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New records of *Loculoascomycetes* from natural protected areas in Sonora, Mexico

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Abstract — Thirty collections of *Loculoascomycetes* from the Ajos-Bavispe National Forest Reserve and Wildlife Refuge, the Pinacate and Great Altar Desert Biosphere Reserve, and the Sierra of Alamos-Rio Cuchujaqui Biosphere Reserve, in Sonora, Mexico were studied. Ten new records for the Mexican mycobiota are presented: *Capronia montana, Chaetoplea crossata, Didymosphaeria futilis, Glonium abbreviatum, Hysterographium mori, Montagnula infernalis, Patellaria atrata, Rhytidhysteron rufulum, Thyridaria macrostomoides, and Valsaria rubricosa.* Photographs of macro-and microscopic characters are given for some species.

Key words: Chaetothyriales, Hysteriales, Melanommatales, Patellariales, Pleosporales

Introduction

The term *Loculoascomycetes* is used for ascomycetes with bitunicate asci and septate ascospores (Kirk et al. 2008). There is some controversy over the taxonomy of the genera in this group, e.g., *Valsaria*, because authors such as Dennis (1978) have placed them in the *Loculoascomycetes* owing to their bitunicate asci while others, such as Barr (1990a), have included them in *Pyrenomycetes* arguing the presence of unitunicate asci. Boehm et al. (2009) studied four nuclear genes in different species of *Loculoascomycetes* and have proposed changes to the current taxonomy, e.g., *Rhytidhysteron rufulum* which was previously included in order *Patellariales* has been tentatively moved to the *Hysteriales*. Thus, *Patellaria atrata*, included in the present study on the mycobiota of Sonora, because of its nearness to *R. rufulum*, might also belong in the *Hysteriales*. As such, future genetic studies to compare and define the taxonomic concepts of these species are recommended.

The ecological importance of these saprophytic fungi is highlighted by their ability to degrade cellulose. They therefore break down organic matter and carry out one of the most important ecological functions, guaranteeing that organic matter is recycled in the ecosystem (Chapin et al. 2002). According to Méndez-Mayboca et al. (2007, 2008), 105 species of *Ascomycetes* have been recorded for Sonora. Of these, 14 are *Loculoascomycetes*, representing 20% of that reported for the Mexican mycobiota. With the 10 new records described in the present study, the current catalog of *Loculoascomycetes* will be comprised of 24 and 80 species for Sonora and Mexico respectively.

Material and methods

Thirty collections of *Loculoascomycetes* from the macromycetes collection of the Centro de Estudios Superiores del Estado de Sonora (CESUES), with some duplicates in the herbarium of the Instituto de Ecología A.C. (XAL) in Xalapa, Veracruz were identified. Specimens were collected in the following protected natural areas of Sonora: the Pinacate and Great Altar Desert Biosphere Reserve, the Sierra of Alamos-Rio Cuchujaqui Biosphere Reserve, and the Ajos-Bavispe National Forest Reserve and Wildlife Refuge.

Specimens were studied following the conventional mycology techniques given by Dennis (1978) and by Breitenbach & Kränzlin (1984), and were identified using specific literature such as the contributions of Scheinpflug (1958), Chesters & Bell (1970), Luttrell (1973), Barr (1978, 1990a,b, 1991), Glawe (1985), Bellemère et al. (1986), Holm & Holm (1988), Aptroot (1995), Ju et al. (1996), Vasilyeva (2000), Delgado-Rodríguez & Checa (2002) and Checa et al. (2007) among others.

Species studied

Capronia montana M.E. Barr, Mycotaxon 41(2): 430 (1991) FIGURES 1–2

Pseudothecia 0.1–0.3 mm diam., black, superficial, globose, gregarious, located on hyphae that form a blackish crust at the edge, centrally depressed with a small papilla, outer surface with setae of $20-45 \times 4-6 \mu m$, sharp, thick and dark; black, thin subiculum at the base. Asci 75–85 × 12–14 μm , bitunicate, claviform, with 8 spores irregularly biseriate, with a broad ocular chamber 9–13 μm long. Ascospores 19–23 × 5–6 μm , fusoid, hyaline when immature, brown when mature, with 5 transverse septa, with terminal cells obtuse, wall smooth. Pseudoparaphyses absent.

SPECIMENS EXAMINED — MEXICO. SONORA: Municipality of Cumpas, EL MEZQUITAL (109°38'23" W 29°57'26" N)—in mesquite vegetation on the fallen branches of *Prosopis* sp. Leg. A. Sánchez. 02.XII.2004 *CESUES 5381*.

OBSERVATIONS — The genus *Capronia* Sacc. is characterized by its spherical or globose ascoma, centrally depressed and with a small papilla, as well as by



FIGS. 1–9. Loculoascomycetes of Sonora, Mexico. 1–2: Capronia montana. 1. Asci. 2. Ascospores fusoid. 3–4: Chaetoplea crossata. 3. Pseudothecia. 4. Ascospores ellipsoid. 5–6: Didymosphaeria futilis. 5. Pseudothecia. 6. Ascospores ellipsoid. 7–8: Glonium abbreviatum. 7. Hysterothecia. 8. Ascospores fusiform. 9: Hysterographium mori. Ascospores muriform.

Scale bar: 1, 2, 8 = 5µm; 4, 6, 9 = 10µm; 3 = 0.5 mm; 5 = 3 mm; 7= 1 mm.

the presence of long, dark setae (Barr 1991). Although the hymenium of the material studied was not completely developed, the morphology of the ascoma and spore agreed with the description of *C. montana* made by Barr (1991).

Chaetoplea crossata (Ellis & Everh.) M.E. Barr, Mem. N. Y. Bot. Gdn. 62: 50 (1990) FIGURES 3–4

Pseudothecia 0.2–0.5 mm diam., black, globose, centrally depressed, with a small papilla, gregarious with a black, thin subiculum at the base. Asci 120–160 × 10–17 μ m, bitunicate, with a short stipe, cylindrical, with 8 spores, uniseriate. Ascospores 14–22 × 8.5–11 μ m, ellipsoid, muriform, olivaceous-brown, with 3 slightly constricted transverse septa, with 1(–3) discontinuous longitudinal septum; the cells at the edges have bifurcate septa, wall smooth. Pseudoparaphyses 1.5–2 μ m diam.

SPECIMENS EXAMINED — MEXICO. SONORA: Municipality of Cumpas, EL MEZQUITAL (109°38'23" W 29°57'26" N)—in mesquite vegetation. Leg. A. Sánchez. 27.VIII.2005 *CESUES* 6325.

OBSERVATIONS — The genus *Chaetoplea* (Sacc.) Clem. is characterized by spherical or globose ascomata, centrally depressed and with a small papilla (Barr 1990b). Owing to its spore morphology, *C. crossata* has been included in this genus. *Pleospora calvescens* (Fr. ex Desm.) Tul. & C. Tul. is similar to *C. crossata*, but can be differentiated by its narrower spores $(5-)6-8(-9) \mu m$ and occasionally without longitudinal septa. The spore width of the material studied is 8.5–11 μm and spores are muriform, characteristics which coincide with Barr's (1990b) description of *C. crossata*; this author also notes that this species has the largest spores in the genus.

Didymosphaeria futilis (Berk. & Broome) Rehm, Hedwigia 18: 167 (1879)

Figures 5–6

Pseudothecia 0.5–5 mm diam., black, semi-immersed to superficial, covered by a clypeus which extends across the substrate forming a blackish surrounding layer, globose, with a small ostiolar papilla, solitary to mostly gregarious. Asci 68–75 × 7–8 µm, bitunicate, cylindrical, octosporous, uniseriate. Ascospores 8–12 × 3–5 µm, ellipsoid, brown, with a transverse septum, occasionally the wall is covered with a gelatinous sheath. Trabecular pseudoparaphyses 1 µm diam.

Specimens examined — MEXICO. Sonora: Municipality of Puerto Peñasco, Sierra Los Tanques (113°20'13" W 31°50'01" N)—sarcocaule scrub vegetation. Leg. A. Sánchez, I. Encinas, J. Miranda. 06.XI.2003 *CESUES 5041*.

OBSERVATIONS — Aptroot (1995) published a monograph of the genus *Didymosphaeria* Fuckel with seven species. This genus is characterized by ascoma immersed in the substrate, globose, ascospores brown and bicellular. *Didymosphaeria futilis* has been included in this genus because of its spore morphology (Luttrell 1973). Although it is a cosmopolitan species that grows on a wide variety of substrates, its morphology is uniform with the exception

of spore size, which is always less than 13 μm in length (Aptroot 1995). The material studied coincides with the descriptions given by Scheinpflug (1958) and Aptroot (1995).

Glonium abbreviatum (Schwein.) L.M. Lohman, Bull. Torrey Bot. Club 64: 64 (1937) FIGURES 7–8

Hysterothecia 0.2–0.9 × 0.1–0.2 mm, black, carbonaceous, elongated, oval to linear, erumpent, gregarious. Peridium composed of pseudoparenchymatous cells. Asci 35–40 × 4–6 μ m, cylindrical-claviform, with 8 irregularly biseriate spores. Ascospores 5–7 × 2.5–3 μ m, fusiform, hyaline, with a slightly constricted median septum, smooth wall. Cellular pseudoparaphyses.

SPECIMENS EXAMINED — MEXICO. SONORA: Municipality of Álamos EL PLATANAR (108°40'40.4" W 26°59'26.7"N)—tropical deciduous forest. Leg. S. Chacón. 15.IX.2006 *CESUES 5764.*

OBSERVATIONS — The genus *Glonium* Muhl. is characterized by hyaline, bicellular spores (Zogg 1962), while *G. abbreviatum* can be distinguished from the other species in the genus by its spore morphology. The main characters of the material studied coincide with the descriptions of Zogg (1962) and Vasilyeva (2000). According to these authors, the distinguishing character of this species is that it has the smallest spores of the genus.

Hysterographium mori (Schwein.) Rehm, Ascomyceten: no. 363 (1876) FIGURE 9

Hysterothecia 0.4–1.2 × 0.2–0.3 mm, black, elongated, linear, erumpent, gregarious, with lines parallel to the longitudinal opening slit. Peridium composed of pseudoparenchymatous cells 4–14 × 3–9 µm, with external cells darker and more sclerotic. Asci 92–130 × (13–)14–16(–18) µm, cylindrical-claviform, with 8 irregularly biseriate spores. Ascospores (16–)17–24 × 6–10 µm, ellipsoid to fusiform, brown, muriform, with 3(–5) transverse septa and 1(–3) longitudinal septa, slightly constricted at the median septum, wall smooth. Cellular pseudoparaphyses 1 µm diam., hyaline.

SPECIMENS EXAMINED — MEXICO. SONORA: Municipality of Álamos HUERTA VIEJA (109°02'53.0" W 27°02'5.4" N)—tropical deciduous forest. Leg. S. Chacón. 14.IX.2006 CESUES 5736, CESUES 5801.

OBSERVATIONS — This taxon can be distinguished from the other species in the genus by its spore morphology. The main characters of the material studied coincide with the descriptions of Zogg (1962), Dennis (1978), Barr (1990b) and Linde (1992). Although the asci are cylindrical-claviform, and the arrangement of the spores is biseriate, some authors such as Delgado-Rodríguez & Checa (2002) and Zogg (1962) have described the asci as cylindrical since this species exhibits much variability in its spore morphology, depending on substrate type and environmental conditions (Linde 1992).

Montagnula infernalis (Niessl) Berl., Icon. Fung. (Abellini) 2: 68 (1896)

Figures 10–11

Pseudothecia 0.1–0.3 mm diam., black, globose, carbonaceous, solitary to gregarious, immersed, erumpent to superficial, covered by a clypeus. Asci 95–121 × 17–22 μ m, claviform, bitunicate, with 8 biseriate spores, occasionally uniseriate. Ascospores 18–22 × 8–11 μ m, ellipsoid, muriform, brown, with 3 transverse septa and the middle one constricted; with 1(–2) discontinuous longitudinal septa, which on rare occasions reach one of the extreme cells of the spore, wall subverrucose, surrounded by a gelatinous sheath. Cellular pseudoparaphyses 2–3 μ m diam., hyaline.

SPECIMENS EXAMINED — MEXICO. SONORA: Municipality of Puerto Peñasco Sierra BLANCA (113°25'40" W 31°31'55" N)—in microphyllous desert scrub. Leg. S. Chacón. 15.IX.2006 *CESUES 5092*.

OBSERVATIONS — The genus *Montagnula* Berl. is characterized by its ascomata immersed in the substrate, with clypeus, globose or spherical; claviform asci; pseudoparaphyses narrow and cellular; ascospores fusoid or ellipsoid, with transverse septa and one or more longitudinal septa (Barr 1990b). It is a genus characteristic of the monocots with a long stem. Owing to its ascospores which measure $20-23(-30) \times 7-10 \mu$ m, with 3 transverse septa, the material studied coincides with *M. infernalis* described by Barr (1990b).

Patellaria atrata (Hedw.) Fr., Syst. Mycol. (Lundae) 2: 160 (1822) FIGURES 12-13

Apothecia 0.5–1 mm diam., solitary or in groups of up to three ascomata, immersed and closed at first, later they open via a longitudinal pore forming a disc, black, superficial or occasionally immersed, sessile, circular or slightly elongated, carbonaceous, black excipulum. Asci $50-83 \times 13-19 \,\mu$ m, cylindrical-claviform, short stipe, bitunicate with 6 to 8 spores. Ascospores $20-27 \times 6-8 \,\mu$ m, claviform, hyaline, with 4–5 transverse septa, regularly biseriate. In the immature ascoma, the hamathecium forms the paraphysoids and in the mature, open ascomata, gives rise to the paraphyses, which are hyaline, $1.5-2 \,\mu$ m diam.; the epithecium reacts with KOH 5% becoming blue-green.

Specimens examined — MEXICO. Sonora: Municipality of Puerto Peñasco, Cráter El Elegante (113°22'55 W 31°51'34" N)—in sarcocaule scrub. Leg. A. Sánchez. 17.I.2004 *CESUES 5130*.

OBSERVATIONS — The genus *Patellaria* Fr. is characterized by hyaline spores and more than one septum (up to more than 5) (Kutorga & Hawksworth 1997). *Patellaria atrata* is distinguished from the other species in this genus by its claviform spore morphology. The main characters of the material studied coincide with the descriptions of Kutorga & Hawksworth (1997), who also highlight for this species apothecium surface black and epithecium dark



FIGS. 10–19. Loculoascomycetes of Sonora, Mexico. 10–11: Montagnula infernalis. 10. Pseudothecia.
11. Ascospores ellipsoid to muriform. 12–13: Patellaria atrata. 12. Apothecia. 13. Ascospores claviform. 14–15: Rhytidhysteron rufulum. 14. Apothecia. 15. Ascospores ellipsoid to fusiform.
16–17: Thyridaria macrostomoides. 16. Pseudothecia. 17. Ascospores subfusoid. 18–19: Valsaria rubricosa. 18. Stroma hypoxyloid. 19. Ascospores ellipsoid.

Scale bar: 10, 16 = 0.5 mm; 11, 13, 15, 17 = 10 μ m; 12 = 0.1 mm; 14, 18 = 1 mm; 19 = 5 μ m.

green. Bellemère et al. (1986) observed that the hamathecium has paraphysoid filaments when the ascoma is young and filaments that are similar to mature paraphyses; observations which concur perfectly with the material studied.

Rhytidhysteron rufulum (Spreng.) Speg., Bol. Acad. Nac. Ci. 25: 79 (1921)

Figures 14–15

Apothecia $1-2.2 \times 0.5-1.5$ mm, solitary to gregarious, erumpent; black when young, linear, elongated, carbonaceous; when mature the fruiting bodies have a disc that is typical in appearance with irregular margins, black to dark brown; epithecium black, dark brown, red or yellow; when they age the margins curl upwards and the ascoma takes the shape of a hysterothecium with cracked lips that are very characteristic. Asci 139–190 × 12–17 µm, bitunicate, with a short stipe, cylindrical, with 8 uniseriate spores. Ascospores $23-31 \times 8-12$ µm, ellipsoid to fusiform, brown, with 1–3 transverse septa, under the light microscope the edges appear with a lighter colored area where the spore germinates, without actually forming germination pores, smooth. Cellular paraphyses 2–6 µm diam. Epithecium amyloid and turns violet with potassium hydroxide 5%.

SPECIMENS EXAMINED — MEXICO. SONORA: Municipality of Álamos PROMONTORIOS (109°02'10.5" W 27°00'54.1" N)—in tropical deciduous forest. Leg. A. Sánchez. 14.IX.2006 CESUES 5711, 5712, 5730, 5812.

OBSERVATIONS — The characteristics that identify the genus *Rhytidhysteron* Speg. are mainly the shape of the fruiting body and spore morphology (Samuels & Müller 1979). The immature ascoma is elongated in shape (lirelliform) with grooves; on maturing it opens along a central longitudinal line, forming an apothecium and finally under dry conditions the margin of the discs collapses becoming hysteriform, triangular or tri-radiate (Kutorga & Hawksworth 1997). Based on these characteristics, and ascospores with 3 transverse septa, the material agrees with *R. rufulum*. Although Samuels & Müller (1979) mention narrower spores $3.5-4.5(-6.5) \mu m$, the observed measurements coincide with those given by Kutorga & Hawksworth (1997): $(22-)25-35(-39) \times (7.5-) 9-12(-14) \mu m$. A related species is *R. hysterinum* (Dufour) Samuels & E. Müll., but its spores have a single transverse septum (Kutorga & Hawksworth 1997).

Thyridaria macrostomoides (De Not.) M.E. Barr, N. Amer. Fl., Ser. 2 (New York) 13: 34 (1990) FIGURES 16–17

Pseudothecia 0.5–1.2 mm, black, solitary, immersed or semi-immersed, covered with a dark layer similar to the clypeus that extends over the substrate; ostiole laterally compressed. Peridium formed by several layers, composed of pseudoparenchymatous cells 7–12 × 3–5 μ m. Asci 145–165 × 20–22 μ m, bitunicate, claviform, with 8 biseriate spores. Ascospores 29–45 × 10–12 μ m, subfusoid, first formed septum supramedian, dark with subhyaline terminal cells, with (3–)4–6(–8) transverse septa, smooth. Pseudoparaphyses trabecular.

Specimens examined — **MEXICO. Sonora: Municipality of Puerto Peñasco**, Cráter El Elegante (113°22'55" W 31°51'34" N)—in sarcocaule scrub. Leg. A. Sánchez. 08.XI.2003 *CESUES 5077, 5078, 5079*. OBSERVATIONS — This species is delimited by the morphology, size and number of septa of its spores. Owing to its laterally compressed ostiole it was previously included in the genus *Lophiostoma*, family *Lophiostomataceae* (Chesters & Bell 1970, Holm & Holm 1988). Barr (1990a) placed it in genus *Thyridaria* Sacc. in family *Platystomataceae*, indicating that the ostiole can be rounded or compressed and the distribution of the ascoma in valsoid groups or the presence of the clypeus or can have colored hyphae on them. These characteristics were observed on the material studied.

Valsaria rubricosa (Fr.) Sacc., Syll. Fung. (Abellini) 1: 743 (1882) FIGURES 18–19 Stroma 0.5–0.8×3–4 mm diam., hypoxyloid appearance, pigmented red-orange and surface spotted with black owing to the ostioles of the abundant superficial perithecia. Asci 90–108 × 10–14 µm, bitunicate, cylindrical, with 8 uniseriate spores. Ascospores $12-16 \times 6-9$ µm, ellipsoid, with a transverse median septum, ornamented wall, covered by gelatinous sheath. Pseudoparaphyses trabecular 3-4 µm diam., with free terminations.

SPECIMENS EXAMINED — MEXICO. SONORA: Municipality of Puerto Peñasco PAPALOTE (113°01'40.5" W 31°55'38.2" N)—in microphyllous desert scrub. Leg. A. Sánchez, I. Encinas, J. Miranda. 07.XI.2003 *CESUES 5059*.

OBSERVATIONS — Valsaria Ces. & De Not. is the subject of some controversy since some authors have placed it in *Loculoascomycetes* with bitunicate asci (Dennis 1978), while others like Barr (1990a) place it in *Pyrenomycetes* with unitunicate asci. The challenging characters which we think this taxon exhibits are bitunicate asci, that Barr (1978) defined as unitunicate with "pseudo Jack-in-the-box" dehiscence and hamathecium composed of paraphyses with free terminations, which together with the presence of the peridium delimiting each fruiting layer of the stroma, makes its placement difficult.

Ju et al. (1996) published a monograph of *Valsaria* and related genera and confirmed that the asci are bitunicate, without specifying taxonomic position. Some authors include *Valsaria* in *Diaporthaceae* (Glawe 1985, Wehmeyer 1923). However, Barr (1978) did not include it in her monograph on *Diaporthales* and later included it in family *Thyridiaceae* (Barr 1990a). Huhndorf (1992) stated that the asci have a refringent apical region that is not amyloid or chitinoid, but this was not observed in the material studied, which did not react with nigrosine.

The species of *Valsaria* produce at least two anamorphic states (Glawe 1985, Huhndorf 1992) as do some species of *Thyridium* Nitschke. Ju et al. (1996) give both genera bitunicate asci and thus assume a close phylogenetic relationship between the two. Based on spore morphology and the bitunicate asci, the material studied concurs with *V. rubricosa*. Its taxonomic position is currently unresolved, so new molecular studies are required.

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Valsaria rubricosa was grown on PDA medium from ascospores obtained from the fruiting body. The fungus grew as indicated by Huhndorf (1992) for *Valsonectria cinnamomi* (Ces.) Huhndorf, which was synonymised with *V. rubricosa*. Thus, after 15 days the mycelium covered the 9 cm Petri dish. In its center, the mycelium became orange and an orange pigment spread over the dish. In some Petri dishes a white pruinose band formed around this color and on the reverse side of the dish concentric circles of black dots were visible. On other dishes at 3 weeks granules 2 mm in diam. appeared, with a yellowgreen or yellow-orange color. According to Huhndorf (1992) these granules are sterile stromata at first and after about two months become fertile stromata with ascospores.

Huhndorf (1992) stated that for the bicellular ascospores, in 2 to 8 hours each one of their cells germinates directly into conidia. The ascospores are surrounded by these conidia that on germination form hyphae that spread out radially from the spore, very quickly initiating a growth colony. Under the light microscope we observed the hyphae of the culture and the presence of ellipsoid to cylindrical, unicellular and hyaline conidia capable of budding from one or several loci to give rise to secondary conidia. In our cultures, we did not observe the presence of star-shaped accumulates composed of dark hyphae that form arthrospores. Huhndorf (1992) observed them in 10 to 25% of the dishes. Sterile stromata are formed by tightly packed hyphae. Also, we observed that when these stromata are formed, a reddish colored pigment spreads across the agar, the same pigment that appears with KOH 5%.

After two months, the black sterile stroma elongate, forming multiple necks spreading at the tip. After three months they are sterile at approx 2 mm diam. and 3 mm high. Under the light microscope they have a pseudoparenchymatous structure, the characteristic red pigment typical of naturally developed stroma does not appear in KOH.

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