mathrm{W}, 1163 \mathrm{~m}, 20 . \mathrm{i} .2007$ ，C．I． Mattoni，R．Pinto－da－Rocha and H．Y．Yamaguti，UV sampling， hills with open rocky savanna， 1 juv．（AMCC［LP 7659］）；road to Igatú，south access， $12^{\circ} 54^{\prime} 11.9^{\prime \prime} \mathrm{S} 41^{\circ} 18^{\prime} 26.2^{\prime \prime} \mathrm{W}, 900 \mathrm{~m}$ ， 21．i．2007，C．I．Mattoni，R．Pinto－da－Rocha and H．Y．Yamaguti， UV sampling，hills with open rocky savanna， 1 \＆（AMCC［LP 7656］）．
Tityus metuendus Pocock，1897：GUYANA：Essequibo River， $01^{\circ} 36^{\prime} 45.7^{\prime \prime} \mathrm{N} 58^{\circ} 38^{\prime} 14.6^{\prime \prime} \mathrm{W}, 240 \mathrm{~m}, 15 . v i i .1999$ ，M．Kuntner， primary forest，collected on tree trunk， $1 \precsim$（AMCC［LP 1546］）． PERU：Loreto：Rio Orosa，E of Iquitos，12．vi．2002，S．Gonzales， on tree trunk at night， 2 \＆（AMCC［LP 1983］）．
Tityus smithii Pocock，1893：ST．VINCENT AND THE GREN－ ADINES：Bequia：Old Road to Hope Beach， $13.006^{\circ} \mathrm{N}$ $61.221^{\circ}$ W，14．vii．2008，L．Esposito and D．Schiff，tropical for－ est，UV detection， 1 subad．$q$（AMCC［LP 9046］）；Cinnamon Garpoan， $13^{\circ} 01.322^{\prime} \mathrm{N} 61^{\circ} 14.126^{\prime} \mathrm{W}, 170 \mathrm{~m}, 5 . \mathrm{v} .2013$ ，CarBio Team，dry scrub， 1 §（AMCC［LP 12976］）；Princess Margaret Beach， $13^{\circ} 00.13^{\prime} \mathrm{N} 61^{\circ} 14.377^{\prime} \mathrm{W}, 20 \mathrm{~m}, 5 . \mathrm{v} .2013$ ，CarBio Team， coastal vegetation， 1 \＆（AMCC［LP 12977］）．
Tityus riverai Teruel \＆Sanchez，2009：U．S．A．：Puerto Rico：Mu－ nicipio Mayagüez：Zoo Dr．Juan A．Rivero，Barrio Miradero， xi．2008，W．Vargas，on avian warehouse，found dead， 1 §， 1 ㅇ（AMNH）；Mayagüez，Zoologico de Puerto Rico（Mayagüez Zoo）， $18^{\circ} 12.948^{\prime} \mathrm{N} 67^{\circ} 08.008^{\prime} \mathrm{W}, 30 \mathrm{~m}, 22 . x .2009$ ，L．Prendini， J．Huff and L．Figueroa，lowland tropical forest in zoological gardens，UV detection on warm，very humid，moonless night， taken on tree trunks and tree holes mostly near bird aviary，some several meters high， 1 subad．， 1 juv．（AMCC［LP 10202］）．
Troglorhopalurus lacrau（Lourenço \＆Pinto－da－Rocha，1997）： BRAZIL：Bahía：Itaeté：Trail between Caves＂Lapa do Bode＂ and＂Lapa Escondida＂， $12^{\circ} 56^{\prime} 9.1^{\prime \prime} \mathrm{S} 41^{\circ} 03^{\prime} 56.2^{\prime \prime} \mathrm{W}, 21 . i .2007$ ， C．I．Mattoni，R．Pinto－da－Rocha and H．Y．Yamaguti，under rocks， 2 甲（AMNH）， 1 subad．$\uparrow$ ， 4 juv．（AMCC［LP 7637］）； ＂Lapa do Bode＂，24．i．2007，C．I．Mattoni，R．Pinto－da－Rocha and H．Yamaguti， 1 subad．$q$（AMCC［LP 10211］）．
Troglorhopalurus translucidus Lourenço et al．，2004：BRAZIL： Bahía：Andaraí：Gruna Canal da Fumaça，GPS，J．E．Gallao，D． Schimonsky and M．E．Bichuette，date， 1 §（LES－4786）．Len－ çóis：Gruta do Lapão，Chapada Diamantina，Lençóis， $12^{\circ} 34^{\prime} 00^{\prime \prime} \mathrm{S}$ $41^{\circ} 22^{\prime} 60^{\prime \prime} \mathrm{W}, 20-29 . i .2009$ ，H．Y．Yamaguti et al．， $1 \$^{\text {®（MZSP），}}$ ［pedipalp and two legs］（AMCC［LP 9668］）．
Zabius birabeni Mello－Leitão，1938：ARGENTINA：La Pampa Prov．：Lihuel Calel，6．i．2003，A．A．Ojanguren－Affilastro， 1 q （AMCC［LP 2261］）．San Juan Prov．：plains near Caucete，base of Sierra de Pie de Palo， $31^{\circ} 42^{\prime} 37.5^{\prime \prime} \mathrm{S} 68^{\circ} 08^{\prime} 24.2^{\prime \prime} \mathrm{W}, 817 \mathrm{~m}$ ， 28．i．2005，C．I．Mattoni and A．A．Ojanguren－Affilastro，UV sam－ pling， 1 ex．［chela］（AMCC［LP 4251］）．
Zabius fuscus（Thorell，1876）：ARGENTINA：Córdoba Prov．： Capilla de Olaen，ca． 11 km W of Molinari， $31^{\circ} 09^{\prime} 44.46^{\prime \prime} \mathrm{S}$ $64^{\circ} 36^{\prime} 24.336^{\prime \prime} \mathrm{W}, 1096 \mathrm{~m}, 29 . x i i .2005$ ，C．I．Mattoni，A．Peretti， P．Carreras，M．Zerda and D．Vrech．，grassland with granitic rocks，under stones and UV， 2 juv．（AMCC［LP 5642］）；La Cum－ bre，iii．2000，K．Biondo， 1 §（AMCC［LP 1869］）；Puesto del Cura，1917，Lillo， 2 q（MNHN RS 3587）．

## 10. Appendix 3

Morphological characters and character states used in phylogenetic analysis of New World buthid subfamily Centruroidinae Kraus, 1955. Nomenclature follows Hjelle (1990) and Sissom (1990), except for carapace and metasomal carination (Vachon 1974; Prendini 2003b), tergite and pedipalp carination (Prendini 2001b), pedipalp trichobothria (VACHON 1974), ovariuterine anatomy (Volschenk et al. 2008), and book lung anatomy (Kamenz \& Prendini 2008).

## Carapace

1. Lateral ocular carina: present (0); absent (1).
2. Central lateral carina: present (0); absent (1).
3. Anterior central submedian carina: present (0); absent (1).
4. Posterior central submedian carina: present (0); absent (1).
5. Anterior and posterior central submedian carinae, fusion: separate (0); fused (1).
6. Central lateral carina and posterior central submedian carina, fusion: fused (0); separate (1).
7. Lateral ocular tubercle, macro-ocelli, count (Stоскwell 1989; Prendini 2000; Soleglad \& Fet 2003): three (0); two (1).
8. Lateral ocular tubercles, posterior micro-ocellus (Stockwell 1989; Prendini 2000; Soleglad \& Fet 2003): present (0); absent (1).

## Chelicerae

9. Fixed finger, dorsobasal setation: present (0); absent (1).
10. Median tubercles on the dorsal surface of the trunk, pattern of arrangement: transverse row (0); spread (1).

## Pedipalps

11. Patella, association between prodorsal and proventral carinae: converging (0); separate, clearly defined (1).
12. Chela dorsal accessory carina: granular (0); smooth (1).
13. Chela median carina: present (0); absent or obsolete (1).
14. Chela ventral accessory carina: present (0); absent or obsolete (1).
15. Chela ventrointernal carina: present (0); absent (1).
16. Chela internomedian carina: present (0); absent (1).
17. Chela dorsointernal carina: present (0); absent (1).
18. Femur retrolateral accessory carina: absent (0); present (1).
19. Chela fixed finger, median denticle row, primary subrows: 8 (0); 9 (1); 13 or more (2).
20. Chela fixed finger, median denticle row, prolateral accessory (supernumerary) granules (Soleglad \& Fet 2003): absent (0); present, well developed (1); present, weak and widely spaced (2).
21. Chela movable finger, median denticle row, number of primary subrows (Soleglad \& Fet 2003; Prendini 2004): 8 (0); 9 (1); 11 (2); $\geq 13$ (3).
22. Chela shape (male) (Prendini 2001b, 2004): incrassate (0); slender (1).
23. Chela shape (female) (Prendini 2001b): incrassate (0); slender (1).
24. Chela fixed finger, shape (male) (Prendini 2001b, 2004): straight, proximal dentate margin linear when fingers closed (0); slightly curved dorsally (1); strongly curved dorsally, proximal dentate margin distinctly emarginate when fingers closed (2).
25. Chela movable finger, shape (male) (Prendini 2001b, 2004): straight, proximal dentate margin linear when fingers closed (0); slightly curved ventrally (1); strongly curved ventrally, proximal dentate margin distinctly emarginate when fingers closed (2).
26. Chela movable finger, proximal lobe (male) (Prendin 2001b): absent (0); present (1).
27. Chela movable finger, median lobe (male) (Prendini 2004): absent (0); present (1).
28. Chela fixed finger, proximal lobe (male) (Prendini 2001b): absent (0); present (1).
29. Chela fixed finger, median lobe (male) (Prendini 2004): absent (0); present (1).
30. Femur dorsal surface, trichobothrium $d_{2}$ (Soleglad \& Fet 2001, 2003): absent (four $d$ trichobothria) (0); present (five $d$ trichobothria) (1).
31. Chela fixed finger, trichobothrium $d b$ position: situated distal to trichobothrium et (0); approximately aligned with et (1); situated between trichobothria est and et (2); approximately aligned with est (3).
32. Chela fixed finger, trichobothrium est position: approximately aligned with trichobothrium $d b \mathbf{( 0 )}$; situated between trichobothria $d b$ and et (1); situated proximal to et (2).
33. Chela fixed finger, trichobothrium et position: situated between trichobothria $d t$ and $d b$ (0); aligned with $d b$ (1); situated between $d b$ and est (2).

## Legs

34. Leg I, prolateral pedal spur: simple (0); bifurcating (1).
35. Leg I, telotarsal setae, arrangement: tufts (0); two discrete rows (1).
36. Leg I, telotarsal setae, form: fine, acuminate (0); thick, acuminate (1); short, stout (2).
37. Leg IV, telotarsal setae, arrangement: tufts (0); two discrete rows (1).
38. Leg IV, telotarsal setae, form: fine, acuminate (0); thickened acuminate (1); short, stout (2).

## Pectines

39. Pectinal teeth, shape: straight, sides almost parallel (0); sides slightly concurve, not straight (1); dorsal surface strongly concurve, sinuate (2).
40. Proximal pectinal teeth, dorsal surface, sculpture: smooth or slightly granular (0); with irregular striations (1); large and regular striations (2).
41. Proximal pectinal teeth, dorsal surface, nodules: single (0); multiple (1); absent (2).
42. Pectinal teeth, dorsobasal surface, macrosetae: present (0); absent (1).
43. Pectinal teeth, peg sensillae, shape: short and blunt (0); elongate and blunt (1); elongate and acuminate (2).
44. Proximal dorsal fulcra, setae: one (0); two (1); three (2); four (3); six or more (4); absent (5).
45. Proximal median lamellae, shape (female) (Prendini 2001b, 2004): not dilated (0); dilated (1).
46. Pectinal plate, anterior margin, sulcus: present (0); absent (1).
47. Pectinal plate, posterior margin (male): curved (0); straight (1).
48. Pectinal plate depressions (male): single median (0); two lateral (1); absent (2).

## Sternites

49. Sternite III, ventral median carinae: broad anterior raised region (0); narrow, elevated anterior carina (1); no elevation anteriorly (2).
50. Sternite III, lateral margins, sculpture: not raised or granular (0); granular carina (1); smooth carina (2).
51. Sternite III, ventrosubmedian surface, sculpture: smooth or slightly granular (0); large, regularly spaced granules (1); small, irregular granules (2). - Granules as characterized in state (1) are known to have a stridulatory function in all/some species where this state was observed.
52. Sternite V, raised, smooth area on posteromedian surface (male) (Prendini 2004): present (0); absent (1).
53. Sternite VI, ventrosubmedian carinae: present (0); absent (1).
54. Sternite VI, ventrolateral carinae: present (0); absent (1).
55. Sternites III-VI, spiracle shape (Kamenz \& Prendini 2008): wide, width $>5 \times$ length (0); compact, width $<3 \times$ length (1).

## Tergites

56. Tergite I, dorsolateral carinae: restricted to anterior 2/3 or less (0); absent (1).
57. Tergite I, dorsosubmedian carinae: present (0); absent (1).
58. Tergite II, dorsolateral carinae: restricted to anterior $2 / 3$ or less (0); absent (1).
59. Tergite II, dorsomedian carina: present (0); absent (1).
60. Tergites III-VI, dorsolateral carinae: present (0); absent (1).
61. Tergites III-VI, dorsosubmedian carinae (Prendini 2004): absent (0); present (1).
62. Tergite VII, dorsomedian carina: narrow, granular carina (0); granular mound, no carina (1); smooth mound, no carina (2).
63. Tergite VII, coloration relative to preceding tergites: paler (0); similar (1).

## Metasoma

64. Segment I, dorsal surface, granulation: sparsely granular (0); densely granular (1).
65. Segment II, lateral inframedian carina (Prendini 2004): continuous (0); posteriorly confined (1); absent (2).
66. Segment III, lateral inframedian carina (Prendini 2004): continuous (0); posteriorly confined (1); absent (2).
67. Segment III, dorsolateral carinae, posterior granules, size relative to anterior granules: similar (0); larger and spiniform (1).
68. Segment IV, lateral inframedian carinae: absent or obsolete (0); distinctly present and well developed (1).
69. Segments IV and V, ventral coloration relative to preceding segments: darker (0); similar (1).
70. Segment V, anal rim granulation: present (0); absent (1).
71. Segment V, dorsolateral carina: present (0); absent (1).
72. Segment V, lateral inframedian carinae: absent (0); present (1).
73. Segment V, ventromedian carina: absent (0); present (1).
74. Segment V, ventrosubmedian carinae: absent (0); present (1).
75. Segment V, ratio of length to width: slightly elongate, length $<2 \times$ width (0); elongate, length $2.5-3 \times$ width (1); strongly elongate, length $>3 \times$ width (2).
76. Segments I-IV, relative width (Lamoral 1978; PRENDIN 2001b, 2003b): narrowing posteriorly, segment I wider than IV (0); slight widening posteriorly, segment I slightly narrower than IV (1); pronounced widening posteriorly, segment I much narrower than IV (2).
77. Segments $I-V$, summed length relative to prosoma + mesosoma length (male): similar ( $<1.5 \times$ )(0); much greater $(\geq 1.5 \times)(\mathbf{1})$.
78. Segments I-V, coloration, dark ventromedian stripe: absent (0); present (1).

## Telson

79. Vesicle shape: spherical, length similar to width (0); slightly ovate, length ca. $1.5 \times$ width (1); ovate, length more than $2 \times$ width (2).
80. Vesicle width relative to width of metasomal segment V (Lamoral 1978; Prendini 2001b, 2003b): approximately equal (0); somewhat narrower (1); considerably narrower, less than half (2).
81. Vesicle ventromedian carina: present (0); absent (1).
82. Vesicle lateral surface, granulation: granular (0); smooth (1).
83. Vesicle subaculear tubercle (Lamoral 1980; Stockwell 1989; Prendini 2000, 2004; Soleglad \& Fet 2003): pronounced, pointed tooth (0); nub-like eminence (1); absent (2).
84. Vesicle subaculear tubercle, dorsal granules: absent (0); present (1).

## Size

85. Male body length, relative to female: smaller or approximately equal (0); much larger ( $>1.5 \times$ )(1).

## Ovariuterus

86. Ovariuterine network, number of loops (Volschenk et al. 2008): 8 (0); 9 (1); 2 (2).
87. Ovariuterus type (Volschenk et al. 2008): simple (0); complex bridged (1).

## Book lungs

88. Lamellar surface (Kamenz \& Prendini 2008): slender venation (0); ribbed venation (1).
89. Lamellar edge (Kamenz \& Prendini 2008): thorns (0); smooth or slightly wrinkled (1).
90. Posterior spiracle edge (Kamenz \& Prendini 2008): hillocks (0); subconical (1).

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