In limbo no longer: a new genus for a rare Amazonian skipper, *Telles pyrex* Evans, 1955 (Hesperiidae: Hesperiinae)

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Abstract: The male of the rare Amazonian skipper *Telles pyrex* Evans, 1955 is described and illustrated for the first time. This species has been without a generic affiliation, *incertae sedis*, since 2009. The generic placement of this species is investigated, considering characters from the male genitalia, wings, and DNA barcodes. *Pseudorphe* A. Warren & Dolibaina **gen. nov.** is proposed to accommodate this species, placed in the tribe Calpodini.

Key words: Amazon, Calpodini, genitalia, modified scales, monotypic

INTRODUCTION

Telles pyrex Evans, 1955 is a relatively large, showy skipper, described from a single female specimen from Amazonian Colombia. Evans (1955) described this species in the previously monotypic genus *Telles* Godman, 1900 based on its mostly yellow ventral hindwing coloration, and general similarities in the antennae and labial palpi to *Telles arcalaus* (Stoll, 1782). The type specimen of *T. pyrex* was figured by Warren *et al.* (2015), in dorsal and ventral aspect, together with images of its labels.

Burns *et al.* (2009) transferred *Telles arcalaus*, the type species of the genus, into *Thracides* Hübner, [1819], therefore rendering *Telles* a synonym of *Thracides*. Those authors reported on and figured a second female specimen of *T. pyrex*, from Amazonian Peru, and discussed its overall dissimilarity to *T. arcalaus*, noting that it clearly does not belong in *Thracides*. Burns *et al.* (2009) left *pyrex* without a genus-level association, *incertae sedis*, in "nomenclatural limbo", where it has remained for the past six years. Recently, without justification, Warren *et al.* (2015) listed *pyrex* as a species of *Orphe* Godman, 1901, due to its general superficial similarity in forewing hyaline markings to the two species of *Orphe*. This action was done merely for convenience, since Burns *et al.*'s (2009) designation as *incertae sedis* precluded its listing anywhere in the faunal list of American Hesperiidae.

Intensive surveys of the Lepidoptera fauna of central Rondônia, Brazil, during the early 1990s, resulted in the collection of one additional female of *T. pyrex* (Fig. 1C-D), as well as one male that was tentatively associated with *T. pyrex* (Fig. 1A-B). This male, while lacking the prominent yellowish ventral hindwing of the female of *T. pyrex*, shares the same rusty ventral ground color as the female, as well as details of the antennae and labial palpi. In addition, the position of hyaline forewing spots on the male closely corresponds to those of the female, although the male has smaller and fewer spots. The genitalia of this male specimen (Fig. 3) are not suggestive of any described genus of Hesperiinae.

In order to confirm the identity of this male specimen as the male of *T. pyrex*, and to glean insight into the genus-level placement for *pyrex*, we sequenced the "barcode region" of the cytochrome oxidase I gene, and compared the sequences to other taxa of American Hesperiinae within the tribe Calpodini (*sensu* Warren *et al.* 2008, 2009).

MATERIALS AND METHODS

Specimens examined in this study are deposited in the following collections: **ADW** (Andrew D. Warren personal collection, Castle Pines, Colorado, USA); **DZUP** (Padre Jesus Santiago Moure collection, Departamento de Zoologia, Universidade Federal do Paraná, Curitiba, Brazil); **MGCL** (McGuire Center for Lepidoptera and Biodiversity, Florida Museum of Natural History, University of Florida, Gainesville, Florida, USA); **NHMUK** (Natural History Museum, London, United Kingdom); and **OM** (Olaf Mielke personal collection, Curitiba, Paraná, Brazil). The abbreviation DZ corresponds to the code number on specimens in the DZUP collection. Additional abbreviations used are: FW–forewing; HW–hindwing; D–dorsal; V–ventral. Terminology used in the descriptions follows Carneiro *et al.* (2012, 2013).

In the "Type Material" section, different labels are identified using "/", and additional information is given between brackets. Genitalia of both sexes were prepared and illustrated using standard methods (Dolibaina *et al.* 2014), and dissected specimens are marked with an asterisk in the "Material Examined" section. The catalogue entry provided for *Pseudorphe pyrex* **comb. nov.** follows Mielke (2005), with updates, and the full bibliography is available in Lamas (2015).

A single leg was used to extract genomic DNA from each specimen with the Macherey-Nagel (MN) NucleoSpin® tissue kit. The details of experimental procedures were described in

Cong & Grishin (2014). The following pairs of primers we used to amplify the barcode in two overlapping segments: sCOIF (forward, 5'-ATTCAACCAATCATAAAGATATTGG-3'), Meg-mCOIR (reverse, 5'- CCAGTWCCTGYACCATTTTCTAC -3'), and Meg-mCOIF2 (forward, 5'-CCTCGWATAAATAAYATAAGATTTTG-3') – sCOIR (reverse, 5'-TAAACTTCTGGATGTCCAAAAAATCA-3').

Sequences and accompanying specimen data were submitted to GenBank and received accession numbers KU239104–KU239109. Additional DNA sequences were downloaded from GenBank <http://genbank.gov/>. Many of these sequences were reported in Janzen *et al.* (2011) and photos of specimens are available from the Area de Conservación Guanacaste (ACG) online database (Janzen & Hallwachs 2009). Sequences were aligned manually since they matched throughout their length without insertions or deletions. The Phylogeny.fr server at <http://www.phylogeny.fr/> was used with the Hamming distance model to compute evolutionary distances from aligned DNA sequences and build dendrograms (Dereeper *et al.* 2008).

RESULTS

Cytochrome Oxidase I DNA "barcodes" from a female and the putative male of *T. pyrex* were compared to those of sampled species of most other genera of Calpodini (Fig. 5). These confirm that the male tentatively associated with the females of *T. pyrex* is indeed the male of *T. pyrex*. In addition, these sequences, considered together with morphological characters from both sexes, demonstrate that while *T. pyrex* does appear to be a member of the tribe Calpodini (*sensu* Warren *et al.* 2009), it is apparently not congeneric with any of the included genera (see discussion). Therefore, we feel justified in describing a new genus for *T. pyrex*.

Pseudorphe A. Warren & Dolibaina, **gen. nov.**
(Figs. 1-4)

Type species. *Telles pyrex* Evans, 1955.

Diagnosis. *Pseudorphe* **gen. nov.** is distinguished from genera of Calpodini (*sensu* Warren *et al.* 2009) belonging to Evans' "O. *Calpodes* group" by the second segment of the labial palpus being quadrantic instead of flattened. This character is shared, however, with most of the genera of Calpodini belonging to Evans' "K. *Carystus* group". The presence of two almost circular patches of modified short and shining dark brown scales on the male, in the postdiscal region of CuA1-2A on the forewing and the submarginal region of Rs-CuA2 on the hindwing (Figs 1A, 2A-B), is unique among Calpodini, as is the well developed, tripartite, distal apophysis of the tegumen, and the distinctly long anterior projection of saccus and aedeagus (Fig. 3).

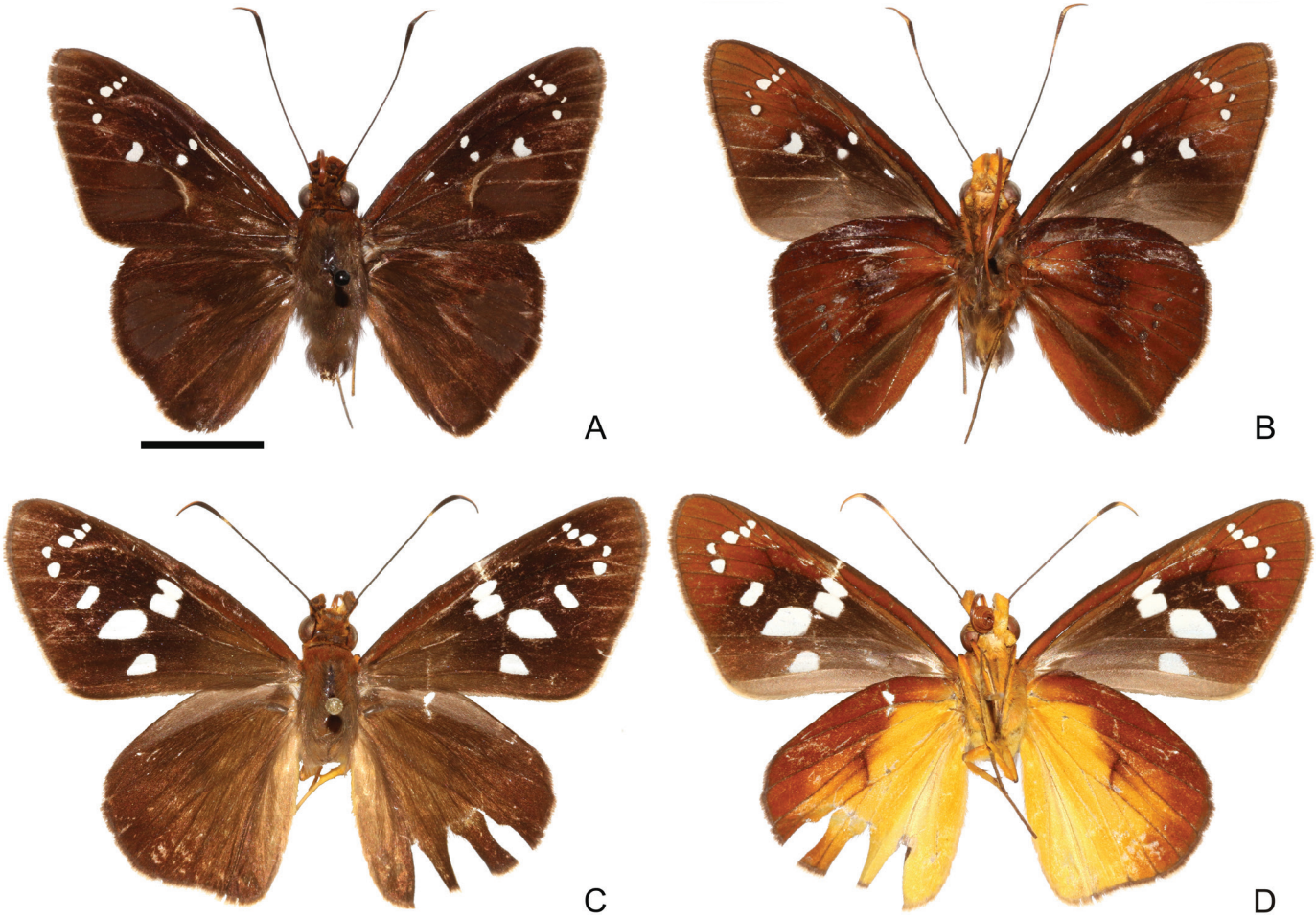

Fig. 1. Adults of *Pseudorphe pyrex*: A-B) male in dorsal and ventral views; C-D) female in dorsal and ventral views. Scale bar = 1 cm.

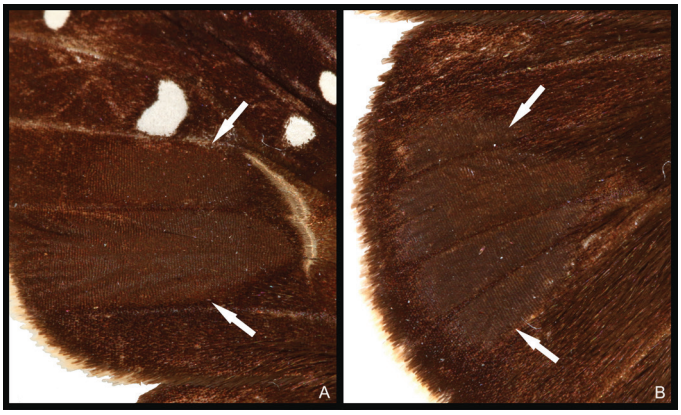

Fig. 2. Modified wings scales of male *Pseudorphe pyrex*: A) left forewing; B) left hindwing. White arrows delimit the areas with modified scales. Scale bar = 1 cm.

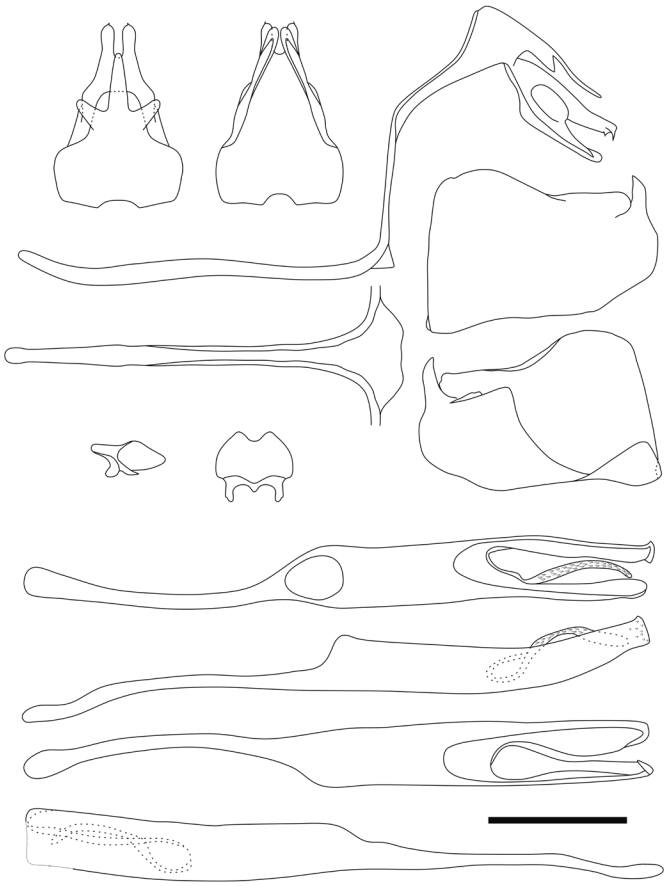

Fig. 3. Male genitalia of *Pseudorphe pyrex*: tegumen and uncus in dorsal and ventral views; tegumen, uncus, gnathos, ventral tegumen arm, dorsal projection of saccus and anterior projection of saccus in left lateral view; anterior projection of saccus in ventral view; external and internal views of the left valva; fultura inferior in left lateral and dorsal views; aedeagus in dorsal, left lateral, ventral and right lateral views. Scale bar = 1 mm.

Description. HEAD: Eyes rufous. Labial palpi with second segment quadrantic, third segment short and conical. Antennae reaching upper end of forewing distal cell; club slender, about 1/5 length of shaft, apiculus slightly longer than club; nudum reddish, with 17 segments, apparently all on apiculus.

THORAX: Mesotibia smooth, with a distal pair of spurs. Metatibia smooth, with two pairs of spurs, proximal pair at midlength of metatibia, inner spur shorter than outer, distal pair at distal margin of metatibia, more developed than proximal pair, inner spur longer than outer.

Forewing discal cell long, 3/4 forewing length, upper half of discal cell distally more projected than lower half; origin of M2 nearer to M3 than to M1; distance between origins of CuA2 and CuA1 longer than distance between origins of CuA1 and M3; male DFW with a transverse, thin, light brown and somewhat disjunct stigma in CuA1-2A, extending a short distance below anal fold; a wide and almost circular patch of modified short and shining dark brown scales in CuA1-2A, longer in CuA2-2A, distally bordering the stigma.

Hindwing with origin of M2 halfway between origins of M1 and M3; M2 poorly developed (represented by a fold); male DHW postdiscal area with a wide ovoid patch of modified short and shining dark brown scales from Rs to CuA2, longer in M1-M3.

MALE GENITALIA: Tegumen rectangular and short, with a long median, distal apophysis. Ventral tegumen arm narrow, shorter than dorsal projection of saccus. Anterior projection of saccus slender, two times combined length of tegumen and uncus. Uncus longer than wide, distal margin half the length of base, distal third bifid, with slender short, parallel arms, each with a pair of spiniform ventral projections at distal margin. Gnathos totally divided, arms slender and reaching end of uncus. Valva broad, narrowing distally, base width 3/4 that of length; sacculus triangular, widest portion at base then narrowing distally; narrow costa; ampulla with a short, rounded projection; harpe broad, with a disto-dorsal horn-shaped projection that exceeds the ampulla dorsally. Fultura inferior short, with a small ventral fold at base and two lateral projections at proximal margin. Aedeagus as long as the combined length of anterior projection of saccus and valva, coecum slender, about 3/7 length of the aedeagus; opening of ejaculatory bulb ovoid; distal portion with a broad dorso-ventral vesica opening, cornutus as a long, narrow strip covered by short thorns.

FEMALE GENITALIA: Sterigma broad, wider than long in ventral view. Lamella antevaginalis comprised of two lateral sclerotized plates fused medially in a narrow strip. Lamella postvaginalis with two lateral plates connected distally in a wide distal sclerotized margin, producing two large lobes ventrally, central area positioned above membranous ostium bursae. Ostium bursae at base of sterigma, with a short, weakly sclerotized dorsal portion. Ductus bursae as long as corpus bursae, with a short, sclerotized ring at the base of sterigma. Corpus bursae with two distinct broad areas, one near ductus bursae and other near terminal. Tergum VIII proximally fused with sterigma, distally broad, with an incomplete spiracular opening at its inferior margin. Papilla analis rectangular, distal half sclerotized, distal margin straight; apophysis posterior slender and short.

Etymology. The name is a reference to *Orphe* Godman, 1901 (Lepidoptera: Hesperiidae), a genus that includes two species with a forewing spot pattern similar to that of *P. pyrex*.

Pseudorphe pyrex (Evans, 1955), **comb. nov.**
(Figs. 1-4)

Telles pyrex Evans, 1955. **Cat. Amer. Hesp. 4**, p. 237; [holo]type male, IV-1932, Florida, Upper Putumayo, [Colombia], G. Klug leg.; BM(NH).- Bridges, 1983. **Lepid. Hesp. 1**, p. 99; **2**, p. 36.- Bridges, 1988. **Cat. Hesp. 1**, p. 157; **2**, p. 60.- Mielke, 2004. Hesperioidea, p. 81, *in* Lamas (ed.). **Checklist: Part 4A, Hesperioidea-Papilionoidea**, *in* Heppner (ed.). **Atlas Neotrop. Lepid. 5A**.- Mielke, 2005. **Cat. Amer. Hesperioidea 5**, p. 1300.- Burns *et al.*, 2009. **J. Lepid. Soc. 63**(3), p. 141, 142, 151, figs 39-40 (female d, v), 152, figs 41-42 (female gen.), 153.

(no genus) *pyrex*; Beattie, 1976. **Rhop. Direct.**, p. 243.- Burns *et al.*, 2009. **J. Lepid. Soc. 63**(3), p. 151, figs 39-40 (female d, v), 152, figs 41-42 (female gen.), 153.

Description. HEAD: uniformly brown, anterior chaetosema yellow, paraocular area and posterior chaetosema yellow-orange. Labial palpi uniformly yellow-orange, distal area of second segment and whole third segment brown. Antenna shaft dark brown, with a light-yellow ring at base of club, dorsally poorly defined; apiculus yellow ventrally.

THORAX: dorsally dark brown, ventrally rufous-brown to yellow. Tegulae and patagia rufous-brown.

Forewing length of male 22.9 mm (n=1); female 21.0, 24.8 mm (n=2); costal margin slightly convex; apex rounded and slightly projected; outer margin convex near apex then straight to tornus; tornus rounded; anal margin straight; both sexes with dark-brown DFW ground color, rufous from costal margin to Radius, extending to end of Sc; eight whitish hyaline spots, two discal, upper and lower, at mid-point of discal cell, separated (in male) or fused with each other forming a single wide spot (in female); one postdiscal in M3-CuA1 near origin of M3, shorter than wide; five small, subapical, circular spots from R3 to M3, first three aligned with each other and inclined to outer margin, last two aligned with each other in a perpendicular angle to wing, that in M1-M2 projected distally; female has two additional whitish hyaline postdiscal spots, one large in CuA1-CuA2 and another, smaller, in CuA2-2A below the anal fold, both aligned with the end of discal cell. DHW costal margin slightly convex; apex rounded and obtuse; outer margin slightly convex from apex to CuA1, then strongly concave; tornus rounded and projected; anal margin slightly convex; ground color uniformly dark-brown. VFW ground color dark brown, light gray from inferior half of CuA2-2A to anal margin, rufous-brown from costal margin to Radius extending to apex and to CuA1. Male VHW ground color rufous-brown, lighter near base, with a large, dark strip traversing the wing before end of discal cell, dark brown along 2A, four small, circular metallic gray spots in postdiscal area from M1 to CuA2. Female VFW ground color yellow, with a small rufous-brown area from base to apex then to submarginal area of CuA1-CuA2. Holotype female with two small, circular metallic gray spots in postdiscal area from M1 to M3, absent in other females examined. The remaining characters of this species are as presented above in the generic description.

Distribution. This species is known from just a few specimens collected in western Amazonia, from Colombia (holotype), Peru and Brazil.

Type Material. The holotype of *Telles pyrex* Evans, 1955 is deposited at the NHMUK with the following labels: / 178 / Type / Florida, R[iver]. Putumayo [Putumayo province, Colombia], April 1932. (G. Klug [leg.]). / Rothschild Bequest B.M.1939-1. / Photo AA / BMNH(E) #806133 /. The holotype was figured by Warren *et al.* (2015).

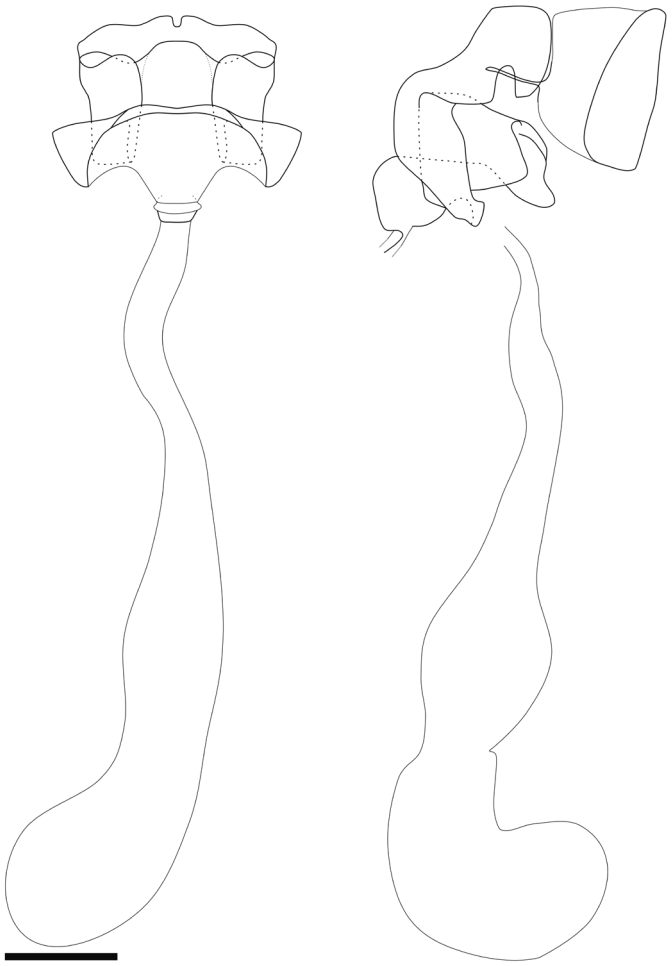

Fig. 4. Female genitalia of *Pseudorphe pyrex*: ventral and lateral views. Scale bar = 1 mm.

Material Examined. COLOMBIA – holotype female cited above. PERU – *Madre de Díos*: Puerto Maldonado (100 Km E), IX-XI-1992, Tello leg. 1 female (OM, figured by Burns *et al.* 2009). BRAZIL – *Rondônia*: Cacaulândia (Fazenda Rancho Grande), 17-VIII-1993, A. D. Warren leg. 1 male (ADW*), 21-VII-1994, G. T. Austin leg. 1 female (MGCL*); Vilhena, 13-XI-1986, C. Elias leg. 1 female (DZ 31.111).

DISCUSSION

Despite its apparently wide distribution in upper Amazonia, including parts of Colombia, Peru and Brazil, *Pseudorphe pyrex* is extraordinarily rare in collections. We've seen only five specimens (four females and one male), and two of these are from a well-surveyed site in Rondônia, Brazil. The single known male, collected by the senior author in Rondônia in 1993, was flushed from trailside vegetation, then settled a few meters away on the underside of a leaf, in the shaded understory of primary rain forest.

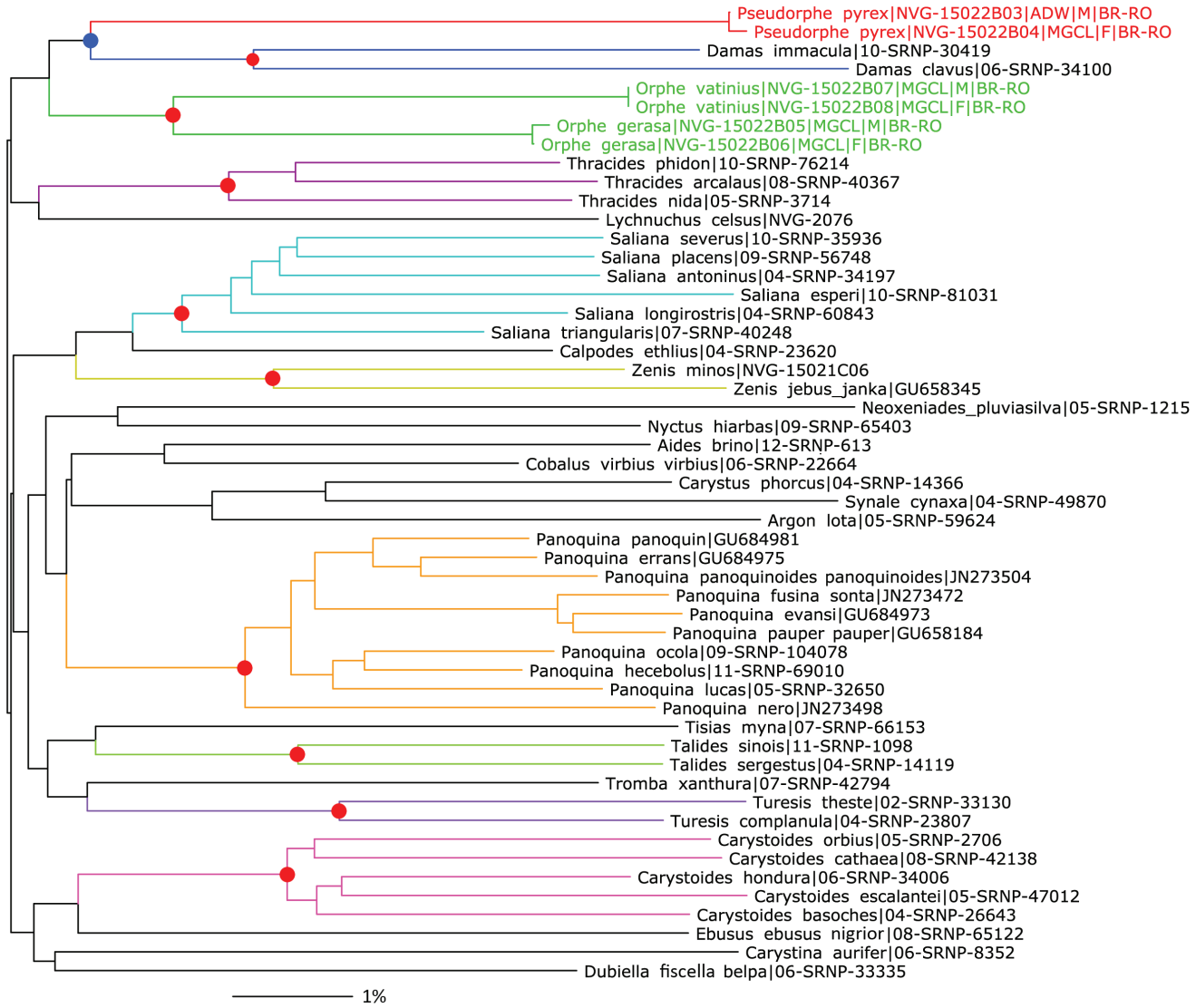

Fig. 5. Distance dendrogram of representative Calpodini. Each sequence is labeled by the species name and sequence ID. IDs with "NVG" are for sequences obtained in this work, "SRNP"-containing IDs are voucher codes from Janzen & Hallwachs (2009), others are GenBank accessions. Collection code, sex, and locality (RO = Rondônia, Brazil) are noted for *Pseudorphe* and *Orphe*. Branches leading to congeners are of the same color. Red dots mark the last common ancestor of included sequences in each genus represented by more than one sequence. The blue dot, marking the last common ancestor leading to *Pseudorphe*, is placed further away from the terminals than any red dot, suggesting that the placement of *pyrex* in a new genus is consistent with how other genera are currently defined in Calpodini.

The discovery of the male of *P. pyrex* has demonstrated significant sexual dimorphism in the species. The extensive forewing and hindwing patches of modified scales on the male are not seen elsewhere within Calpodini or Hesperiinae. Other characters that define *Pseudorphe* are found in the male genitalia, including the well developed, tripartite, distal apophysis of the tegumen, and the distinctly long anterior projections of saccus and aedeagus. In addition, sequence data from the "barcode" region of the COI gene indicate the uniqueness of *P. pyrex*; while *Pseudorphe* is clearly a member of Calpodini, its barcodes are quite distinct from those of apparently closely related genera *Damas* Godman, 1901 and *Orphe* (Fig. 5). The COI barcode difference between the two *Orphe* species is about 7%, which is about the largest divergence within a genus of Calpodini, but *P. pyrex* is different from either of these or *Damas* by 10-11%, a distance consistent with its assignment to a different genus.

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