

Discovering unrecorded genera of *Helotiales* in the Canary Islands

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Abstract: We report the first records for the Canary Islands of species of three genera of *Helotiales*, *Dematiocypha dematiicola*, *Hamatocanthoscypha ocellata* and *Hyalopeziza nectrioidea*. Macro-, micromorphological and ecological data are provided for these species.

Keywords: Ascomycota, diversity, *Hyaloscyphaceae*, *Leotiomyces*, Macaronesia, *Pezizellaceae*, taxonomy.

Resumen: Tres especies de tres diferentes géneros de *Helotiales* no citados con anterioridad para las Islas Canarias se describen en este artículo, *Dematiocypha dematiicola*, *Hamatocanthoscypha ocellata* y *Hyalopeziza nectrioidea*. Incluimos aquí descripciones macro y micromorfológicas, así como información sobre la ecología de cada especie.

Palabras clave: Ascomycota, diversidad, *Hyaloscyphaceae*, *Leotiomyces*, Macaronesia, *Pezizellaceae*, taxonomía.

Introduction

The Canary Islands are recognized worldwide as an important hot-spot of diversity of living beings (MYERS *et al.*, 2000). More than two thousand species of fungi have been reported for this archipelago with 7447 km² of emerged land. Nearly seven percent of the species occur there exclusively (BELTRÁN-TEJERA *et al.*, 2019). *Helotiales* Nannf. ex Korf & Lizoň, with more than two thousand species, is the most diverse order in *Leotiomyces* O.E. Erikss. & Winka. What we know about the order in the Canary Islands is based on scattered reports through history rather than from monographic treatments of families and genera. The number of reports has increased constantly since the first report in 1874 (BERKELEY, 1874). Before 1980 there were fewer than 20 publications that included species from the area. The pace of discovery quickened after Korf and his disciples started working on Macaronesian fungi (KORF, 1978; KORF *et al.*, 1978; OUELETTE & KORF, 1979; GREENLEAF & KORF, 1980; KORF, 1981; ITURRIAGA & KORF, 1992, 1997, 1998; LIZOŇ *et al.*, 1998), and also thanks to the important effort of Beltrán-Tejera and collaborators in the last 47 years. Today there are c. 84 research papers that summarize 76 species in 41 genera in 14 families in the archipelago (QUIJADA, 2015; BELTRÁN-TEJERA *et al.*, 2019). Most of the genera have two or three species, a third of the genera have only one species and the most speciose are *Cistella* Qué. (6 spp.), *Mollisia* (Fr.) P. Karst. (5 spp.), *Arachnopeziza* Fuckel, *Hyaloscypha* Boud., *Lachnum* Retz. and *Pezicula* Tul. & C. Tul. (4 spp. each) (BELTRÁN-TEJERA *et al.*, 2019). Unfortunately, most of the citations of *Helotiales* (76%) have no accurate information linked with the report. Only 24% of the reports, mainly those made by Beltrán-Tejera and collaborators, have specific information about the locality (coordinates, altitude) and ecology (substrate type, host-plant, vegetation type, etc.) of the helotialean fungi (QUIJADA, 2015). According to the revision of QUIJADA (2015) more than 70% of the reports come from lignicolous species. The most common host plants are: *Castanea sativa*, *Erica arborea*, *Laurus novocanariensis*, *Pinus canariensis*, *Chamaecytisus proliferus*, *Eucalyptus globulus*, *Ilex canariensis* and *Morella faya*. All of them with more than five reports of associated fungi (QUIJADA, 2015). These results from common hosts clearly lead to the conclusion that laurel and pine forest are the ecosystems most explored for this group of fungi, but also there is an important number of reports in disturbed ecosystems such as *Castanea* and *Eucalyptus* plantations (QUIJADA, 2015).

Hyaloscyphaceae Nannf. is the most diverse family in the Canary Islands (BELTRÁN-TEJERA *et al.*, 2019). In its old concept, *Hyaloscyphaceae* Nannf. (*sensu lato*), is characterized by its small to large, brightly coloured, superficial to suberumpent, sessile to stipitate apothecia, covered outside by conspicuous hairs. But the old concept fell apart when HAN *et al.* (2014) published their review of

the family. Their extensive taxa sampling using four genes, showed that the morphological resemblances among genera in *Hyaloscyphaceae* evolve independently in ten different clades throughout the *Helotiales* (HAN *et al.*, 2014). Today, the family has 24–26 genera, and some genera traditionally placed in it such as *Calycina* Nees ex Gray, *Ciliarina* Svrček, *Hamatocanthoscypha* Svrček, *Psilachnum* Höhn., etc. have been transferred to the family *Pezizellaceae* Velen. (BARAL, 2016). The aim of this paper is to expand the knowledge of *Helotiales*, families *Hyaloscyphaceae* and *Pezizellaceae*, in the Canary Islands, reporting three species in genera not previously recorded.

Material and methods

Descriptions of the localities sampled, vegetation characteristics and methods of study of the collections are those of QUIJADA (2015) and QUIJADA *et al.* (2015). All samples were collected from detached branches lying on the ground. All collections were photographed in the fresh conditions, on the same day or only two or three days after they were collected. The distribution of the species was explored using GBIF (The Global Biodiversity Information Facility) and the references contained in this article. Specimens are part of the Mycological Section of the Herbarium of the University of La Laguna (TFC). Color terminology refers to ANONYMOUS (1976). Locality names were taken from the web IDECanarias version 3.0 (<http://visor.grafcn.es/visorweb/>). Abbreviations: * = living state; † = dead state; CR = aqueous congo red; CRB = aqueous cresyl blue; H₂O = water; idem = the same; IKI+ = pore amyloid in MLZ or LUG with or without KOH pre-treatment; KOH = potassium hydroxide; LBs = lipid bodies; LCI = lipid content index according to BARAL & MARSON (2005); LUG = Lugol's solution; MLZ = Melzer's reagent; VBs = vacuolar bodies.

Descriptions

Dematiocypha dematiicola (Berk. & Broome) Svrček, *Česká Mykol.*, 31: 193 (1977). Fig. 1

Apothecia 0.1–0.2 mm in diam., up to 0.8 mm high, scattered, not erumpent, broadly sessile, discoid with narrow attachment to urceolate, yellow Gray (93.yGray) to grayish greenish yellow (105.gy.gY), margin strong hairy and whitish. **Hairs** tapered, aseptate, straight to rarely sinuous; surface smooth with loosely attached amorph matter; without changes in KOH, LUG, MLZ or CR; at upper flank *43–53.5(56.7) µm long, 2.4–3.1 µm broad at base; at margin *44.5–69.3 × 2.6–3.1 µm long, 2.6–3.1 µm broad at base; apex tapering to 0.5–0.9 µm; 1/3–2/3 of length of the hair with solidified walls (0.1–

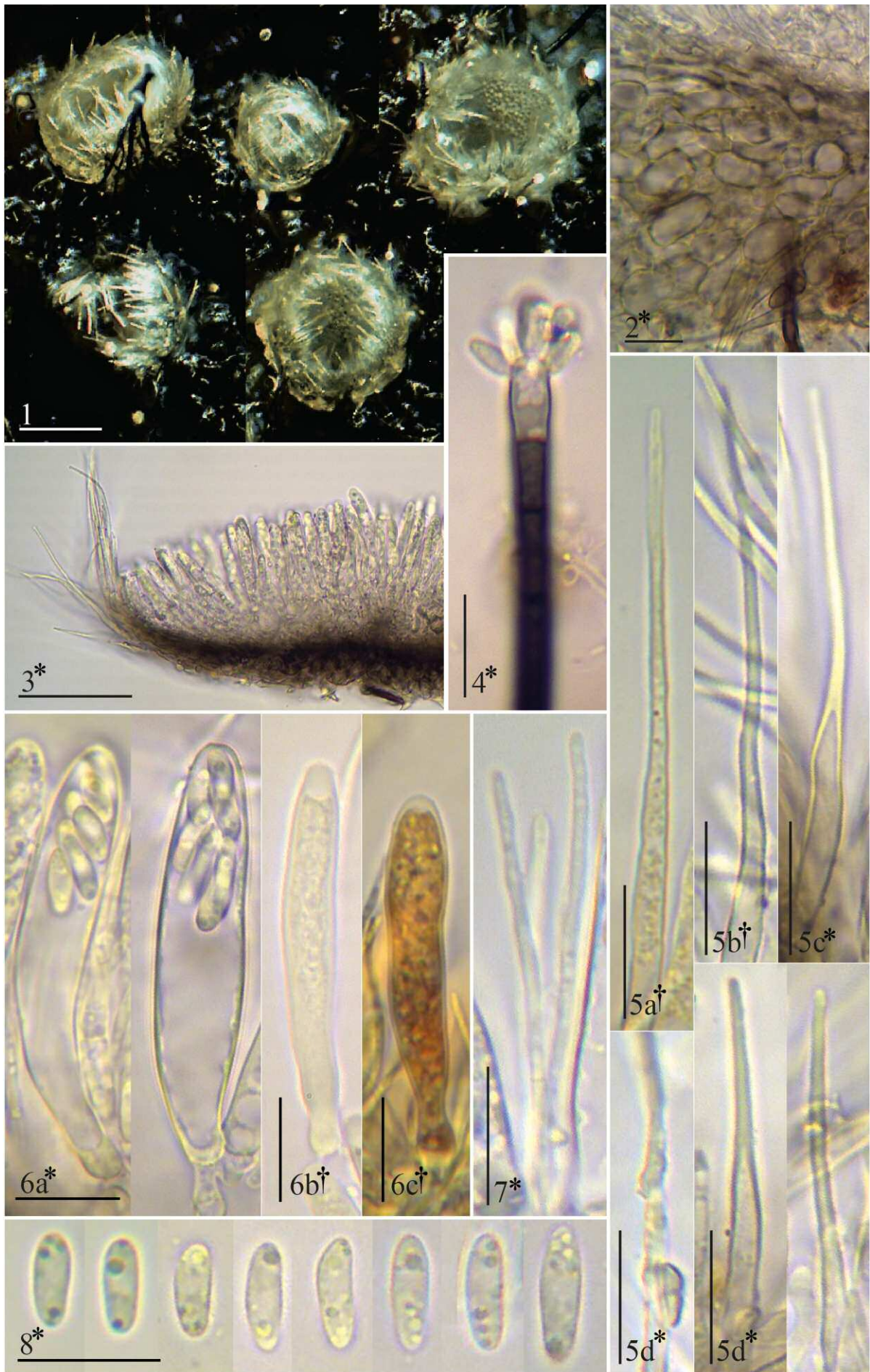


Fig. 1. – Morphological features of *Dematioscypha dematiicola*. 1. Fresh apothecia. 2. Excipular tissues in section. 3. Section of apothecia. 4. Anamorph. 5. Hairs. 6. Asci. 7. Paraphyses. 8. Ascospores. Scale bars: 100 µm= 1; 50 µm= 3; 10 µm= 2, 4, 5a-e, 6a-c, 7, 8. Mounted in: H₂O= 2, 3, 4, 5c-e, 6a, 7, 8; KOH= 5b; MLZ= 5a-6c. All photos from TFC Mic. 23687.

0.7 µm), not solidified at base and remaining in KOH. **Asci** *(39)41–43.4(45) × 6–7.8 µm, †32–37(41) × 4–6 µm; cylindrical-clavate, 8-spored, ascospores biseriata, *pars sporifera* *15–22.5 µm, pore IKI; base short arising from croziers. **Ascospores** *(6.5)7–8.5(10) × 2.2–2.9 µm, †5.7–7.2 × 1.6–2.1 µm; subcylindrical to clavate, straight or very slightly curved, 0-septate, hyaline, thin-walled, with 2–4(6) guttules (*0.4–1.2 µm diam.), LCI (7)12–20(30)%. **Paraphyses** uninflated, cylindrical, hyaline, 2–3-septate; terminal cell *(12)15–18(19) × 1–1.4 µm, second cell *7.5–10.5(13) × 1–1.8 µm; branched near base, thin-walled, guttules scanty in terminal and second cell. **Ectal excipulum** at base and middle flanks *textura prismatica-angularis*, *12–21 µm thick; at margin and upper flank *textura prismatica*, *6.5–11 µm; brown black (65.brBlack) to deep olive brown (96.d.OlBr), slightly gelatinised, without crystals, exudates or guttules. Ectal cells *(6.9)8–10.4 × (3)4–5.5 µm at middle flank, wall thickness *0.4–0.8 µm; *7.5–9(10) × (2.7)3.5–5 µm at margin.

Specimens studied: SPAIN, Tenerife, Santa Cruz de Tenerife, Anaga Rural Park, Barranco de la Mina, 28°33'12"N, 16°11'05"W, 781 m, hygrophilous evergreen laurel forest, on *Prunus lusitanica* subsp. *hixa*, 24 October 2012, L. & C. Quijada (TFC Mic. 23687).

Distribution and ecology: *Dematioscypha dematiicola* is distributed in the northern hemisphere in Europe (Austria, Belgium, Denmark, Estonia, Germany, Great Britain, Netherlands, Russia, Spain, Sweden), Asia (Japan) and North America (Canada, Mexico United States of America). In the southern hemisphere in New Zealand. It grows on hardwoods, usually found on *Betula*, *Carpinus*, *Corylus*, *Fagus*, *Quercus* and *Rhododendron*. There is only one report on *Cordyline australis*. Occurring from spring to autumn (GBIF, 2019; HOSOYA & OTANI, 1997; HUHTINEN, 1987; RAITVIIR, 2004; SPOONER, 1987).

Remarks: Our sample fits quite well the current concept of the species (HOSOYA *et al.*, 1997; HUHTINEN, 1987; RAITVIIR, 2004; SPOONER, 1987; SVRČEK, 1977). According to BARAL (2016) the genus has four species, but *Dematioscypha catenata* (Preuss) P.R. Johnst. was combined in the genus *Haplographium* Berk. & Broome based on phylogenetic data [*Haplographium catenatum* (Preuss) Hol.-Jech.] (SOMRITHIPOOL *et al.*, 2017). The latter is only known in its asexual morph. *Dematioscypha richonis* (Boud.) Huhtinen and *D. galanii* Raitv. differ from *D. dematiicola* in their hairs with dark-coloured incrustation (RAITVIIR, 2004). *Dematioscypha olivacea* (Velen.) Baral differs in its smaller asci and ascospores (VELENOVSKÝ, 1934).

Hamatocanthoscypha ocellata Huhtinen, *Karstenia*, 29(2): 194 (1990). Fig. 2

Apothecia 0.1–0.2(0.3) mm in diam., up to 0.1 mm high, scattered to gregarious, not erumpent, sessile to short stipitate (up to 40 µm high), white (263.White) to yellow white (92.yWhite), margin hairy. **Hairs** cylindrical, tapering to a blunt apex, 1–2(3)-septate, uncinata to rarely slightly curved; surface smooth, sometimes with dispersed warts, not dissolving or changing in CR, KOH, LUG or MLZ; at upper flank *20–31.5 µm long, *1.9–3 µm broad at base, at margin *(24)26.4–33(35.2) µm long, *1.9–3.1 µm broad at base; apex tapering to *1.1–1.6 µm. **Asci** *(23)25.5–27.7(30) × 4.2–5.3 µm, †(18)21–23.4(28) × 2.7–4 µm; cylindrical, 8-spored, ascospores biseriata, *pars sporifera* *11–17.4 µm, pore amyloid in MLZ or LUG with or without KOH pre-treatment; arising from croziers. **Ascospores** *(4.6)5.2–5.8(6.4) × 1.7–2.3 µm, †4–5.6 × 1.5–1.9 µm; cylindrical-subcylindrical to ovoid-clavate, 0-septate, hyaline, thin-walled, few tiny guttules or lacking, LCI (0)1–7(12)%. **Paraphyses** uninflated, cylindrical, 2–3-septate; terminal cell *(13)14.4–16.3(17.7) × 1.7–2.5 µm, cell below *(4.4)5.7–7.3(8.8) × 1.4–2.5 µm; not branched, thin-walled, without guttules in terminal cell and the cell below. **Ectal excipulum** at base

and middle flanks *textura globulosa-angularis* to *prismatica*, *19–25 µm thick; at margin and upper flank *textura prismatica*, *6–18 µm; hyaline, to deep orange yellow at the stipe (72.d.OY), not gelatinised, without crystals or exudates. Ectal cells *(7)8.5–11.5(13.5) × 3–6.6 µm at middle flank, wall thickness *0.3–0.6 µm; *7–9.4 × 2.7–3.2 µm at margin.

Specimens studied: SPAIN, Tenerife, La Orotava, Teide National Park, 28°19'51"N, 16°29'37"W, 2060 m, meso-oromediterranean summit broom scrub, on *Pinus pinaster*, 21 February 2013, L. & C. Quijada (TFC Mic. 23871). *Idem*, 23 March 2014, L. & C. Quijada and R. Negrín (TFC Mic. 24520).

Distribution and ecology: *Hamatocanthoscypha ocellata* is distributed in the northern hemisphere in Europe (Estonia, Finland, Netherlands, Russia) and Asia (Japan). In the southern hemisphere in New Zealand. Growing on softwood (*Picea*, *Pinus*, *Pseudotsuga*). Occurring from autumn to winter (GBIF, 2019; HUHTINEN, 1989; NZ-FUNGI, 2002–2019; RAITVIIR, 2004).

Remarks: The genus *Hamatocanthoscypha* is easily distinguished by its hyaline hairs with non-thickened walls and hooked apices (RAITVIIR, 2004). Our sample fits well with *H. ocellata*, although the ascospores and asci are in lower range of the variation with respect to the original description (4–6.4 × 1.5–2.3 µm vs. 4–12 × 2–2.3 µm, 18–30 × 2.7–5.3 µm vs. 28–45 × 3.4–5.8 µm, respectively). All the other morphological features and the ecology fit well (HUHTINEN, 1989). The most similar species is *H. obsoleta* Huhtinen, but it has yellowish to brown apothecia, excipulum covered with hyaline to brown crystals, and shorter hairs (HUHTINEN, 1989; RAITVIIR, 2004).

Hyalopeziza nectrioidea (Rehm) Raschle, *Sydowia*, 29 (1–6): 199 (1977). Fig. 3

Apothecia 0.1–0.3 mm in diam., up to 0.2 mm high, scattered to gregarious, not erumpent, broadly sessile, deep olive green (126.d.Ol.G) to olive black (114.OlBlack), margin hairy, white to light yellow green (135. l. y G). **Hairs** cylindrical to conical, tapering or not, with a blunt apex, 1–2(3)-septate, at flank they are curved downwards, but at margin they are straight to slightly sinuous near apex, usually constricted at the septa; surface smooth, sometimes with disperse granules of resinous exudates, not dissolving or changing in CR, KOH, LUG or MLZ¹; at upper flank *(27.3)41–56 (70.7) µm long, *2.5–3.9(4.9) µm broad at base, at margin *(35.6)49–58(67.7) µm long, *2.7–3.6(4.3) µm broad at base; apex tapering to *(0.9)1.4–1.7(2.4) µm. **Asci** *(29.7)32.7–36.4(43.5) × (6.9)7.8–8.6 (9.5) µm, †(21)26.4–30.7(36.6) × (5.1)5.9–6.7(7.5) µm; cylindrical, 8-spored, ascospores biseriata, *pars sporifera* *18–26 µm, pore amyloid in MLZ or LUG with or without KOH pre-treatment; arising from simple septa. **Ascospores** *(6.7)8–9.1(10.7) × 2.2–3 µm, †5.5–7.5(8.6) × 1.7–2.5 µm; cylindrical to sub-cylindrical, when young slightly clavate, without septa, hyaline, thin-walled, without drops or sparsely multiguttulate, LCI (4)7–12(20)%. **Paraphyses** uninflated cylindrical to medium clavate, 2–3-septate; terminal cell *(10.4)13–15.3(17) × 1.5–2.3 µm, cell below *(5.7)7–9(11.7) × 1.3–2.4 µm; not branched or bifurcate at base, thin-walled, without guttules. **Ectal excipulum** at base and middle flanks *textura globulosa-angularis* to *prismatica*, *51–85 µm thick; at margin and upper flank *textura prismatica*, *9–18 µm; medium brown (58.m.Br) to brown black (65.brBlack) at base and lower flank, vivid deep blue green (166. v. d. b G) at margin, not or slightly gelatinised, without crystals or exudates. Ectal cells *(6.6)9.7–11.6(16.2) × (4.5)6–7.6(10.7) µm at middle flank, wall thickness *0.5–1.1 µm; *(5.8)6.7–7.6(8.8) × (2.8)3.8–4.5(5.4) µm at margin.

¹ RASCHLE (1977: 199) observed a dextrinoid reaction.

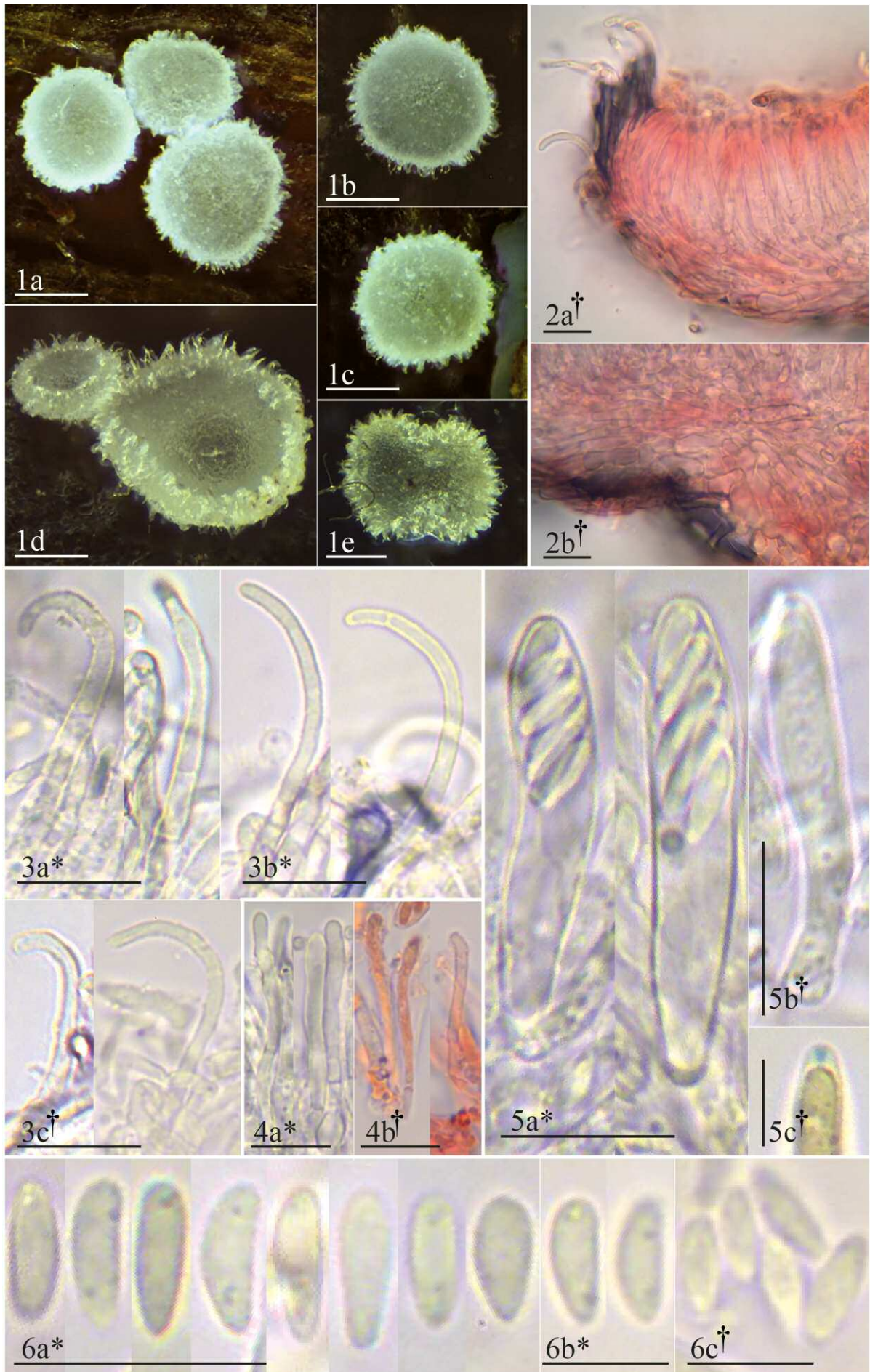


Fig. 2. – Morphological features of *Hamatocanthoscypha ocellata*. 1. Fresh apothecia. 2. Excipular tissues in section. 3. Hairs. 4. Paraphyses. 5. Asci. 6. Ascospores. Scale bars: 100 μm = 1a-e; 10 μm = 2a-b, 3a-c, 4a-b, 5a, 6a; 5 μm = 5b-c, 6b-c. Mounted in: CR= 2a-b, 4b; H_2O = 3a-b, 4a, 5a, 5c, 6a-b; KOH= 3c, 5b, 6c; MLZ= 5c. Photos: TFC Mic. 23871= 1a-c, 3a, 5a-c 6a; TFC Mic. 24520= 1d-e, 2a-b, 3b-c, 4a-b, 6b-c.

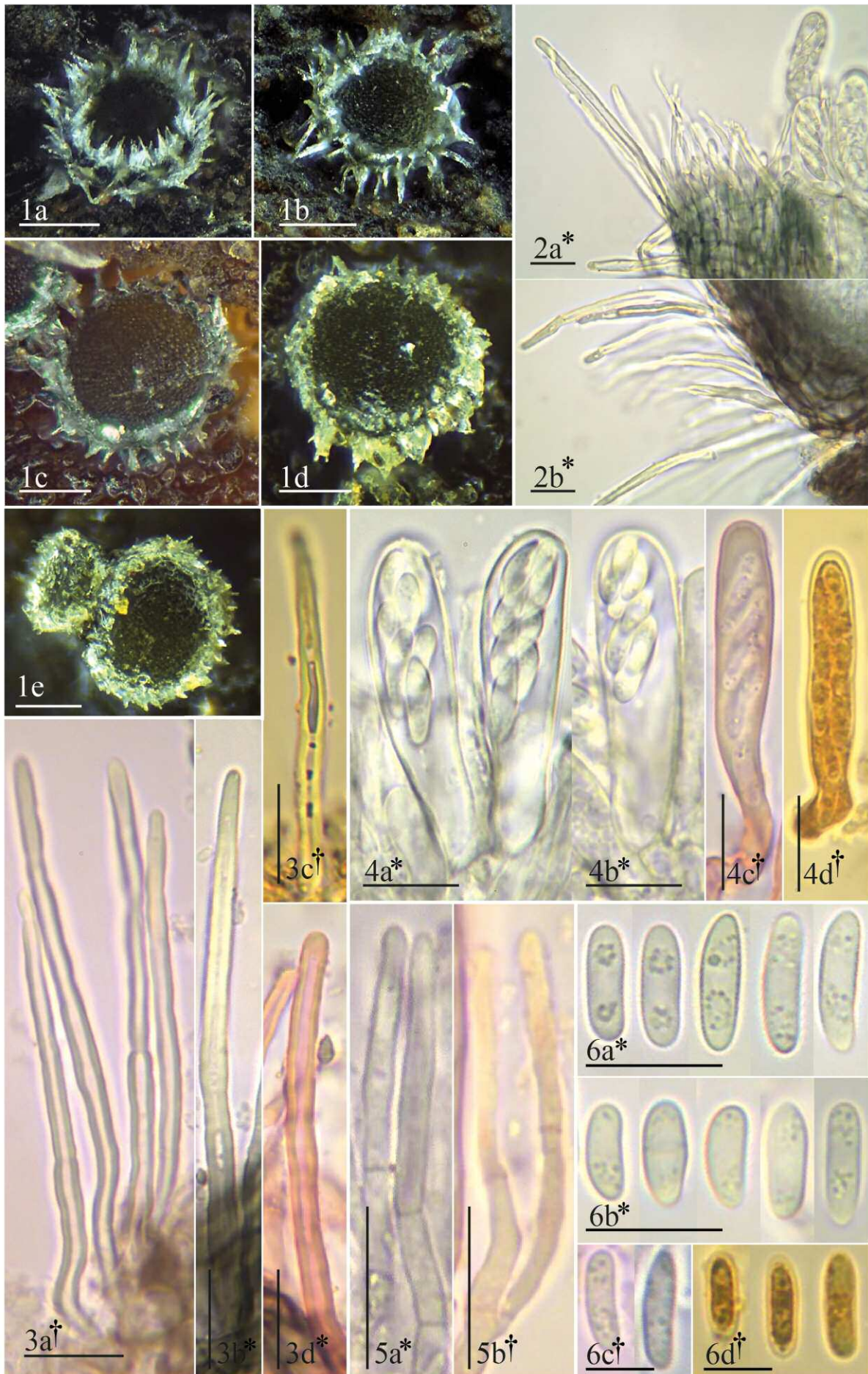


Fig. 3. – Morphological features of *Hyalopeziza nectrioidea*. 1. Fresh apothecia. 2. Excipular tissues in section. 3. Hairs. 4. Asci. 5. Paraphyses. 6. Ascospores. Scale bars: 100 μm = 1a-e; 10 μm = 2a-b, 3a-d, 4a-d, 5a-b, 6a-d. Mounted in: CR= 3d, 4c, 5b; H₂O= 2a-b, 3b, 4a-b, 5a, 6a-b; KOH= 3a, 6c; MLZ= 3c, 4d, 6d. Photos: TFC Mic. 23877= 1a-b, 3b, 4a, 5b, 6a; TFC Mic. 24524= 1c-e, 2a-b, 3a, 3c-d, 4b-d, 5a, 6b-d.

Specimens studied: SPAIN, Tenerife, La Orotava, Teide National Park, los Lajones, 28°19'51"N, 16°29'37"W, 2060 m, meso-oromediterranean summit broom scrubs, on *Spartocytisus supranubius*, 21 February 2013, L. & C. Quijada (TFC Mic. 23877). *Idem*, 23 March 2014, L. & C. Quijada (TFC Mic. 24524).

Distribution and ecology: *Hyalopeziza nectrioidea* is distributed in the northern hemisphere in Europe (Andorra, Austria, Spain, Switzerland). Growing on *Alnus*, *Rhododendron*, *Caluna* and *Salix*. Occurring from summer to autumn (GALÁN *et al.*, 1997; GBIF, 2019; RAITVIIR, 2004; REMLER, 1979; SENN-IRLET *et al.*, 2012).

Remarks: *Hyalopeziza nectrioidea* is an alpine species that grows on hardwood. As far we know it is a European taxon. The species differs from others by its dark ectal excipulum, long hairs (up to 100 µm length), inamyloid asci, and ellipsoid to ovoid ascospores without drops. There are four similar species: (1) *H. alni* E. Müll. with longer hairs, amyloid asci and fusoid ascospores; (2) *H. rubefasciens* L. Holm differs in its shorter ascospores (5.5–7.5 × 1.7–2.5 µm vs. 4–6 × 1.5 µm) and ecology, growing on *Lycopodium*; (3) *H. trichodea* (W. Phillips & Plowr.) Raitv. and (4) *H. valesiaca* Raschle have longer hairs (RAITVIIR, 2004).

Our sample fits well with descriptions by RAITVIIR (2004) and GALÁN *et al.* (1997). Our report seems to be the southernmost record in the northern hemisphere, but the ecology is the same because it appears in ecosystems located at high altitude, but not to alpine level in the biogeographical sense.

Discussion

Here we treat species of three genera in *Hyaloscyphaceae* (2 spp.) and *Pezizellaceae* (1 sp.). *Hyaloscyphaceae* is the most diverse family of *Helotiales* in the Canary Islands with five genera: *Cistella* (6 spp.), *Fuscolachnum* J.H. Haines (1 sp.), *Hyaloscypha* (4 spp.), *Hyphodiscus* Kirscht. (3 spp.) and *Psilocistella* Svrček (1 sp.). On the other hand, *Pezizellaceae* has only one genus (*Calycina*) with two species (BELTRÁN-TEJERA *et al.*, 2019). Species of all these genera were found in forest ecosystems (laurel and pine forest) except the genus *Cistella* which is more diverse in mesomediterranean summit broom scrubs (QUIJADA *et al.*, 2015). The genus *Hamatocanthoscypha* is reported for the first time to the Canary Islands, but also to the Macaronesian region since no reports exist in Azores, Madeira or Cape Verde (MELO *et al.*, 2010; MELO & CARDOSO, 2010; BAÑARES-BAUDET, 2005). We add here a additional genus to the family *Pezizellaceae* (*Hamatocanthoscypha*). *Hamatocanthoscypha ocellata* was found in the arid ecosystems at high altitude (meso-oromediterranean summit broom scrub) on *Pinus pinaster* (an introduced plant). The host is native to the western mediterranean basin (VIÑAS *et al.*, 2016), where no reports exist as far as we know for this fungus. *Dematioscypha* and *Hyalopeziza* are also both new reports to the Canary Islands and Macaronesia (MELO *et al.*, 2010; MELO & CARDOSO, 2010; BAÑARES-BAUDET, 2005). *Hyalopeziza nectrioidea* shares ecosystems with *Hamatocanthoscypha ocellata*, but it grows on an endemic substrate (*Spartocytisus supranubius*). *Dematioscypha dematiicola* was found in the most humid ecosystem, the hygrophilous evergreen laurel forest on *Prunus lusitanica* subsp. *hixa*, an endemic tree which is abundant in the northeastern laurel forest of Tenerife and in other islands of the archipelago (Hierro, La Palma, Gomera and Gran Canaria). These three new records increase the current knowledge of *Helotiales* in the Canary Islands, a group of fungi with a high undiscovered diversity in the Macaronesian region, as the results of QUIJADA (2015) and QUIJADA *et al.* (2015) point out.

Acknowledgements

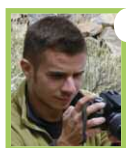
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