Psathyrella ephemera A. H. Smith, une espèce rare ou méconnue des milieux bourbeux , découverte en Belgique et en France

D. Deschuyteneer, P. Tanchaud, D. Wächter

Ecologie - Place of find

Introduction

De nombreuses récoltes de cette espèce ont été réalisées par le premier auteur, au cours des mois d'octobre 2018 et 2019 et septembre 2020 à Malines-Mechelen (B), dans le fond vaseux, induré mais encore légèrement humide, d'un étang asséché par l'été torride. Les basidiomes, le plus souvent isolés ou en petits groupes d'une dizaine d'exemplaires répartis sur plusieurs mètres carrés, apparaissaient dans la vase ou fixés à des branches enfouies, à proximité immédiate de vieux troncs de feuillus immergés depuis des années.

Le second auteur a découvert en Charente-Maritime (Fr), au fond d'un fossé asséché mais encore humide, une autre récolte que nous avons également pu identifier, dont l'écologie était strictement similaire et, semble t'il, distinctive.

Photos in situ de récoltes réalisées en Belgique (D. Deschuyteneer) - Voucher: DD-RUBE - Genbank Accession number/Version: **MN493776.1**

Introduction

Many samples of this species were collected by the first author in October 2018, 2019 and September 2020 in Mechelen (B), in the muddy, indurated but still slightly damp bottom of a pond dried out by the hot summer. The basidiomes, most often isolated or in small groups of about ten specimens spread over several square metres, appeared in the mud or attached to buried twigs, in the immediate vicinity of old deciduous trunks that had been submerged for years.

The second author discovered in Charente-Maritime (Fr), at the bottom of a dry but still wet ditch, another collection that we were also able to identify, whose ecology was strictly similar and, apparently, distinctive.

Spécimens adultes

Primordia et spécimens jeunes - Primordia and young specimens

Photos in situ de récoltes réalisées en Belgique (D. Deschuyteneer) - Voucher: DD-RUBE - Genbank Accession number/Version: **MN493776.1**


35 mm

Photo in situ (Patrice Tanchaud)

Récolte séquencée du 16-09-2019 - La Pichauderie, Saintes, Charente-Maritime – France ; dans un fossé asséché, mais encore humide.

ITS = 99.84% Psathyrella ephemera (MN493776)

Chapeau mesurant de 20 à 35(40) mm de diamètre, glabre, d'un brun marron chaud, devenant par la suite noisette et beige grisâtre, nettement plus pâle au niveau de la marge (pouvant apparaître blanchâtre), non strié au stade précoce, mais présentant une striation peu marquée au cours de la croissance, s'étendant depuis la marge jusqu'à mi-rayon ; conico-campanulé au stade précoce devenant plan-convexe, et alors souvent pourvu d'un large mamelon obtus. Hygrophane, il décolore en beige crème pâle. Chair concolore ; odeur sans particularité.
Lames au nombre de 45 à 55, alternant avec une ou plusieurs lamelles ou lamellules, assez larges (5mm), ventrues, largement adnées. Au stade précoce, beige pâle avec un léger reflet lilacin, moyennement serrées, devenant gris brunâtre et subdistantes ; l'arête fimbriée est blanche ou concolore.
Stipe mesurant 40-70 x 3-7 mm, le plus souvent court, épais et courbé, creux, fibrilleux blanchâtre, dont la base plus large est clavée ; pruineux au sommet, parfois teinté par la sporée, présentant une nette zone subannulaire au niveau du tiers inférieur ou à proximité de la base.
Voile fibrilleux blanchâtre, abondant, et reliant la marge du chapeau au stipe sur les primordia ; volatile, il persiste provisoirement au cours de la croissance sous forme de fibrilles éparses, nettement plus denses au niveau de la marge, où il peut former un mince filet aranéeux.

Cap measuring from 20 to 35(40) mm in diameter, glabrous, warm brown, later becoming hazelnut and greyish beige, distinctly paler at the margin (may appear whitish), not striated in the early stage, but with a slight striation during growth extending from the margin to mid-radius; conico-campanulate in the early stage becoming plano-convex, and then often with a wide obtuse mamelon. Hygrophanous, it discolours to pale creamy beige. Flesh concoloured; odour not distinctive.

Gills 45 to 55 in number, alternating with one or more lamellae or lamellulae, 5 mm wide, ventricose, largely adnate. In the early stage, pale beige with a slight lilacin sheen, moderately closed, becoming brownish grey and subdistant; Edge fimbriate white or concoloured.
Stipe measuring 40-70 x 3-7 mm, usually short, thick and curved, hollow, whitish, fibrillous, with a broader clavate base ; pruinose at top, sometimes spore-stained, with a distinct subannular zone in the lower third or close to the base.
Veil : whitish fibrillous, abundant and connecting the margin of the cap to the stipe on the primordia; volatile, it persists temporarily during growth in the form of scattered fibrills, much denser at the margin, where it can form a thin araneous net.

Spores mesurant (7,9)8,3-9,7(10,8) × (4,5)4,7-5,5(5,8) µm ; Q = (1,6)1,7-1,9(2,1) ; Me = 9 × 5 µm ; Qe = 1,8 ; brunes, non opaques, lisses, oblongues, ellipsoïdes et ovoïdes de face, légèrement phaséoliformes de profil, parfois avec une légère dépression supra-hilaire ; pore germinatif central, étroit ; apex conico-convexe, non tronqué .

Basides clavées, tétrasporiques.

Cheilocystides mesurant (33,5) 36 - 55,8 (61,2) × (8,6) 10,2 - 16 (17,4) µm ; Me = 45,7 × 13,4 µm ; nombreuses, denses ou éparses et alors regroupées sous forme de petits « clusters », majoritairement lagéniformes à lagéno-utriformes et parfois ventrues, peu ou pas stipitées, à col court et large, à sommet largement obtus, rarement fourchu, à paroi fine ou souvent modérément épaissie en particulier au niveau du sommet qui présente souvent de très nombreuses inclusions granulaires cristallines réfringentes intra et extracellulaires, et qui, sur le frais, est recouvert d'importants exsudats mucoïdes se colorant en vert dans l'ammoniaque à 10%, rappelant les dépôts également observés au niveau de l'arête de *Psathyrella tephrophylla*.

Paracystides clavées et sphéropédonculées sont peu nombreuses et leur fréquence est fonction de la densité des cheilocystides.

Pleurocystides mesurant (7,9)8,3-9,7(10,8) × (4,5)4,7-5,5(5,8) µm ; Me = 9 × 5 µm ; très nombreuses, lagéniformes, lagéno-utriformes, cylindriques, à paroi fine ou modérément épaissie, à col court et large, à sommet largement obtus et parfois incrusté ou contenant des granules réfringents, souvent longuement stipitées.

Médiostrate nettement pigmentée brun-clair.

Pileipellis constitué d'une à deux assises de cellules globuleuses, clavées et sphéro-pédonculées.

Pileitrame à hyphes cylindriques pigmentées.

Voile : fibrilles cylindriques adhérentes à la surface du chapeau .

Caulocystides analogues aux pleuro- et cheilocystides, ou versiformes, parfois fourchues, au sommet souvent épaissi, contenant également des granulations cristallines réfringentes et exsudant à frais de nombreuses gouttes mucoïdes colorées en vert dans l'ammoniaque 10%.

Boucles présentes.

Örstadius qui a examiné mes exsiccata : (communication personnelle): Your result of the examined collection hardly differed from mine.
I found the spores to be 9-10 x 5.2-6 µm, av. 9.5 x 5.5 µm.
Pleurocystidia 42-62 x 13-20 µm, utriform, lageniform, numerous.
Cheilocystidia 25-52 x 10-18 µm, similar in shape and frequency to the pleurocystidia; no clavate cells seen.
It´s difficult for me to give further comments as I have not examined Smith´s type.

Spores measuring (7,9)8,3-9,7(10,8) × (4,5)4,7-5,5(5,8) µm; Q = (1,6)1,7-1,9(2,1); Me = 9 × 5 µm; Qe = 1,8; medium red-brown, non-opaque, smooth, oblong, ellipsoid and ovoid in face view, slightly phaseoliform and sometimes with a slight suprahilar depression in profile; germ pore central, narrow, apex conico-convex, not truncate .
Basidia: clavate, 4spored.
Cheilocystidia measuring (33.5) 36 - 55.8 (61.2) × (8.6) 10.2 - 16 (17.4) µm; Me = 45.7 × 13.4 µm; numerous, dense or scattered and then grouped in small "clusters", mostly lageniform to lageno-utriform and sometimes clavate, slightly or not stipitate, with a short and wide neck and a wide obtuse apex, rarely forked, thin-walled or often moderately thickened, especially near the apex, which very often presents numerous intra- and extracellular refractive crystalline granules, and which in fresh specimens is covered with large mucoid drops turning green in 10% ammonia, reminiscent of the deposits also observed at the edge of *Psathyrella tephrophylla.*
Paracystidia clavate and spheropedunculate are scattered and their frequency depends on the density of the cheilocystidia.
Pleurocystidia measuring (7.9)8.3-9.7(10.8) × (4.5)4.7-5.5(5.8) µm; Me = 9 × 5 µm; very numerous, lageniform, lageno-utriform, cylindrical, thin-walled or moderately thickened, short and broad-necked, with largely obtuse apex which is sometimes encrusted or contains refractive granules; often lengthy stipitate.
Mediostrate distinctly light brown pigmented.
Pileipellis consisting of one or two layers of globulous, clavate and sphero-pedunculate cells.
Pileitrama: light brown pigmented cylindrical hyphae.
Veil: white**,** fibrillous.
Caulocystidia similar to pleuro and cheilocystidia, or versiform, sometimes forked, with often thickened tops, also containing or covered with refractive crystalline granulations and numerous green-coloured mucoid drops in 10% ammonia.
Clamps: present

Örstadius who examined my exsiccata: (personal communication)
Your result of the examined collection hardly differed from mine.
I found the spores to be 9-10 x 5.2-6 µm, av. 9.5 x 5.5 µm.
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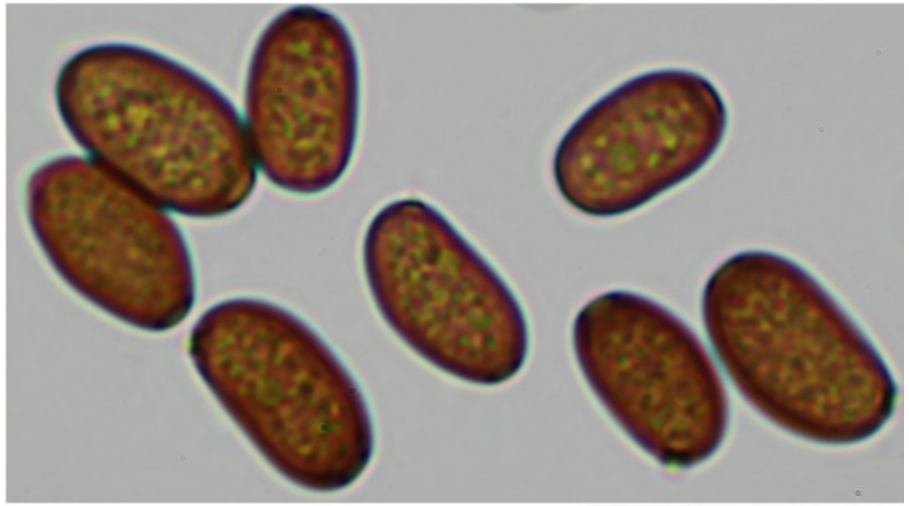
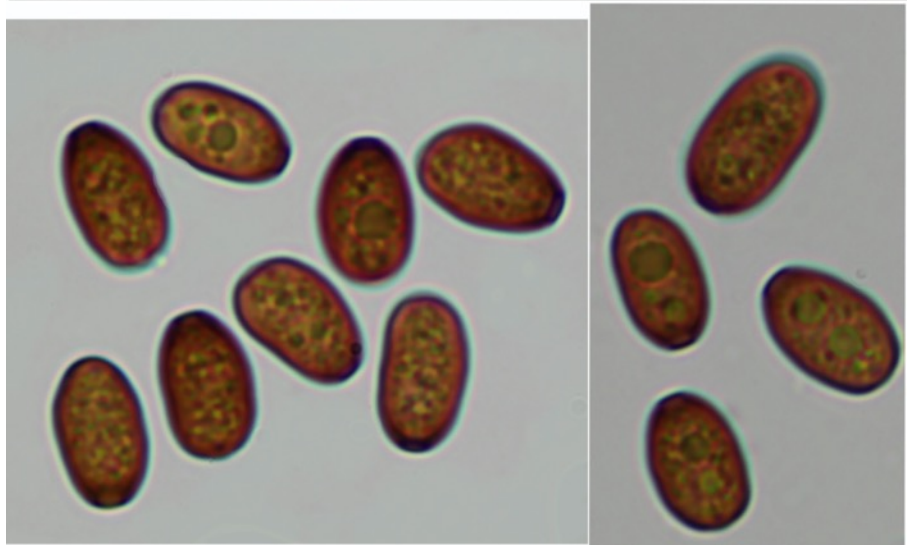
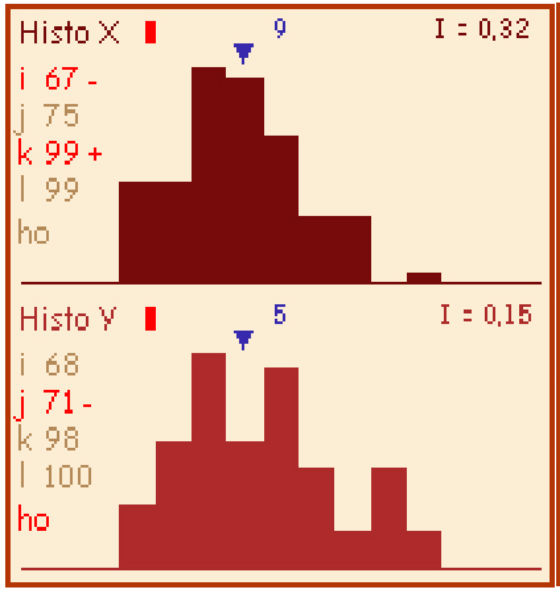


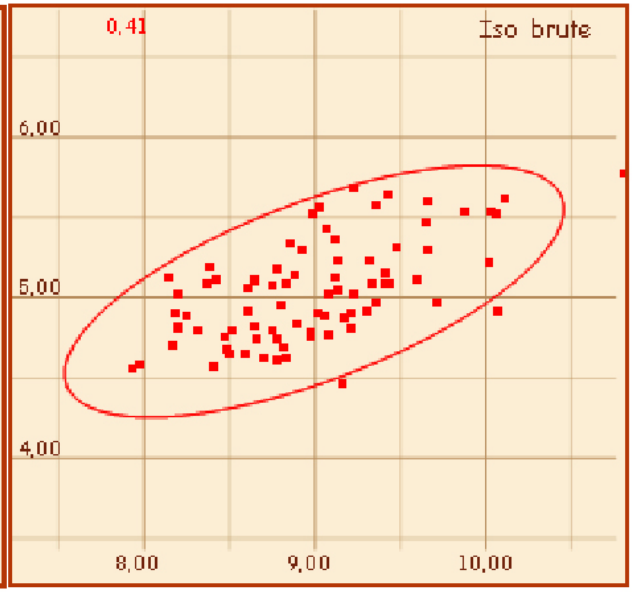


Piximètre: (N=80)
(7,9)8,3-9,7(10,8) × (4,5)4,7-5,5(5,8) µm
Q = (1,6)1,7-1,9(2,1) ; Me = 9 × 5 µm ; Qe = 1,8

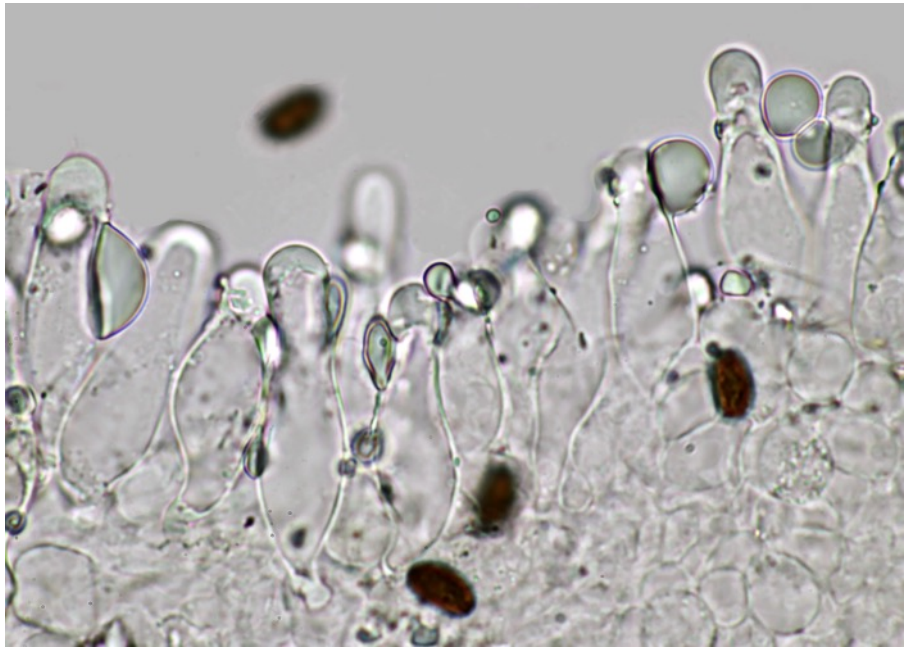
Melzer : sur base d'une récolte allemande (G) Mecklenburg-Vorpommern, Rehna, 31.10.06, leg. T. Richter (AM1755). Spores : 8,7-10,5 x 5-6,2 µm, av. 9,5-9,7 x 5,4 µm, av. Q=1,76-1,80. In water reddish brown, in ammonia medium brown, in KOH dirty brown. Rarely phaseoliform. Usually with large, conspicuous oil drops. Germ pore small, but distinctly, apiculus tiny.

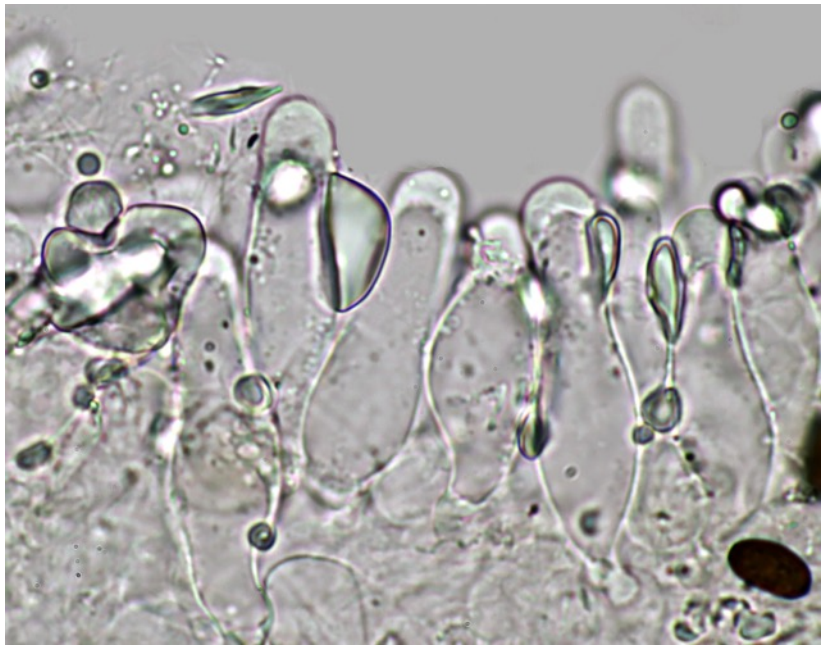
Smith: 8-10 x 5-6 µm

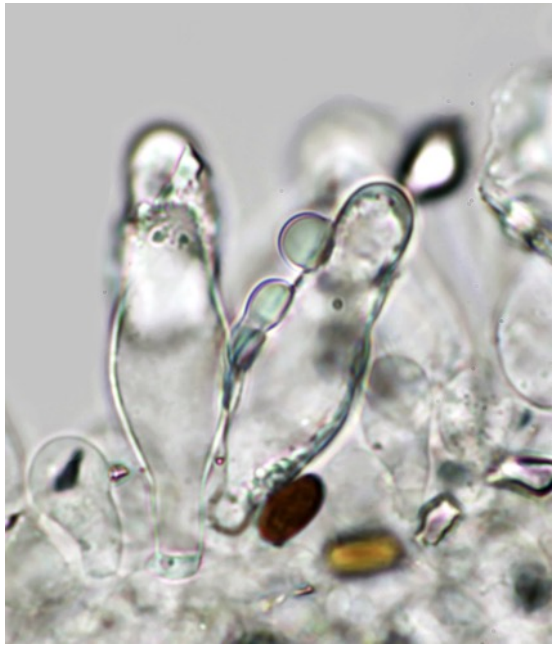


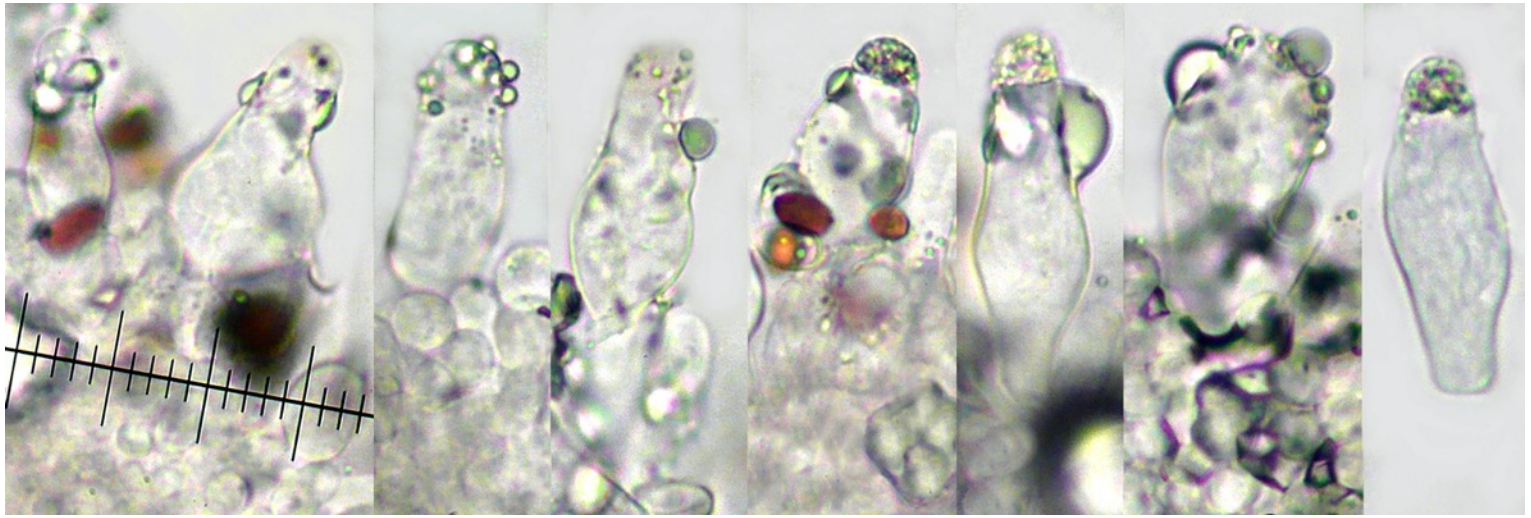


Cheilocystides mesurant (33,5) 36 - 55,8 (61,2) × (8,6) 10,2 - 16 (17,4) µm ; Me = 45,7 × 13,4 µm ; polymorphes, sub-lagéniformes à utriformes, souvent et abondamment **incrustées** au sommet, recouvertes de **dépôts mucoïde**s verdissant dans l'ammoniaque.



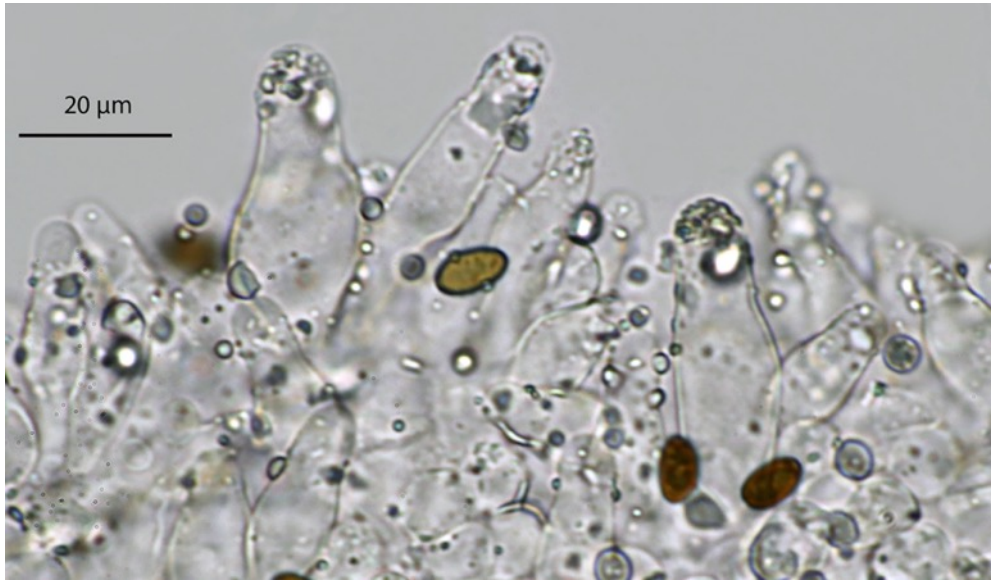


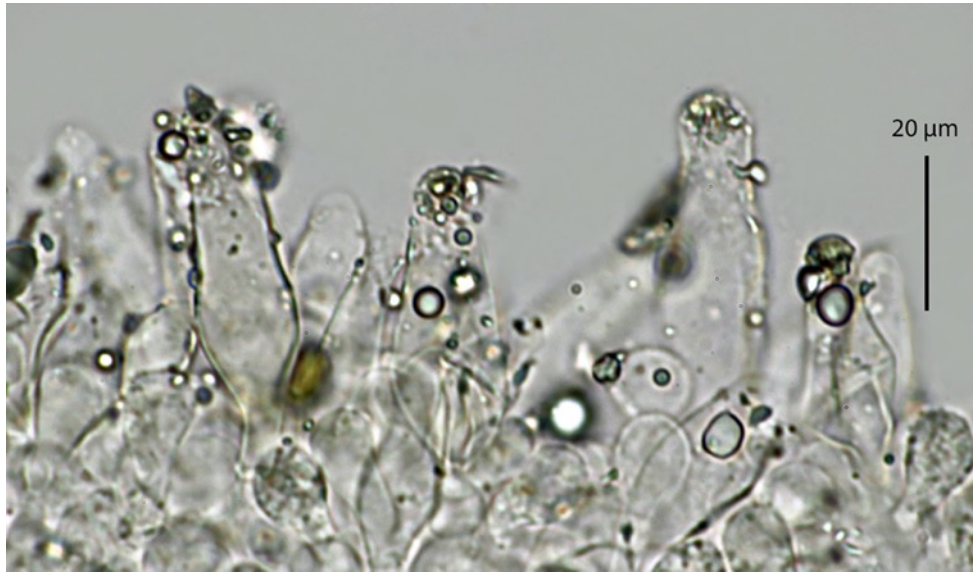


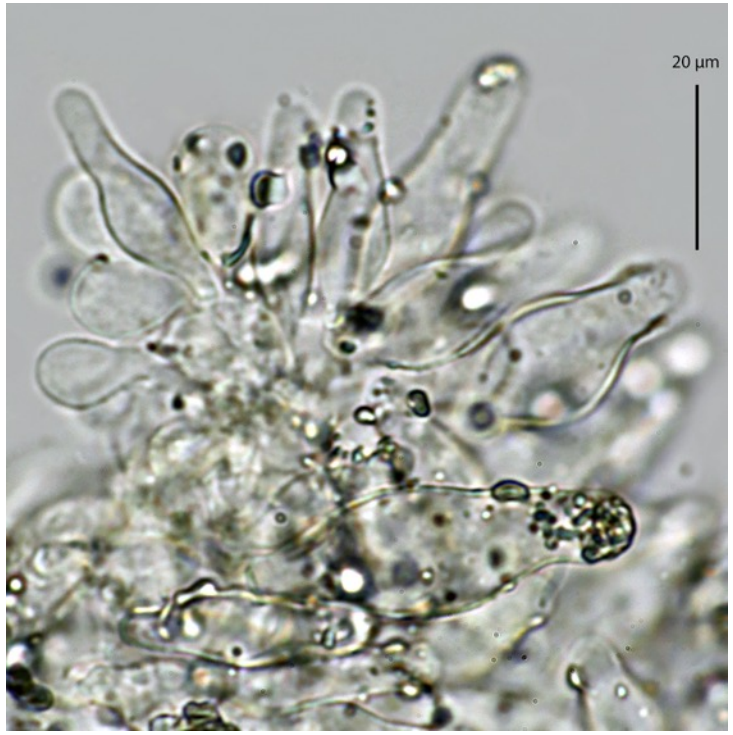


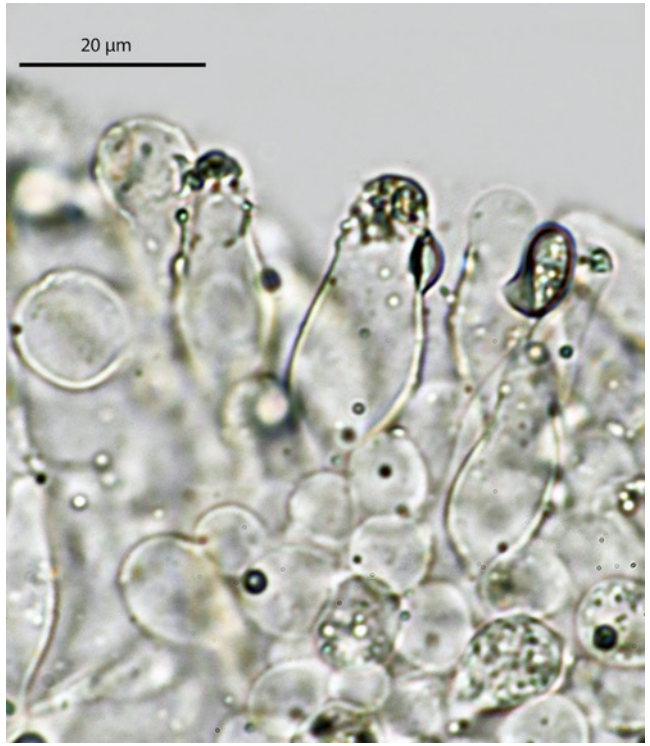
Ci-dessus cheilocystides de la récolte de P. Tanchaud nettement incrustées au sommet

Cheilocystides incrustées au sommet et couvertes de gouttes mucoïdes dans NH4OH 10%

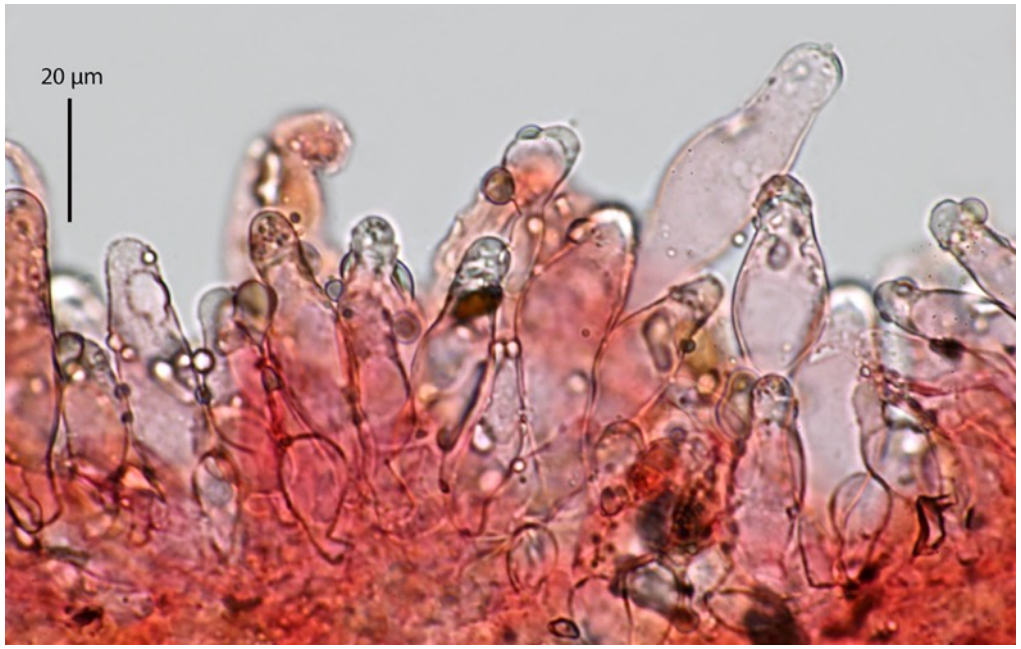


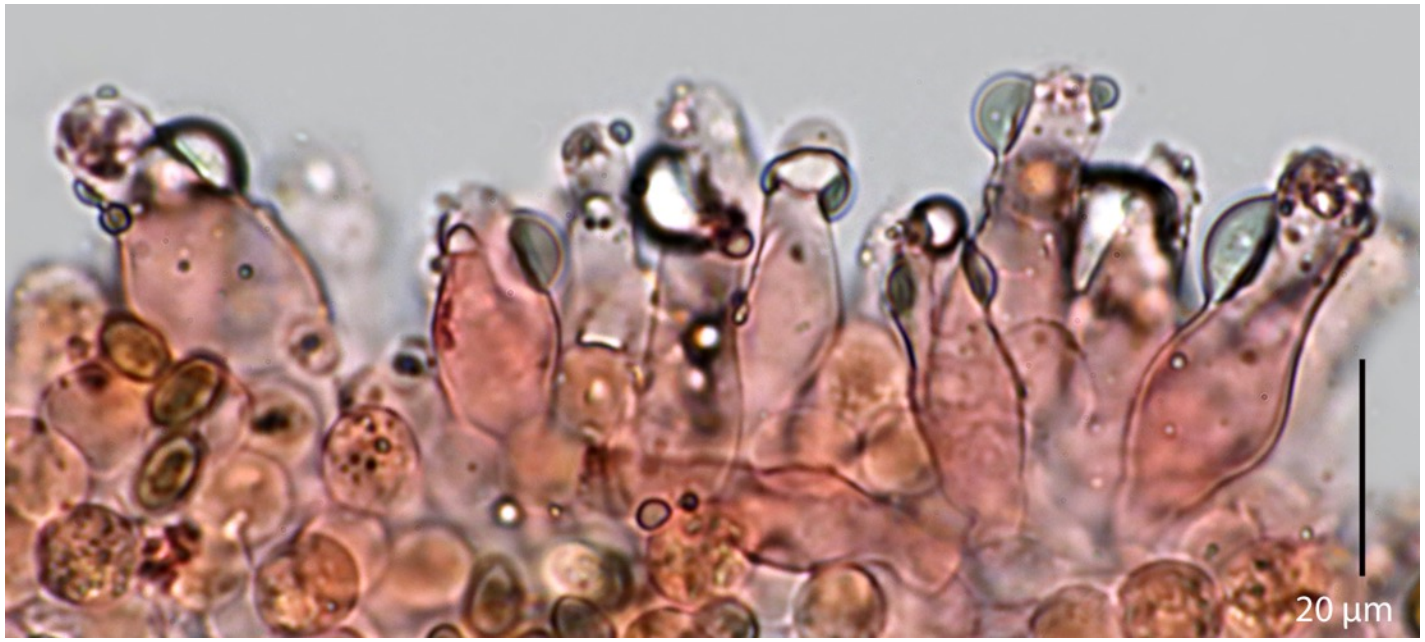


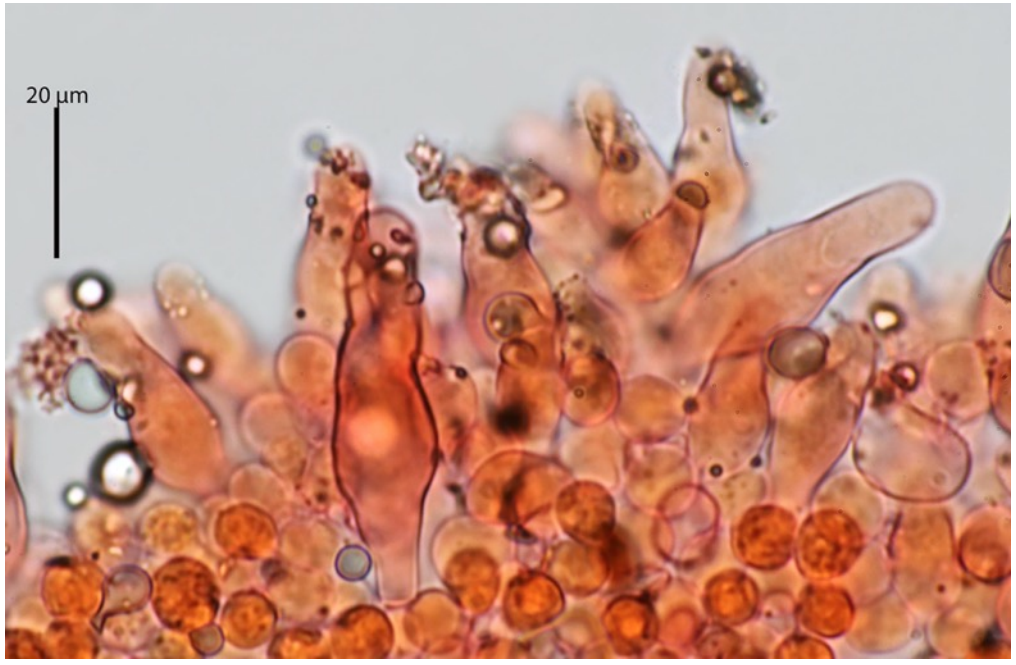


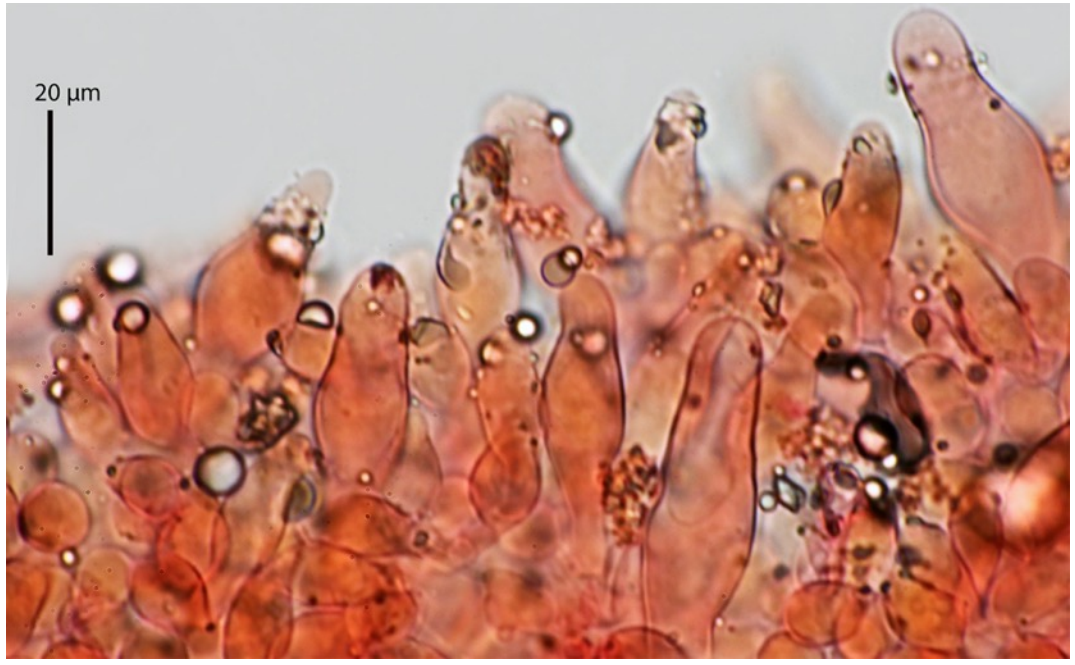


Incrusted cheilocystidia covered with drops

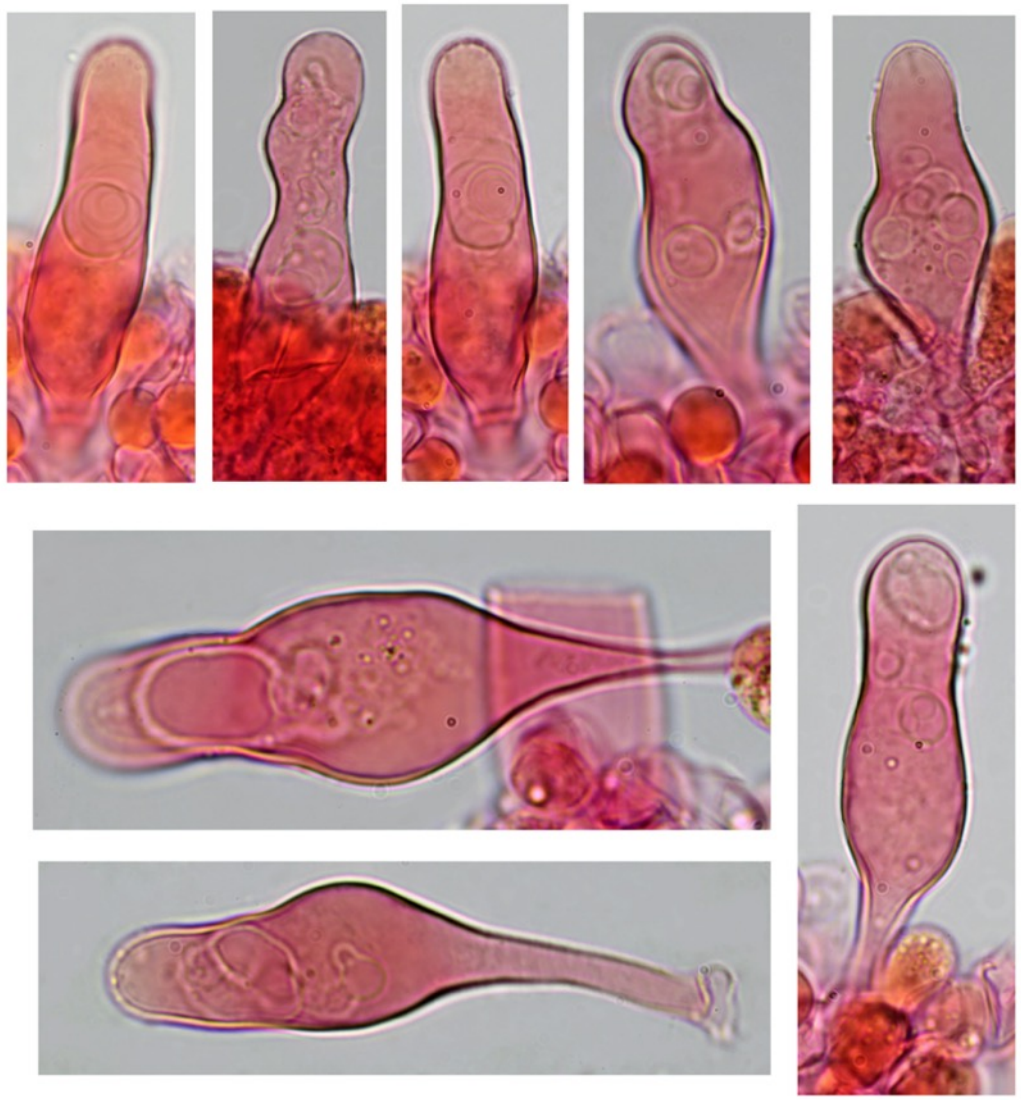


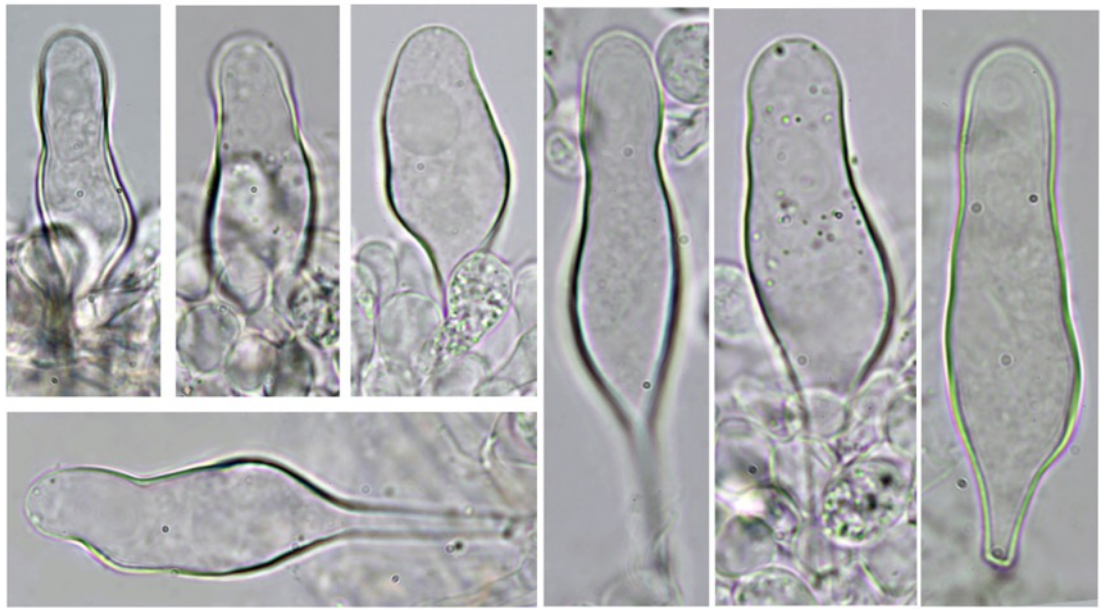


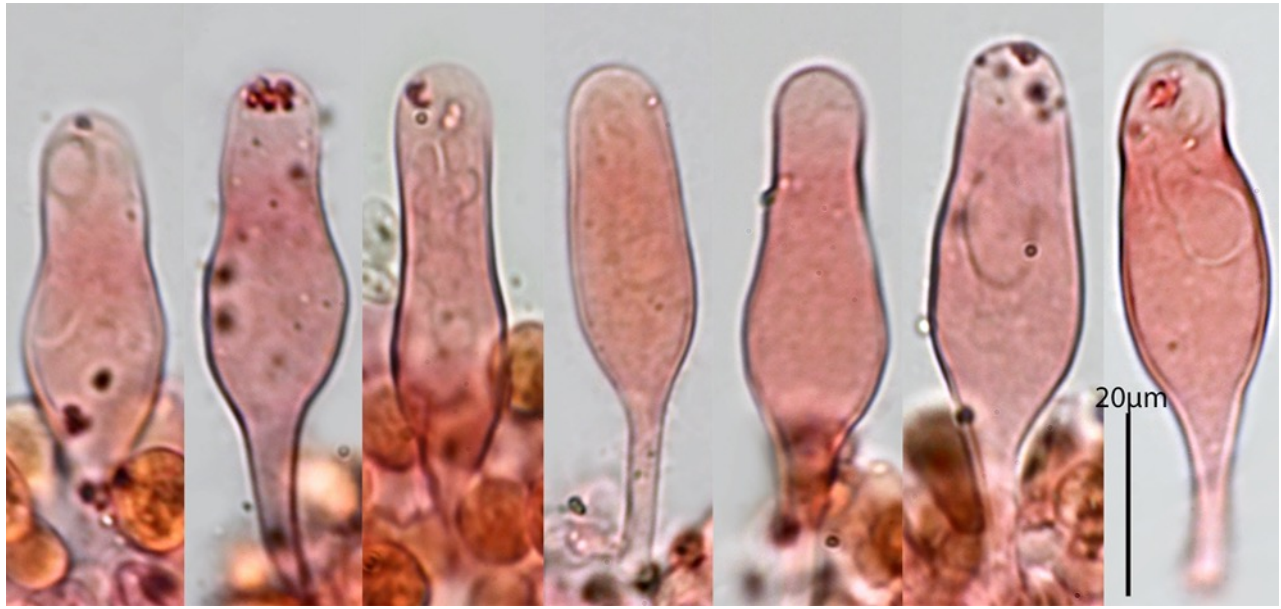




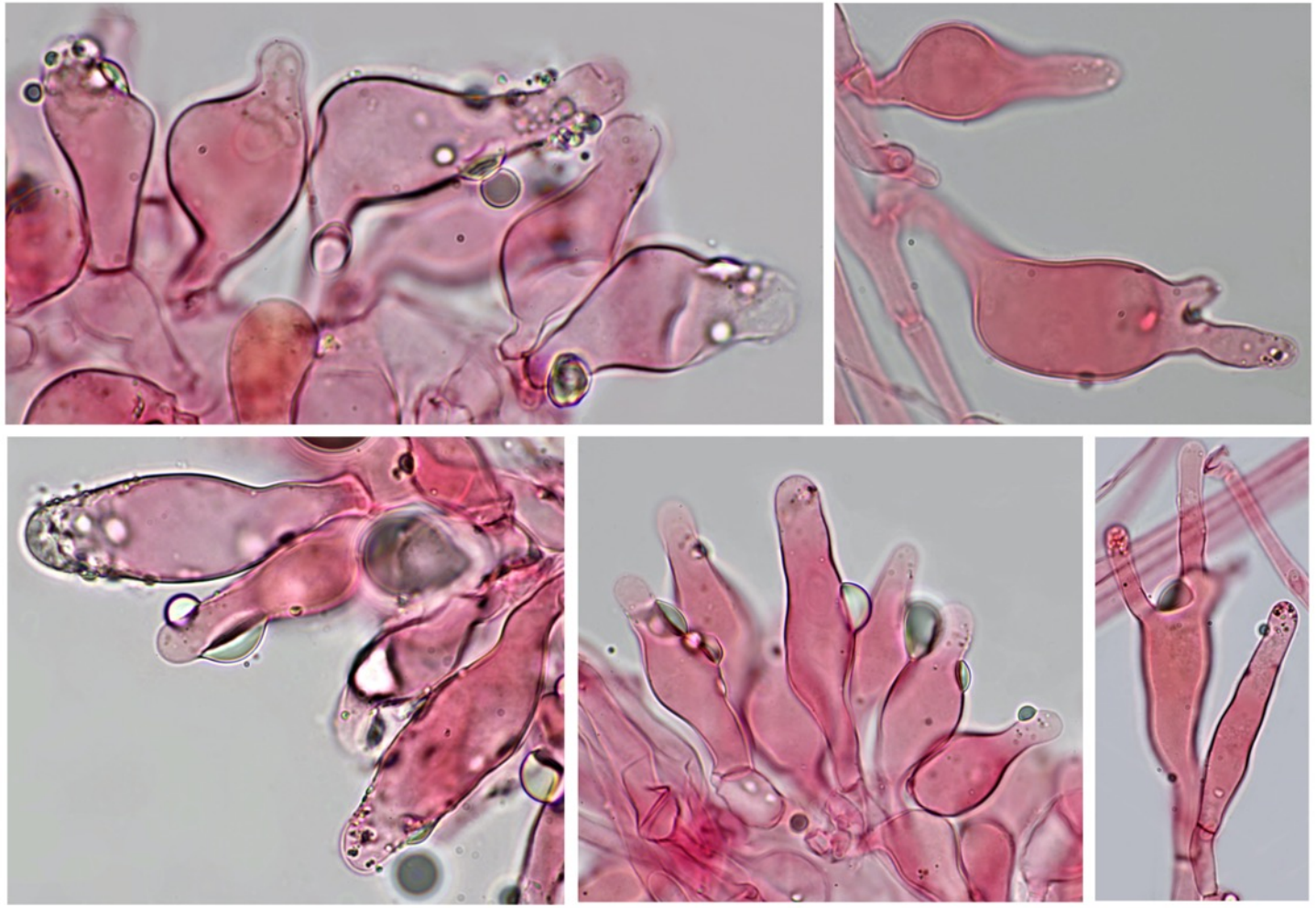
Pleurocystides mesurant (43,1) 47 - 62,6 (66,1) × (13,2) 13,7 - 17,5 (18,6) µm ; Me = 55,2 × 15,4 µm ; très nombreuses, lagéniformes, lagéno-utriformes, cylindriques ou ventrues, à paroi fine ou modérément épaissie, à col court et large, à sommet largement obtus, souvent longuement pédicellées et contenant ou tapissée parfois d' incrustations réfringentes au sommet.







Caulocystides analogues aux pleuro- et cheilocystides ou versiformes, parfois fourchues, au sommet souvent épaissi contenant également des granulations cristallines réfringentes et exsudant, sur le frais, de nombreuses gouttes mucoïdes colorées en vert dans l'ammoniaque 10%.



Voile prélevé sur un primordia
43 - 94,38 (94,4) × (6,3) 6,32 - 6,9 µm

DNA extraction, amplification and sequencing of the fungus was performed by Alvalab (Oviedo, Spain). The phylogenetic analysis was done by Dieter Wächter (Thiersheim, Germany). The genomic DNA was extracted from dried and fresh fruiting bodies. Amplification of the ITS region was performed with the ITS4 primer [1]. The initial base calling was done with FinchTV [2]. The nucleotide sequences were checked manually for errors, as well as the base calling at unsafe regions (trails, low confidence scores, stutters and polymorphs) on the basis of existing sequences of the /saponacea s.l. clade by divergence matrix and corrected if necessary. In the present case only a trimming of the trails was necessary. The following molecular phylogenetic markers were used for the phylogenetic analysis ITS1 (Internal Transcribed Spacer 1), 5.8S (5.8S rRNA Gene), ITS2 (Internal Transcribed Spacer 2), LSU (Large Subunit 28S rRNA Gen), β-tub (exons of the β-tubulin gene), ef-1α (exons of the ef-1α gene). The nucleotide sequences for the tree inference were taken from NCBI [3] and Unite [4] (essential ones of the /saponacea s.l. clade see Table 1). Region boundaries for the ITS- and LSU-region were carried out with ITSx [5] and HMMER [6] including the databases. As outgroup, the sequence sets of the most closely related clade of the ingroup were used, i.e. the /stridvallii clade, the /arenosa s.l. clade and the /gordonii s.l. clade. Due to the rapidly evolving, indel-rich areas of the ITS region, it can only be aligned veridical by using an iterative multigene-guide tree. The initial alignment of the ITS region was performed with Mafft [7] using the FFT-NS-2 method. The initial alignments of the LSU-, β-tub and ef-1α genes was carried out using E-INS-method. The indel matrices for the ITS and LSU regions were each coded with SeqState [8] using the SIC = "Simple Indel coding" [9] method. After each alignment step, an ML analysis with RAxML [10] (model: GTRCAT, refining under GTR+G for DNA, GTR2+G with acquisition bias correction according to Lewis [11] for indel partitions) was carried out and the resulting best tree was used as a guide tree for the refinement of the ITS1 and ITS2 MSA. The iterative alignments were done with Prank [12], whereby the switches -once and -uselogs were set. Tracing values were recorded, evaluated statistically and thus the end of the iteration loop of the alignment was determined. The partitioning of all alignments and the indel matrices as well as the model selection for the DNA alignments was done with Partitionfinder [13]. For the final partitioning, the guide tree of the last iteration step was used. As information criterion the Bayesian Information Criterion (BIC) [14] used was after comparison with the Corrected Akaike Information Criterion (AICc) [15] and evaluation with respect to over- or under-partitioning. The partitioning scheme of the final phylogeny was:

- DNA-partition 1: ITS1 + ITS2
- DNA-partition 2: 5.8S
- DNA-partition 3: LSU + β-tub-Codon 1
- DNA-partition 4: β-tub Codon 2 + ef-1α Codon 2
- DNA-partition 5: β-tub Codon 3 + ef-1α Codon 3
- DNA-partition 6: ef-1α Codon 1
- Binary partition (gap matrices): ITS1 + ITS2 + LSU

The final maximum likelihood analysis was done with RAxML 8.2.10 [10]. For all DNA partitions, the GTR substitution matrix [16] under the CAT model [10] was used. The final optimization took place under gamma distribution [10]. For the binary partitions, the "Two State Time-Reversible Model" with acquisition bias correction [11] was used. 1000 ML bootstrap inferences were calculated. Of these, 1000 trees were sampled and the best tree was labeled with the ML bootstrap support values and collapsed to the ML bootstrap value of 50%. The phylogram in Fig. 1 was edited with Treegraph [17]. The Outgroup has been collapsed for a better view.

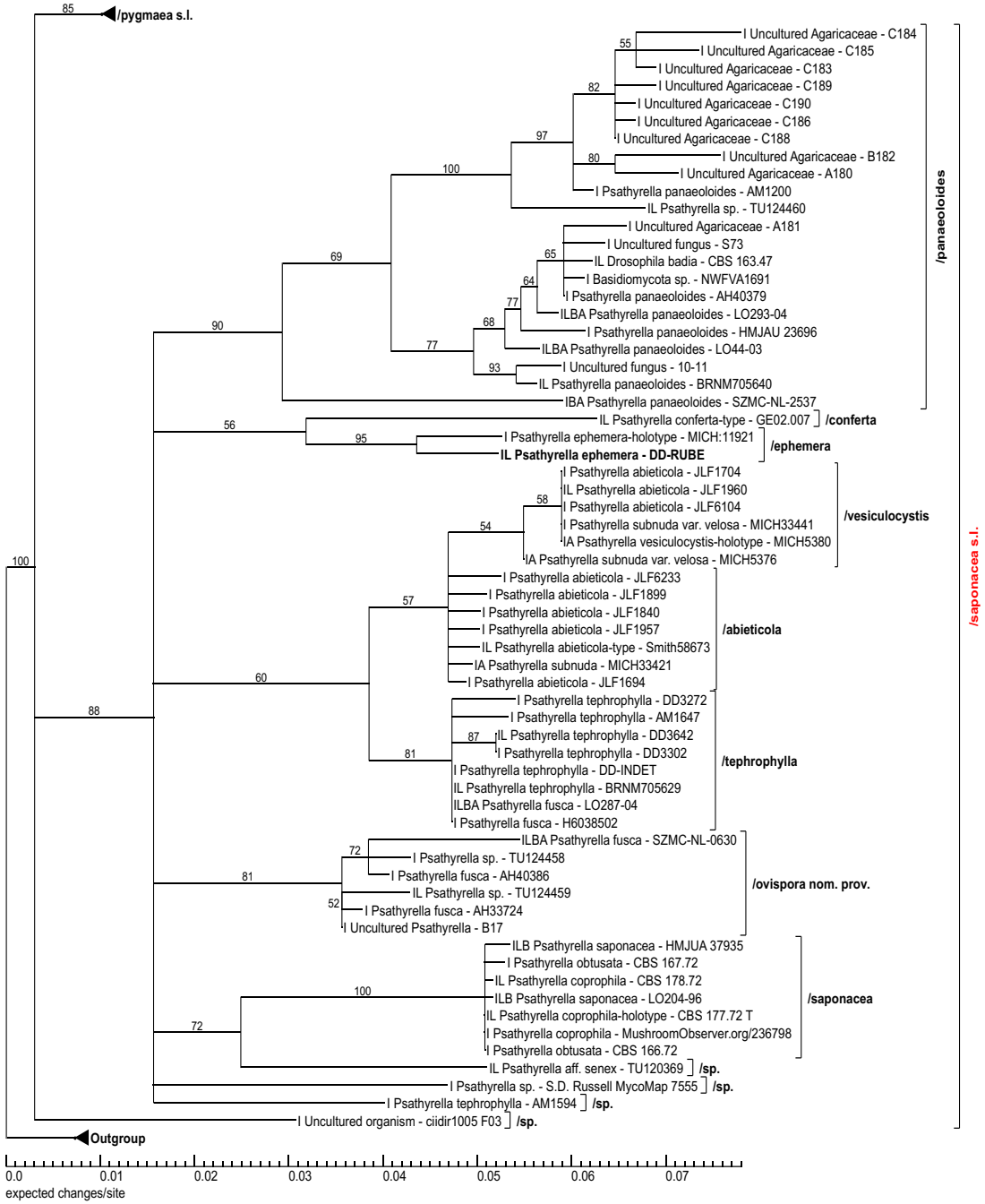


Fig. 1 50% collapsed maximum likelihood consensus phylogram. The values on the branches are ML bootstrap values. Abbreviations: I: ITS region, L: LSU region, B: β-tubulin region, A: ef-1α region.

Table 1 List of relevant sequences used in this publication

Species	Ref.-ID	ITS	LSU	β-Tub	ef-1α
Uncultured Agaricaceae	C189	AM076648.1			
Uncultured Agaricaceae	C190	AM076649.1			
Uncultured Agaricaceae	C188	AM076647.1			
Uncultured Agaricaceae	C186	AM076645.1			
Uncultured Agaricaceae	C185	AM076644.1			
Uncultured Agaricaceae	C183	AM076642.1			
Uncultured Agaricaceae	C184	AM076643.1			
Uncultured Agaricaceae	A180	AM076639.1			
Uncultured Agaricaceae	B182	AM076641.1			
Psathyrella panaeoloides	AM1200	MK045663.1			
Psathyrella sp.	TU124460	UDB028411	UDB028411		
Uncultured Agaricaceae	A181	AM076640.1			
Psathyrella panaeoloides	LO293-04	KC992894.1	KC992894.1	KJ664874.1	KJ732783.1
Uncultured fungus	S73	FJ820561.1			
Psathyrella panaeoloides	HMJAU 23696	MG734733.1			
Psathyrella panaeoloides	LO44-03	DQ389719.1	DQ389719.1	KJ664873.1	KJ732782.1
Uncultured fungus	10-11	KM374327.1			
Psathyrella panaeoloides	BRNM705640	AM712271.1	AM712271.1		
Psathyrella panaeoloides	SZMC-NL-2537	FM878022.1		FN396317.1	FM897255.1
Drosophila badia	CBS 163.47	MH856198.1	MH867725.1		
Basidiomycota sp.	NWFVA1691	KU712241.1			
Psathyrella panaeoloides	AH40379	MF966501.1			
Psathyrella conferta	GE02.007	KC992890.1	KC992890.1		
Psathyrella ephemera	MICH:11921	NR_161023.1			
Psathyrella ephemera	DD-RUBE	MN493776.1	MN493779.1		
Psathyrella vesiculocystis	MICH5380	MF326007.1			MF521772.1
Psathyrella abieticola	JLF6104	MK996301.1			
Psathyrella abieticola	JLF1960	MK996300.1	MN031141.1		
Psathyrella subnuda var. velosa	MICH33441	MF326006.1			
Psathyrella abieticola	JLF1704	MK996294.1			
Psathyrella subnuda var. velosa	MICH5376	MF326005.1			MF521773.1
Psathyrella abieticola	Smith58673	KC992891.1	KC992891.1		
Psathyrella abieticola	JLF6233	MK996302.1			
Psathyrella abieticola	JLF1840	MK996296.1			
Psathyrella abieticola	JLF1957	MK996299.1			
Psathyrella abieticola	JLF1694	MK996292.1			
Psathyrella subnuda	MICH33421	MF326000.1			MF521778.1
Psathyrella abieticola	JLF1899	MK996298.1			
Psathyrella fusca	H6038502	UDB021182			
Psathyrella fusca	LO287-04	KC992892.1	KC992892.1	KJ664870.1	KJ732779.1
Psathyrella tephrophylla	BRNM705629	AM712270.1	AM712270.1		
Psathyrella tephrophylla	DD3642	MK577902.1	MK577901.1		
Psathyrella tephrophylla	DD3302	MK577904.1			
Psathyrella tephrophylla	AM1647	MK087035.1			
Psathyrella tephrophylla	DD3272	MK577903.1			
Psathyrella tephrophylla	DD-INDET	MK583508.1			
Psathyrella tephrophylla	AM1594	MK045664.1			
Psathyrella fusca	AH40386	MF966503.1			
Psathyrella fusca	AH33724	MF966497.1			
Psathyrella sp.	S.D. Russell MycoMap 7555	MK532815.1			
Psathyrella sp.	TU124458	UDB028409			
Psathyrella fusca	SZMC-NL-0630	FM878030.1	FM876288.1	FN396270.1	FM897217.1
Uncultured Psathyrella	B17	JX135080.1			
Psathyrella sp.	TU124459	UDB028410	UDB028410		
Psathyrella saponacea	LO204-96	DQ389717.1	DQ389717.1	KJ664871.1	
Psathyrella coprophila	CBS 177.72 T	NR_160137.1	NG_064095.1		
Psathyrella saponacea	HMJUA 37935	MH155965.1	MH155960.1	MH161167.1	
Psathyrella obtusata	CBS 166.72	MH860427.1			
Psathyrella coprophila	CBS 178.72	MH860434.1	MH872164.1		
Psathyrella obtusata	CBS 167.72	MH860428.1			
Psathyrella coprophila	MushroomObserver.org/236798	MG983995.1			
Psathyrella aff. senex	TU120369	UDB024759	UDB024759		
Uncultured organism	ciidir1005_F03	JN660650.1			

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