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A new species of *Rhytidhysteron* (Ascomycota: Patellariaceae) from Colombia, with a provisional working key to known species in the world

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Abstract

We describe a new species, *Rhytidhysteron columbiense* Soto-Medina & Lücking, from Colombia. It is a saprobic species found on decaying branches in montane rain forest remnants, characterized by boat-shaped ascomata up to 3 mm long and 1.8 mm wide, with brown-black disc and yellowish-green pruina, perpendicularly striate margins, and by 3-septate, ellipsoid to fusiform, red-brown ascospores 38–52 × 13–18 µm in size. The new species differs from all other species in the genus by the yellowish-green pruina on the margins of the ascomata and from *R. rufulum* s.lat. also in the larger ascospores, whereas *R. brasiliense* has similarly long ascospores, but differs in their broader width and their pigmentation, as they remain light brown at maturity. A provisional working key to known species of the genus in the world is provided based on literature data. © 2017. Acad. Colomb. Cienc. Ex. Fis. Nat.

Key words: *Rhytidhysteron;* Ascospore; Pruina; Colombia.

Una nueva especie de *Rhytidhysteron* (Ascomycota:Patellariaceae) de Colombia, con una clave provisional para las especies conocidas en el mundo

Resumen

Se describe la nueva especie *Rhytidhysteron columbiense* Soto-Medina & Lücking para Colombia. Es una especie saprobia encontrada sobre ramas en descomposición en remanentes de bosque montano lluvioso; se caracteriza por ascomas en forma de canoa de cerca de 3 mm de longitud y 1,8 mm de ancho, con disco marrón negruzco y pruina verde amarillosa, márgenes estriados perpendicularmente, y tres esporas septadas, elipsoides a fusiformes, marrón rojizas, de 38–52 × 13–18 µm de tamaño. Esta nueva especie difiere de las otras del género por la pruina verde amarillosa sobre los márgenes del ascoma, y de *R. rufulum* s.lat. por las esporas más grandes, mientras que *R. brasiliense* tiene las esporas grandes, pero difiere por sus ascosporas más anchas y por su pigmentación, que es de color pardo claro en la madurez. Se agrega una clave provisional para las especies del género en el mundo con base en datos de la literatura. © 2017. Acad. Colomb. Cienc. Ex. Fis. Nat.

Palabras clave: *Rhytidhysteron;* Ascospora; Pruina; Colombia.

Introduction

Rhytidhysteron is a small genus of about 20 species typified by *R. brasiliense* Speg. (**Spegazzini**, 1881; **Silva-Hanlin & Hanlin**, 1999). The genus includes saprobic to weakly pathogenic fungi growing on woody plants, well-characterized by its rather large, conspicuous ascomata, which are usually elongate and boat-shaped and feature a prominent, perpendicularly striate margin, in combination with pigmented, sparsely septate to submuriform ascospores. The genus appears to have a subcosmopolitan distribution, with species described from all continents (**Spegazzini**, 1881; **Rosatto**, 1996; **Hsieh, *et al.***, 1997; **Tanaka & Hosoya**, 2006; **Murillo, *et al.***, 2009; **Méndez-Mayboca, *et al.***, 2010; **Almeida, *et al.***, 2014; **Yacharoen, *et al.***, 2015; **Doilom, *et al.***, 2016; **Thambugala, *et al.***, 2016). *Rhytidhysteron* has received attention for its secondary chemistry (**Murillo, *et al.***, 2009; **Pudhom & Teerawatananond**, 2014; **Pudhom, *et al.***, 2014; **Chokpaiboon, *et al.***, 2016) and the human-pathogenic properties detected in some species (**Spatafora, *et al.***, 1995; **Wipf, *et al.***, 2004; **Chowdhary, *et al.***, 2008; **Mahajan, *et al.***, 2014; **Mishra, *et al.***, 2014; **Chander, *et al.***, 2016).

The genus was revised by **Samuels & Müller** (1980), who reduced the 11 names listed in the genus at the time to only two species, namely *R. rufulum* (Spreng.) Speg. (with *R. brasiliense* as synonym) and *R. hysterinum* (Dufour) Samuels & E. Müll. However, it was later shown that

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their conclusions were based on superficial observations or ignorance of the character of the corresponding type specimens, including ascoma morphology and ascospore size (**Kutorga & Hawksworth**, 1997; **Thambugala, *et al.***, 2016). Further studies including molecular and chemical data indicated that *Rhytidhysteron* is more diverse than assumed and that *R. rufulum* is species complex (**Boehm, *et al.***, 2009; **Murillo, *et al.***, 2009; **Yacharoen, *et al.***, 2015; **Doilom, *et al.***, 2016; **Thambugala, *et al.***, 2016). As a result, about 20 species are currently recognized, but based on the rather local studies by **Murillo, *et al.*** (2009) in Costa Rica and **Doilom, *et al.*** (2016) and **Thambugala, *et al.*** (2016) in Thailand, many more species are to be expected in this genus. Here we describe a new species discovered in Colombia, which even without the support of molecular data differs from all other accepted species, featuring large ascomata and the longest ascospores known in the genus and a unique ascoma morphology, with yellowish-green-pruinose margins.

Materials and methods

Material was collected during a field trip to Cerro San Antonio in a mountainous area of Cali, as part of the III Meeting of the Colombian Group of Lichenologists (GCOL). This locality, also known as Cerro de la Horqueta, is 22 km from the city of Cali, at an altitude of 2000–2200 m, with the coordinates 76º58'N and 03º29'W. The Cerro San Antonio is characterized by a rather well-preserved montane forest and has been the subject of several studies of flora (**Alvarez, *et al.***, 1987).

We took pictures of the fungus both in the field and the laboratory with a Nikon D3100 and a Canon PowerShot SX160 digital camera. Ascomata sections were mounted for microscopic identification in a water medium. Measurements of ascomata, hamathecium, exciple, ascospores and asci were made in water. Photographs of the hamathecium, exciple, and ascospores, were taken with an Olympus microscope and Canon PowerShot SX160 camera. The material was deposited in the herbarium of the universidad del Valle.

Results

Rhytidhysteron columbiense Soto-Medina & Lücking, **sp. nov.** Figure 1. Index Fungorum IF552900

Diagnosis. A new species similar to *Rhytidhysteron brasiliense*, particularly in ascospore size, but differing in the larger ascomata with yellowish-green pruina on the margin.

Type. COLOMBIA. Valle del Cauca: Cali, Cerro de San Antonio (Horqueta); 76º58'14"N, 5º44'55"W, 2000–2200 m; on fallen, decaying woody branch; April 2013, E. A. Soto-Medina & R. Lücking s.n (holotype CUVC 62421).

Etymology. The species epithet, *columbiense*, refers to the country where the material was collected.

Description. Saprobic on decaying branches in montane rain forest remnants. *Ascomata* 1.5–(2.66)–3.0 mm long, 1.2–(1.59)–1.8 mm wide, 0.6–0.7 mm high (n = 14), hysterothecial, boat-shaped, crowded to aggregate, superficial, brown-black but with distinct, yellowish-green pruina on the margins, with exposed, lenticular to irregular, brown-black disc when wet, folded along the margins when dry and forming an elongate slit, perpendicularly striate. *Exciple* 60–90 µm wide (n = 8), composed of dark brown to black, thick-walled cells of *textura angularis*. *Hamathecium* comprised of dense, septate pseudoparaphyses, branched and forming a dark epithecium above the asci, fused and slightly swollen at the apex and enclosed in a gelatinous matrix. *Asci* 175–190 × 14–18 µm, 6–8-spored, bitunicate, cylindrical, with short, furcate pedicel, rounded at the apex, with distinct ocular chamber. *Ascospores* 38– (43.5)–52 × 13–(14.7)–18 µm (n = 17), uniseriate, slightly overlapping, hyaline when immature, becoming reddish-brown when mature, ellipsoidal to fusiform, rounded to slightly pointed at both ends, (1–)3-septate, with smooth wall. *Asexual morph*: not observed.

Discussion

Except for medical and chemical studies or studies relating to economically important crops (**Pena**, 1967; **Restrepo**, 1970; **Chaparro, *et al.***, 2009; **Vega, *et al.***, 2010), ascomycetous fungi have received little attention in Colombia, with only a few reports of conspicuous families such as Cordycipitaceae, Nectriaceae, Sarcoscyphaceae, and Xylariaceae. This is actually the first report of the genus *Rhytidhysteron* and the family Patellariaceae for Colombia (**Vazco & Franco Molano**, 2014), and it is striking that this first find resulted in the discovery of a species new to science.

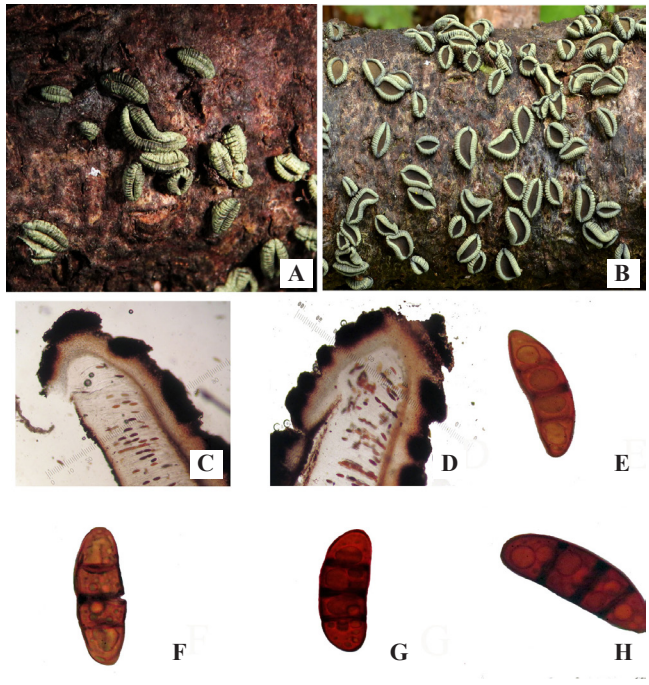

Figure 1. Dry Ascomata of *Rhytidhysteron columbiense* **A**, appearance of hysterothecia on host **B**, Hamathecium detail and striations **C**, asci with ascospores **D**, ascospores **E-H.**

Rhytidhysteron columbiense agrees with most other species in the genus in the more or less boat-shaped, sessile ascomata with a perpendicularly striate margin. This combination of characters is also found in *R. rufulum*, which has recently been shown to be a collective taxon including several species (**Murillo, *et al.***, 2009; **Thambugala, *et al.***, 2016). However, *R. columbiense* is not another segregate of the *R. rufulum* aggregate, since it differs from the latter in important features. All species or lineages currently distinguished in the *R. rufulum* aggregate have ascomata with non-pruinose, black margins and either black or colored (red-brown to orange) disc, quite different from the yellow-green pruinose margins and non-pruinose, brown-black disc in *R. columbiense*. The only exception is material of *R. rufulum* s.lat. reported from Japan and Taiwan (**Hsieh, *et al.***, 1997; **Tanaka & Hosoya**, 2006), which exhibits chocolate-brown discs combined with a grey margin [http://chawantake.sakura.ne.jp/data/Rhytidhysteron_rufulum.html; http://old.taibif.tw/en/namecode/140476] and almost certainly represents an undescribed taxon. In addition, ascospores in species of the *R. rufulum* aggregate are 20–37 × 7–14 µm in size, whereas in *R. columbiense* they are much larger, 38–52 × 13–18 µm. Thus far, only two species of *Rhytidhysteron* are known with ascospores of a similar, large size: *R. brasiliense* has entirely black, non-pruinose ascomata and its ascospores are slightly broader and lack the characteristic red tinge of those of *R. columbiense* (**Spegazzini**, 1881; **Kutorga & Hawksworth**, 1997; **Thambugala, *et al.***, 2016); *R dissimile* (P. Karst.) Magnes also features black, non-pruinose ascomata and has predominantly 5-septate ascospores that remain yellowish.

Even without molecular data, this finding highlights the urgent need for a deeper study of microfungi in Colombia, which are supposed to be much more diverse than macrofungi, but their potential richness in the country has barely been addressed by taxonomic study and hence their potential importance and uses cannot be currently assessed. In addition, this result supports the notion that microfungi are in important source for unrecognized species richness, and the genus *Rhytidhysteron* appears to be a prime example how such diversity has not previously been recognized, with potentially dozens of species waiting to be discovered. This is of particular interest considering the potential chemical and medical implications of fungi in this genus (**Spatafora, *et al.***, 1995; **Wipf, *et al.***, 2004; **Chowdhary, *et al.***, 2008; **Murillo, *et al.***, 2009; **Mahajan, *et al.***, 2014; **Mishra, *et al.***, 2014; **Pudhom & Teerawatananond**, 2014; **Pudhom, *et al.***, 2014; **Chander, *et al.***, 2016; **Chokpaiboon, *et al.***, 2016).

Provisional working key to known species of Rhytidhysteron in the world. In the following key, all currently listed names in the genus *Rhytidhysteron* are keyed out, with the exception of *R. minor* (Cooke) A. Pande [comb. inval.], which likely represents a species of *Opegrapha*, and *R. viride* Speg. (Spegazzini 1881), which appears to be an unrelated lichenicolous fungus; the taxonomic affinities of *R. beccarianum* (Ces.) Bat. & Valle are also uncertain although it is included below. The key provides access to the taxa thus far assigned to *Rhytidhysteron* but does not conform a taxonomic revision.

1a. Ascospores submuriform, with 3–5 transverse and 0–3 longitudinal septa per segment; ascomata with red-brown disc and black margins; type: U.S.A. ..... ***R. opuntiae*** (J.G. Br.) M.E. Barr (**Brown**, 1953; **Barr**, 1990)

1b. Ascospores transversely (1–)3(–5)-septate; ascomata variable 2

2a. Ascospores partially 5-septate, 30–46 × 12–20 µm; ascomata black with black, striate margins; type: Finland ..... ***R. dissimile*** (P. Karst.) Magnes (**Karsten**, 1866; **Magnes**, 1997)

2b. Ascospores at most 3-septate, variously sized; ascomata variable 3

3a. Ascospores consistently 1-septate, 20–28 × 12–15 µm; ascomata (red-)brown with brown margins; type: France ..... ***R. hysterinum*** (Dufour) Samuels & E. Müll. (**Samuels & Müller**, 1980)

4a. Ascospores 38–52 × 13–21 µm 5

4b. Ascospores (12–)19–37 × (5–)8–14 µm 6

5a. Ascomata brown-black with yellow-green, striate margins; ascospores red-brown; type: Colombia ..... ***R. columbiense*** Soto-Medina & Lücking (this paper)

5b. Ascospores black with black, striate margins; ascospores brown; type: Brazil ..... ***R. brasiliense*** Speg. (**Spegazzini**, 1881)

6a. Ascospores 12–15 × 5–6 µm; ascomata brown with olive margins; type: Sri Lanka ..... ***R. beccarianum*** (Ces.) Bat. & Valle (**Batista & Maia**, 1964)

6b. Ascospores 19–37 × 7–14 µm 7

7a. Ascoma disc (brown-)black or grey with (brown-)black margin 8

7b. Ascoma disc yellow- to red-brown or cinnabar-red with (brown-)black margin 13

Notes. If ascoma disc chocolate-brown with grey margins, see **Hsieh, *et al.*** (1997) and **Tanaka & Hosoya (**2006), which represents an undescribed species.

8a. Ascoma disc slate grey to dark grey 9

8b. Ascoma disc (brown-)black 10

9a. Ascoma margins striate; ascospores 25–30 × 12–14 µm, about 2 times as long as broad; type: U.S.A. ..... ***R. fuscum*** (Ellis & Everh.) J.L. Bezerra & Kimbr. (**Ellis**, 1889; **Ellis & Everhart**, 1892; **Bezerra & Kimbrough**, 1979)

9b. Ascoma margins smooth; ascospores 30–32 × 10–12 µm, about 3 times as long as broad; type: India ..... ***R. indicum*** (Anahosur) M.P. Sharma & K.S. Thind (**Anahosur**, 1971; **Sharma & Rawla**, 1986)

10a. Ascospores 30–37 × 10–14 µm; ascoma margins striate 11

10b. Ascospores 20–31 × 9–12 µm; ascoma margins variable 12

11a. Ascospores with reddish tinge; ascomata up to 2 mm long; type: Indonesia (Java) ..... ***R. javanicum*** Penz. & Sacc. (**Penzig & Saccardo**, 1897)

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11b. Ascospores lacking reddish tinge; ascomata up to 4 mm long; type: Paraguay ..... ***R. guaraniticum*** Speg. (**Spegazzini**, 1888)

Notes. *Rhytidhysteron* clade II from Costa Rica in the study of **Murillo, *et al.*** (2009) appears to belong to this taxon; it agrees in ascoma morphology and ascospore size.

12a. Ascomata margins striate; ascospores brown; type: U.S.A. ..... ***R. prosopidis*** Peck (**Peck**, 1894)

12b. Ascoma margins smooth; ascospores yellowish brown; type: Thailand ..... ***R. thailandicum*** Thambug. & K.D. Hyde (**Thambugala, *et al.***, 2016)

13a. Ascoma margin striate; ascoma disc orange- to red-brown (*R. rufulum* complex) 14

13b. Ascoma margin smooth; ascoma disc yellow-brown or orange to cinnabar-red without brown tinge 17

14a. Ascospores 23–24 × 8–9 µm; material: Costa Rica ***R.* clade IV** (**Murillo, *et al.***, 2009)

Notes. *Rhytidhysteron* clade IV from Costa Rica in the study of **Murillo, *et al.*** (2009) was considered to represent genuine *R. rufulum* but the ascospores are too small for that species; instead, clade III fits *R. rufulum* well (see below). If disc cinnabar-red, compare *R. quercinum* (see below).

14b. Ascospores 25–36 × 8–13 µm 15

15a. Ascospores 25–27 × 10–12 µm; ascospores dark brown; type: Australia ..... ***R. scortechinii*** Sacc. & Berl. (**Saccardo & Berlese**, 1885)

15b. Ascospores 28–36 × 9–13 µm; ascospores (reddish-) brown 16

16a. Ascoma disc orange-brown; ascospores brown; type: Paraguay ..... ***R. discolor*** (Speg.) Speg. (**Spegazzini**, 1880, 1919)

16b. Ascoma disc red-brown; ascospores reddish brown; type: Puerto Rico ..... ***R. rufulum*** (Spreng.) Speg. (**Spengel**, 1820; **Spegazzini**, 1921)

Notes. *Rhytidhysteron* clade III from Costa Rica in the study of **Murillo, *et al.*** (2009) seems to represent genuine *R. rufulum*, not clade IV as suggested in that study.

17a. Ascoma disc yellow-brown 18

17b. Ascoma disc orange to cinnabar-red 19

18a. Ascospores 27–34 × 7–12 µm, about 3–4 times as long as broad; ascomata rather stout, up to 1.8 mm long and 1.3 mm broad; ascospores reddish brown; type: Thailand ..... ***R. neorufulum*** Thambug. & K.D. Hyde (**Thambugala, *et al.***, 2016)

18b. Ascospores 19–31 × 8–13 µm, about 2–3 times as long as broad; ascomata elongate, up to 3.4 mm long and 0.7 mm broad; ascospores dark brown; type: Thailand ..... ***R. tectonae*** Doilom & K.D. Hyde (**Doilom, *et al.***, 2016)

19a. Ascoma disc cinnabar-red; ascospores 19–25 × 7.5–11.5 µm; type: India ..... ***R. quercinum*** (B.G. Desai & V.N. Pathak) M.P. Sharma & Rawla (**Desai & Pathak**, 1970; **Sharma & Rawla**, 1986)

19b. Ascoma disc orange; ascospores 28–37 × 9–14 µm; material: Costa Rica ..... ***R.* clade I** (**Murillo, *et al.***, 2009)

Notes. This clade likely represents an undescribed species.

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Conflict of interests

The authors declare no conflict of interest.

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