Patinella hyalophaea Sacc. – Rediscovered in New Brunswick, Canada

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Summary: Patinella hyalophaea (type species of Patinella) is redescribed from the type material from Italy and from a recent collection from Canada. Further records of this apparently rare species could not be traced. Its ecological amplitude involves decorticated branches of both angio- (Fagus) and gymnosperms in montane, humid beech forests mixed with conifers. A possible adaptation to flooding by streams is discussed. The affinities of Patinella within the Helotiales are unsettled, whereas an earlier proposed relation to the Orbiliomycetes based on the capitate paraphyses is considered as quite improbable.

Keywords: Discomycetes, Helotiales, semiaquatic.

Introduction

During a collecting trip in August 2012 to the Caledonia Gorge, New Brunswick (NB), Canada, the junior author found a non-descript black discomycete and tucked it away for further study. The collecting trip was, in fact, a two week intensive "Bioblitz" organized by the NB Museum with a goal to record the biodiversity in the Caledonia Gorge Protected Natural Area (PNA) (http://www.nbm-mnb.ca/index.php?option=com_content&view=article&id=598<e mid=1326). The NB Museum sponsors and supports a yearly Bioblitz

to larger PNAs with a goal to sample all these locales within twenty years. Biologists with various backgrounds including mammology, entomology, mycology, botany and invertebrate zoology congregate for two intensive weeks of collecting with the culmination of an Open House providing an opportunity for sharing and discussing the results with the public. The black discomycete was not part of the "show and tell" but turned out to be one of the more interesting mycological Patinella hyalophaea Sacc.

During a long-lasting monographic study of the *Orbiliomycetes* by the senior author, the type material of *Patinella hyalophaea* (type species of *Patinella* Sacc.) from Italy was examined in 2003, because Nannfeldt (1932) considered the genus to belong in the family *Orbiliaceae*. The result of this examination is presented here and a comparison with the Canadian specimen is made, a portion of which was examined by the senior author. Despite differences in the

substrate (angiosperm vs. gymnosperm wood), the Canadian specimen fully concurs with the type of *Patinella hyalophaea*. The phylogenetic relationship of this fungus is unknown. A similarity to the genus *Orbilia* based mainly on the capitate paraphyses and the ex-

cipulum of *textura angularis* is very probably a result of convergence. Judging from ascus morphology the species belongs in the *Helotiales*, with unclear affiliation in a family, though a relation to genera such as *Patellariopsis* Dennis etc. seems possible. A portion of the Canadian specimen was sent to Luis Quijada (Tenerife) for sequencing, but the result was not available when the present paper was submitted.



Fig. 1 – Collection habitat of *Patinella hyalophaea* (Crooked Creek, Caledonia Gorge, New Brunswick, Canada). Photograph: S. Clayden

Abbreviations: * = living state, † = dead state, |K| = Lugol's solution (~1% |2, ~3% KI), KOH = potassium hydroxide (~10%).

Results

Patinella hyalophaea Sacc., Grevillea, 4(29): 22 (1875).

DESCRIPTION OF THE SPECIMEN FROM New Brunswick (based on air dried material).

Apothecia rehydrated 0.25-1.2 mm diam, 0.16-0.3 mm thick (receptacle 0.1-0.2 mm), scattered to gregarious in small groups, black, superficial, sessile or with a short and broad, stout stipe up to 0.1 \times 0.6 mm, lens-shaped, flat to medium convex, finally centrally depressed, more or less round, margin indistinct, smooth, not protruding, dry with a thin, slightly incurved margin. **Asci** \dagger (40-)42–53(–65) \times (4–)4.5–5(–5.5) µm, 8-spored, spores uniseriate or biseriate in upper part, pars sporifera †23–30 µm long, entire ascus iodine negative in Lugol's solution (IKI, with or without KOH-pretreatment), apex hemispherical to subconical, with abrupt apical wall thickening (†2–3.5 μm thick), at base narrowed in a short to medium long stalk, arising from croziers. **Ascospores** *4.8–5.7 \times 2.5–2.7 μ m, †3.5– $5 \times 2.2 - 2.6 \,\mu\text{m}$, hyaline, aseptate, smooth, ellipsoid to slightly ovoid, straight or slightly inequilateral, containing 1(-2) inconspicuous, KOH-inert oil drops 0.3-0.8 μm diam in each half. Paraphyses filiform, branched below, sometimes also at upper septum, hyaline, with a slightly to strongly swollen, globose or gradually inflated (clavate) apex, upper half to third of the swollen apex with a 0.5-3(4) µm thick, bright olive-brown wall (pigment unchanged in KOH), protruding by ~3–6 μm beyond the dead asci, terminal cell $14-23 \times 3-4.8 \,\mu\text{m}$, lower cells $8-12 \times 1-1.5 \,\mu\text{m}$. **Subhymenium** 20-30 µm thick, of a small-celled, bright to dark olive-brown, textura angularis, cells $+6-9 \times 3-5 \mu m$. **Medullary excipulum** $80-200 \mu m$ thick at the base, cells $\pm 15-25(-30) \times 11-18(-23) \mu m$, of a thin-walled, large-celled textura (globulosa-)angularis with some intermingled hyphoid elements 2-6 µm wide, subhyaline to light yellowish-olive-brown, dark brown near attachment to substrate, pigment located in the intercellular exudate. Ectal excipulum very indistinctly delimited, ~40-60 µm at flanks, of indistinctly vertical orientation (70-90°), 15-25 µm thick at mid flanks and margin, bright olive-brown especially in cortex, cells at flanks $10-25 \times 7-$ 17 μ m, cortical cells at margin and flanks †8–11(–13) × 5–7(–9) μ m, more or less clavate, more or less freely protruding, with 1–1.5 μm thick, two-layered, olive-brown wall thickening (corresponding to the thickened wall of the paraphysis apices). Anchoring hyphae forming a dense, up to 60 µm thick, light ochre-brown layer of horizontally oriented hyphoid elements restricted to the place of attachment, cells $+5-14 \times 1.5-4 \mu m$, thin-walled, smooth. – Entire tissue without crystals, without IKI-reaction, olive-brown pigment unchanged in KOH (but turning a bit more yellowish), almost discoloured in potassium hypochlorite (KCIO) by turning pale yellowish-olivaceous. - Desiccation-tolerance: A few spores were still alive when the dry specimen was reexamined ~12 weeks after it was dried.

Locality and Habitat: Canada, New Brunswick, 33 km S of Moncton, 6 km NNW of Riverside-Albert, Caledonia Gorge Protected Natural Area, Crooked Creek Road, 102 m a.s.l., N 45° 47′ 42″ W 64° 46′ 27″, 21-Aug-2012, *leg*. A. Carter. The material was found on a portion of decorticated coniferous wood (6 cm diam) lying on the wet bank of a small stream (Crooked Creek) in a wet muddy area between two bridges. It is quite likely that the wood was submerged by water for a period of time, possibly due to flooding, but since it was not heavily decayed we feel that it was deposited recently by the side of the stream. The photo in Fig. 1 provides an idea of the habitat involved. Specimens are deposited in the NB Museum (A. Carter CGAC-147; NBM) and in the private herbarium of H.O. Baral (H.B. 9739).

DESCRIPTION OF THE SPECIMEN FROM ITALY (holotype).

Apothecia rehydrated 0.3-1 mm diam, 0.16–0.22 mm thick (receptacle 0.11–0.13 mm), sub- to densely gregarious, black, superficial, with a stipe-like base, lens-shaped, flat to slightly convex, round, finally slightly undulating and centrally depressed, margin indistinct,

smooth, not protruding. **Asci** $†41-54 \times 4.3-4.7(-5) \mu m$, 8-spored, spores uniseriate, sometimes subbiseriate in upper part, pars sporifera †21–34 µm long, entire ascus inamyloid (IKI), apex hemispherical to subconical, with abruptly thickened, 2 -> 1 μm thick wall, with a short to medium long stalk arising from croziers. **Ascospores** †3.8– $4.7 \times 2.4 - 2.5(-2.6)$ µm, hyaline, aseptate, smooth, ellipsoid to slightly ovoid, straight, containing 1–3 small LBs (lipid bodies) grouped in each half (0.2–0.6 μm diam). **Paraphyses** narrowly cylindrical, unbranched, hyaline, apically abruptly capitate to clavate, with a (0.5)1–2 μm thick, bright brown wall, protruding by ~2–4 μm beyond the dead asci, terminal cells $15-25 \times 3-5.5 \mu m$, lower cells $\pm 12 - 14 \times 1 - 1.3(-1.7) \, \mu \text{m}$. **Subhymenium** 15–30 μm thick, small-celled, dark (olivaceous-)brown. **Medullary excipulum** 90–130 μm thick at the base, of thin-walled, light (olive-)brown textura angularis(-prismatica). **Ectal excipulum** not clearly delimited, of textura angularis, cells $\dagger 9-22 \times 7-12 \,\mu\text{m}$, 15 μm thick at margin, light to bright (olive-)brown, pigment unchanged in KOH. Anchoring hyphae forming a dense, 5–15 μm thick layer, pale ochre-brown, †1.3–2 μm wide hyphae, wall 0.2(-0.3) µm thick, smooth. Entire tissue without crystals, without IKI-reaction.

Locality and Habitat: ITALY, Veneto, Belluno, \sim 45 km NNE of Treviso, SE of Belluno, Bosco del Cansiglio, \sim 1000–1500 m, N \sim 46° 4′ E \sim 12° 25′, on decorticated, somewhat eroded, ?6–10 cm thick branch of *Fagus sylvatica*, probably lying on the ground, on entirely slightly to strongly decayed wood, X.1874, collector not stated, Herb. mycol. P.A. Saccardo (PAD, holotype).

Discussion

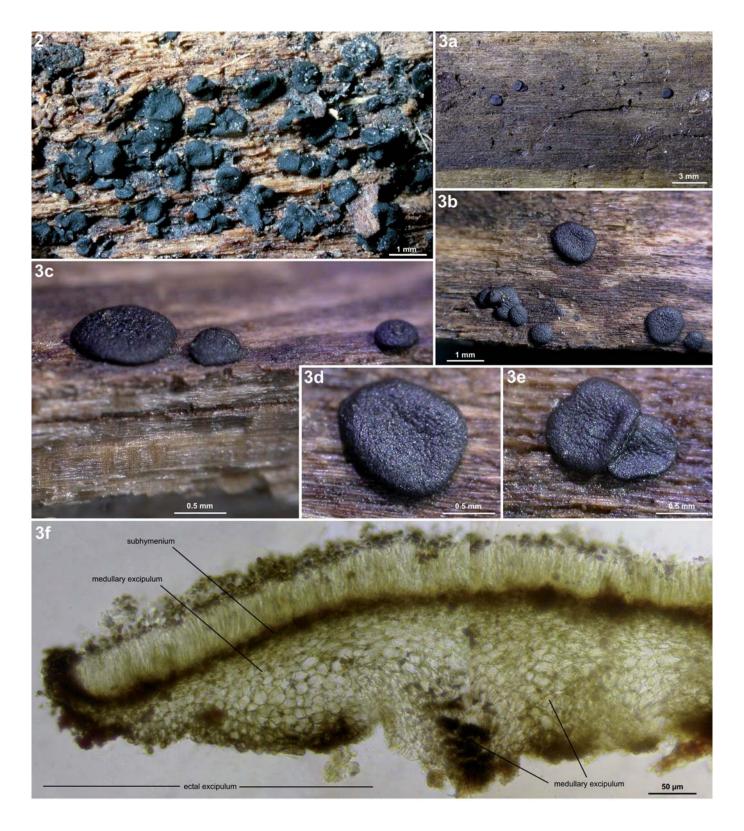
SACCARDO (1875a: 22) described the new genus *Patinella* without macro- and microscopic measurements, but with a remark ("edenda") that the single included species will be described elsewhere. The description of the species is found in SACCARDO (1875b: 129) and a copy of it in SACCARDO (1889: 769), and includes the following features: apothecia 1/2-3/4 mm diam (erroneously as 1/2-2/4 mm in SACCARDO, 1889); asci $50-55\times4.5$ µm, pars sporifera 30-35 µm long; spores 4×3 µm, ovoid; paraphyses filiform, apically bearing a globose, fuscous conidium 4 µm wide, often containing a single central guttule. A colour plate which includes the given measurements is found in SACCARDO (1877, fasc. 1, tab. 3, see Fig. 6). Here the ascospores are figured eguttulate, and the guttule in the "conidia" (= swollen apices of paraphyses) has a size of $\sim 1-1.3$ µm.

The present study of the holotype confirms Saccardo's diagnosis but revealed the ascospores a little narrower, and the paraphyses as eguttulate. Also, Saccardo depicted the asci as distinctly protruding beyond the paraphyses, though the asci were undoubtedly dead, judging from the uniseriate spores occupying most of the ascus volume. The wider spores noted by Saccardo might be due to the study of living spores, though their length lies at the lower end of the range of the present measurement in the type. Also the guttules in the paraphyses are apparently only recognizable in the living state and might represent either vacuolar (VBs) or cytoplasmic bodies (SCBs).

The Canadian record corresponds in all respects with the holotype, which is remarkable considering the geographic distance and the difference in the substrate. Although the living spores were distinctly longer in the present collection, the dead spores match the type very well in their size.

One of the most interesting structural features of *P. hyalophaea* is the nature of the paraphyses. They are darkly pigmented at the swollen apex while the rest of the paraphysis is hyaline. The pigment is located in the cell wall which is considerably and abruptly thickened in the upper half or third of the swollen apex. In older apothecia some paraphyses showed even 2/3 or the entire volume of the swollen apex occupied by the dark wall thickening (Fig. 4 I, n).

This character reminds one of the genera *Amandinea* M. Choisy (*Caliciales*) or *Catillaria* A. Massal. (*Lecanorales*), lichenized fungi with 1-septate ascospores and strongly amyloid, saccate asci of the leca-



Figs 2–3. – *Patinella hyalophaea*. 2, 3a–e. Apothecia *in situ* (after rehydration); 3f. Median section of apothecium (in KOH). – 2. Holotype (Italy, Veneto, on wood of *Fagus sylvatica*); 3. Specimen from New Brunswick (Canada, on wood of indet. gymnosperm). Photographs: H.-O. Baral.

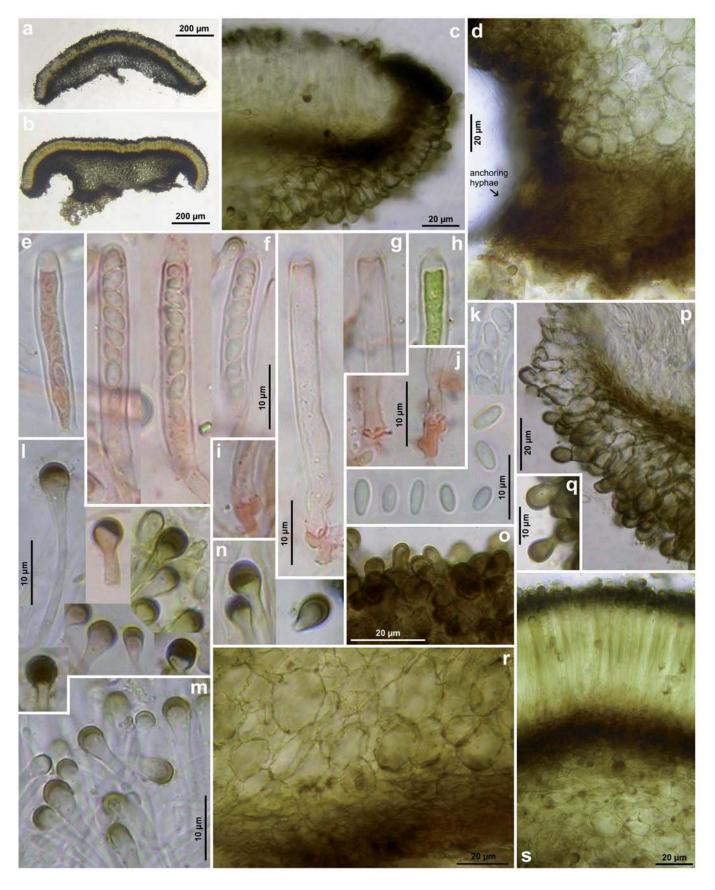


Fig. 4. – *Patinella hyalophaea* from New Brunswick. a–b. Median section of apothecia; c. Dto., marginal region, d. Dto., region of stipe-like base, with thick layer of anchoring hyphae; e, h. Immature asci; f. Mature asci; g. Emptied asci; i–j. Croziers at ascus base; k. Ascospores; l–n. apices of paraphyses; o. Margin in external view; p. Median section of marginal excipulum; q. Detail of protruding cortical cells, r. Median section of basal excipulum; s. Median section of hymenium, subhymenium and medullary excipulum. All elements in dead state (in KOH, e–g, i–j in KOH+CR, h in KOH+IKI, a in H₂O). Photographs: H.-O. Baral.

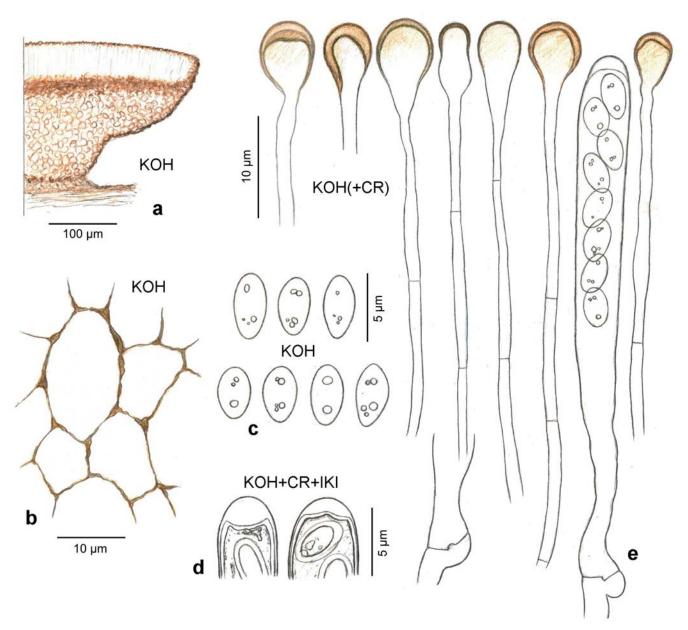


Fig. 5. – *Patinella hyalophaea*, illustrations from the holotype collection. a. Median section of apothecium; b. Dto., cells of ectal excipulum near base; c. Ascospores; d. Ascus apices; e. Ascus and paraphyses, ascus base arising from croziers. Drawings: H.-O. Baral.

noralean type. We assume that the thickened brown wall of the paraphysis apices emerged by convergence in these undoubtedly unrelated genera. The possible functional significance of the pigment cap is unknown; perhaps it plays a protective role, deterring feeding by invertebrates, or in regard to UV-absorption, although the site at Crooked Creek is rather shady.

Saccardo found the swollen parts of the paraphyses to detach soon from their filiform stalk, and he believed that they function as "conidia". We have occasionally observed detached apices of paraphyses, either after strong squashing, or when performing a section with a razor blade (see Fig. 4 l, n). However, Saccardo's interpretation as conidia is certainly incorrect, also because the uppermost septum of the terminal cell is always far below the apical inflation, hence breakage at the thin parts below the swollen apices inevitably leads to the death of these cells.

The swollen apices of the paraphyses, small asci and spores, and the angular texture of the excipulum are reminiscent of the *Orbiliaceae*. Indeed, NANNFELDT (1932) suggested an affinity with that family which was placed in the *Helotiales* at that time. However, most spe-

cies in the *Orbiliaceae* have asci with a furcate, simple-septate ascus base, and those with pronounced croziers have hyaline, translucent apothecia (genus *Hyalorbilia* Baral & G. Marson). Living spores of *Orbiliaceae* mostly contain a refractive structure that is unstable in lethal media, and no such structure could be seen in the few living spores of the Canadian material. We feel the fungus is better placed in the Helotiales, and here perhaps in the vicinity of genera such as *Patellariopsis* Dennis, *Patinellaria* P. Karst., *Phaeangella* (Sacc.) Massee, *Phaeopyxis* Rambold & Triebel, or perhaps *Bulgariella* P. Karst.

Many further species have been transferred to *Patinella* in the past. The Index Fungorum lists no less than 76 taxa (!), and most of them are unsolved in regard to their modern generic position. Whether any of them shows a close phylogenetic relationship to *P. hyalophaea* cannot be answered at present. The characteristic morphology of the paraphyses is unknown to us in any of them. For a more detailed historical review of the genus *Patinella* see BARAL *et al.* (in prep.).

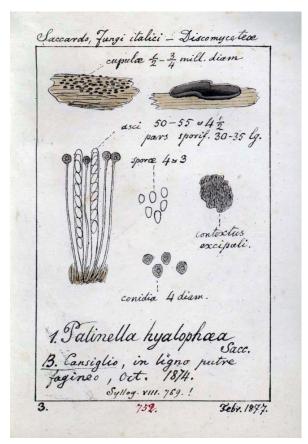


Fig. 6. – Original illustration of *Patinella hyalophaea* (SACCARDO, 1877, fasc. 1, tab. 3).

Ecology

The only collection studied by Saccardo was made in October 1874 in Bosco del Cansiglio in northeast Italy on partly strongly rotted decorticated wood of *Fagus sylvatica*, while ours was collected in August 2012 at Caledonia Gorge, New Brunswick, Canada on conferous wood and although decorticated not well rotted. Both substrates were confirmed by us by microscopic examination of the wood anatomy. The coniferous substrate might have been a manmade piece of lumber since there appear to be saw marks on some portions, perhaps fashioned as a structural piece for one of the bridges. There was no evidence of a preservative in the wood. While not found submerged in water, the numerous sand grains and small colony of *Massarina aquatica* J. Webster on the substrate strongly suggest association with an aquatic habitat including occasional flooding.

Based on our knowledge this report represents the first North American record of *Patinella hyalophaea* and probably the first report in over 130 years since its first detection. This leads to some speculation. Has the microhabitat not been thoroughly investigated? However, freshwater habitats are frequently investigated by

mycologists, and the present fungus, though not brightly coloured, should not have escaped notice due to its comparatively large size. More importantly, why is it found in such disparate areas? Are there similarities between Bosco Del Cansiglio (Italy) and Caledonia Gorge (Canada)?

Despite their wide geographic separation and contrasting elevations, the occurrences of *P. hyalophaea* in coastal eastern Canada and montane northeastern Italy are in broadly similar bioclimatic settings. Beech is an important component of the forest vegetation in both areas: *Fagus grandifolia* in Canada, *F. sylvatica* in Italy. The forests of the Caledonia Gorge PNA are hardwood and mixed coniferhardwood associations dominated by *Acer saccharum, Betula alleghaniensis*, *F. grandifolia*, *Picea rubens*, and *Abies balsamea*. In structure and composition, these stands have close affinities with humid montane forests in the Appalachian mountains, reflecting similar temperature and precipitation regimes (CLAYDEN *et al.*, 2011). In the southern (and northern) Alps, a gradient of climate and vegetation analogous to that in the Appalachians is present, with forests of beech giving way with increasing elevation to beech–fir (*Abies alba*) and spruce (*Picea abies*) (WALTER, 1985).

Whatever the reasons for the apparent rareness of *Patinella hyalophaea*, it is hoped that by bringing new observations and questions, mycologists might find similar collections in the field or buried in herbaria.

Acknowledgements

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