Description d'une récolte insolite de Psathyrella lacuum Huijsman 1955

in Fungus 25: 39

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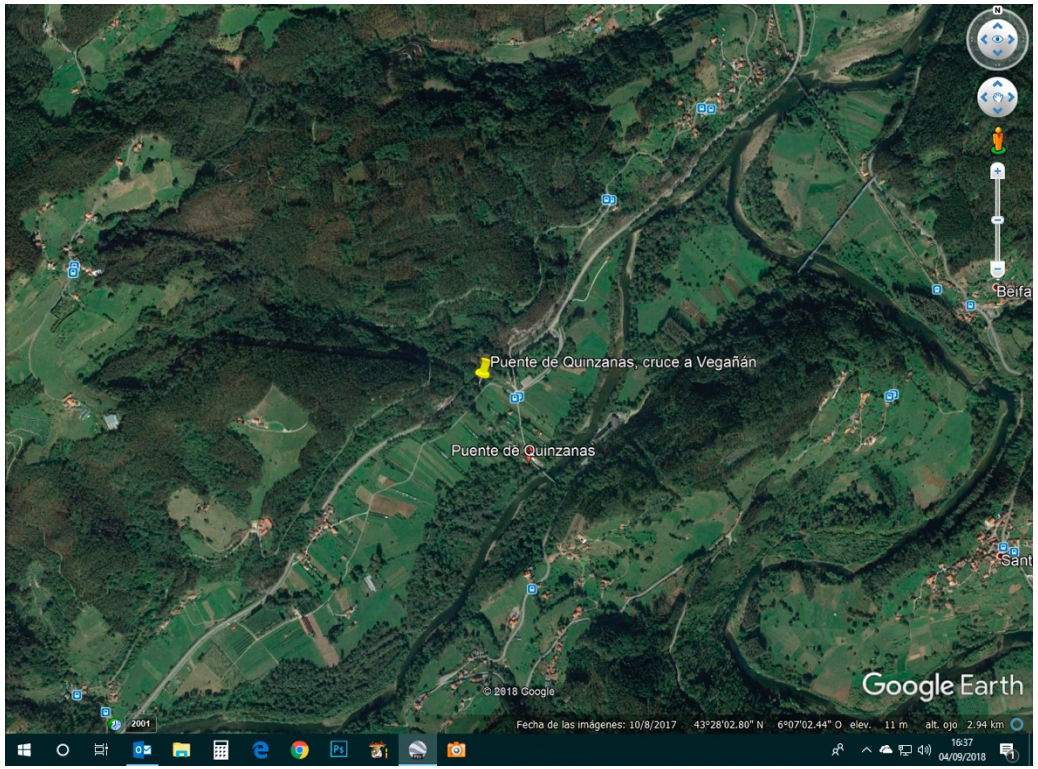
Une dizaine d'exemplaires, à différents stades d'évolution ont été récoltés le 31/08/2018 par le second auteur en Espagne au lieu dit :
Puente de Quinzanas, cruce a Vegañán (Pravia-Asturias).

La récolte a été réalisée en milieu boueux, en présence de *Paratrichophaea parvispora* sur fragments de bois et feuilles pourrissantes au sol, *d'Alnus glutinosa et salix sp*. L'endroit est régulièrement submergé suite aux débordements de la rivière Nalón passant à proximité.



Elle avait été éditée pour identification sur le forum de discussion Mycologia Europaea et à cette occasion Pietro Voto avait suggéré fort à propos que cette récolte puisse correspondre à *Psathyrella lacuum* malgré l'écologie tout a fait inhabituelle.

A notre connaissance, cette rare espèce n'avait été décrite jusqu'à ce jour que comme une espèce « indoor » se développant sur Yuca palm et débris de coco.

Son identification a été confirmée après analyse biomoléculaire de deux spécimens pour lesquels les séquences ITS, LSU et TEF ont été étudiées par le troisième auteur.

GPS position: **43.466335, -6.115580 = 43.466335 N 6.115580 W**

Close-up des primordia

Photo in situ – Leg : Enrique Rubio – Voucher : ERD7651 = DD-LACU

Description macroscopique

Chapeau mesurant de 3 à 30 mm de diamètre, brun foncé et conico-campanulé chez les sujets jeunes, devenant au cours de la croissance brun-noisette, progressivement convexe à plan-convexe et finement strié jusqu'au 2/3 du rayon, à partir de la marge. Hygrophane, il décolore, pour finalement prendre une tonalité crème.

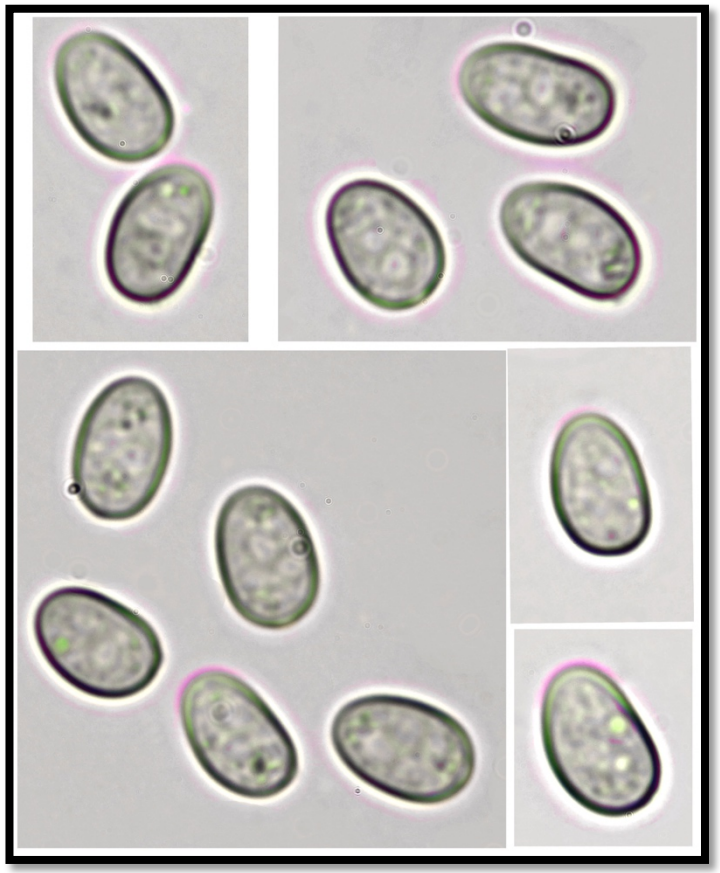
Stipe mesurant 9-30 mm x 1-2,5 mm, blanchâtre, cylindrique, creux, fragile, pruineux au sommet, lisse dans les 2/3 inférieurs mais finement fibrilleux sur les primordia suite à la persistance de fibrilles vélaires. Il est souvent un peu courbé à la base qui est modérément dilatée, strigueuse et teintée de beige.

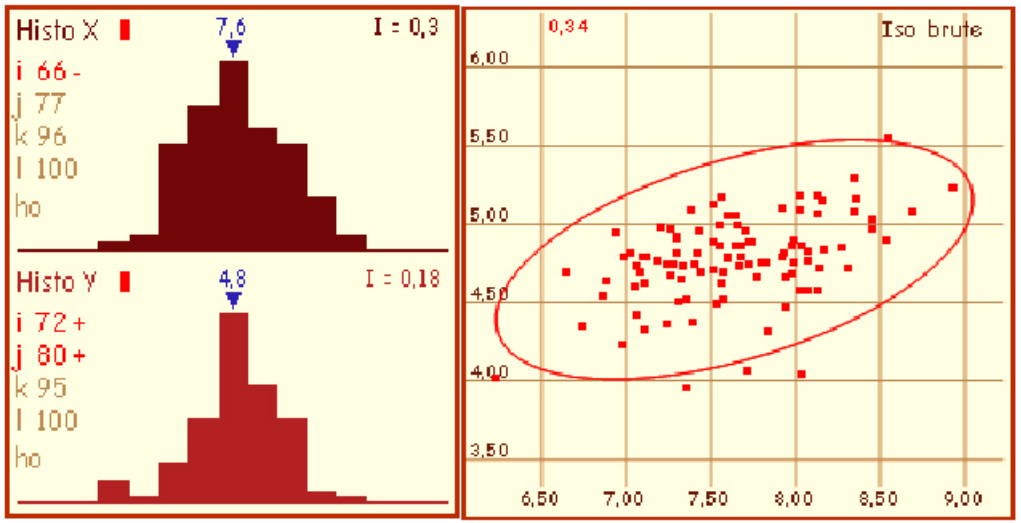
Lames droites à légèrement ventrues, largement adnées, nombreuses et serrées, très pâles et crème chez les exemplaires jeunes, devenant légèrement beiges ; l'arête blanchâtre est fimbriée.

Voile constitué de fibrilles blanchâtres. Sur les primordia, il est abondant et apparaît sous forme de fibrilles éparses, formant au niveau de la marge un filet aranéeux qui relie la marge au stipe. Rapidement volatile, il ne persiste que sous forme de fibrilles disséminées, mais plus présentes au niveau de la marge du chapeau des exemplaires adultes.

Chair fine, hyaline. **Odeur** sans particularité. **Saveur** non testée.

Basides : 14-19 x 7-10 µm, clavées, tétrasporiques.
Spores : 6,2)7,1-8,3(8,9) × (4)4,4-5,1(5,5) µm, très pâles, translucides, d'un jaune très léger dans le NH_4OH, et à peine un peu plus grisâtre dans le KOH à 5% ; paroi légèrement épaissie, en particulier au sommet qui est dépourvu de pore germinatif ; ellipsoïdes et ovoïdes de face, légèrement phaséoliformes de profil.
Boucles: présentes

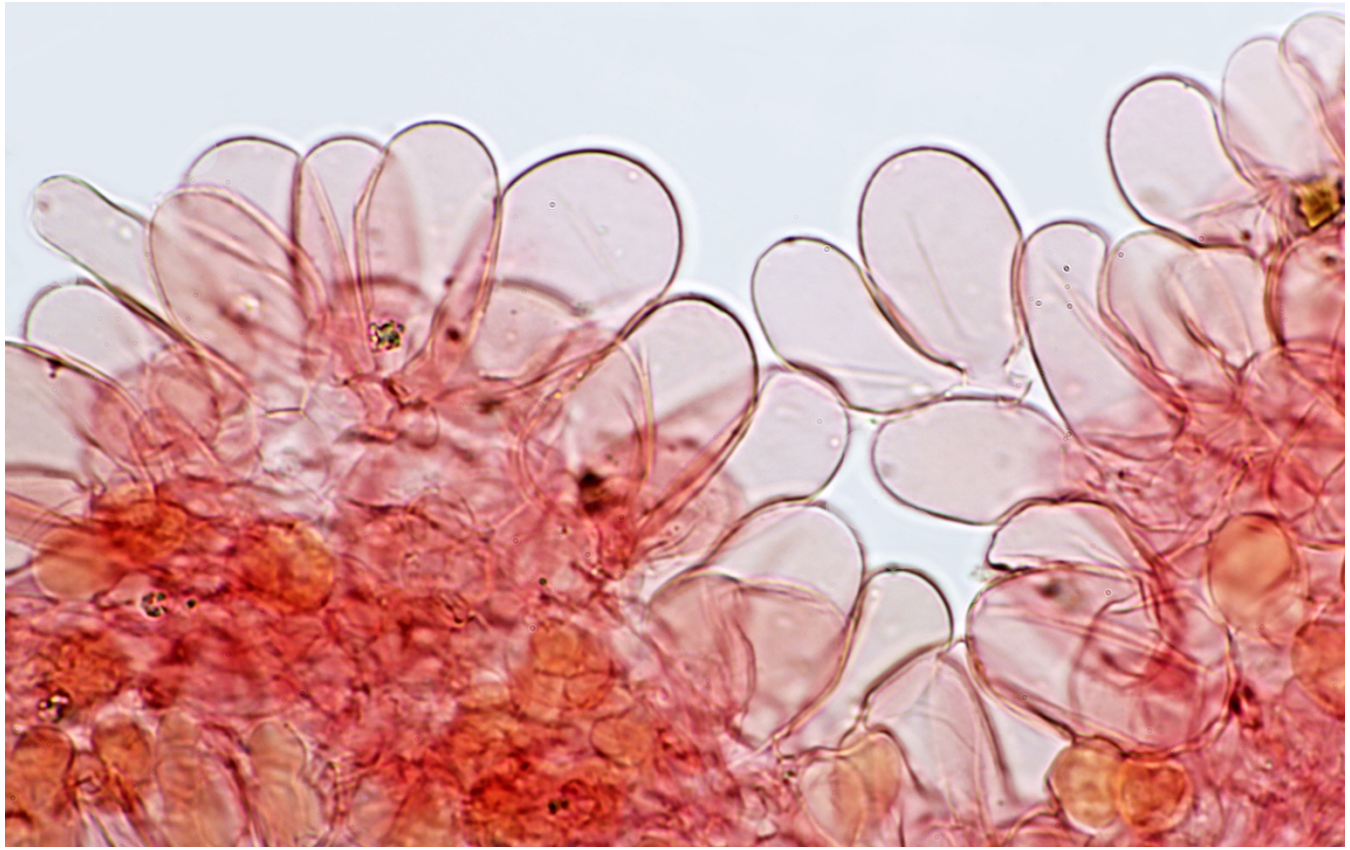


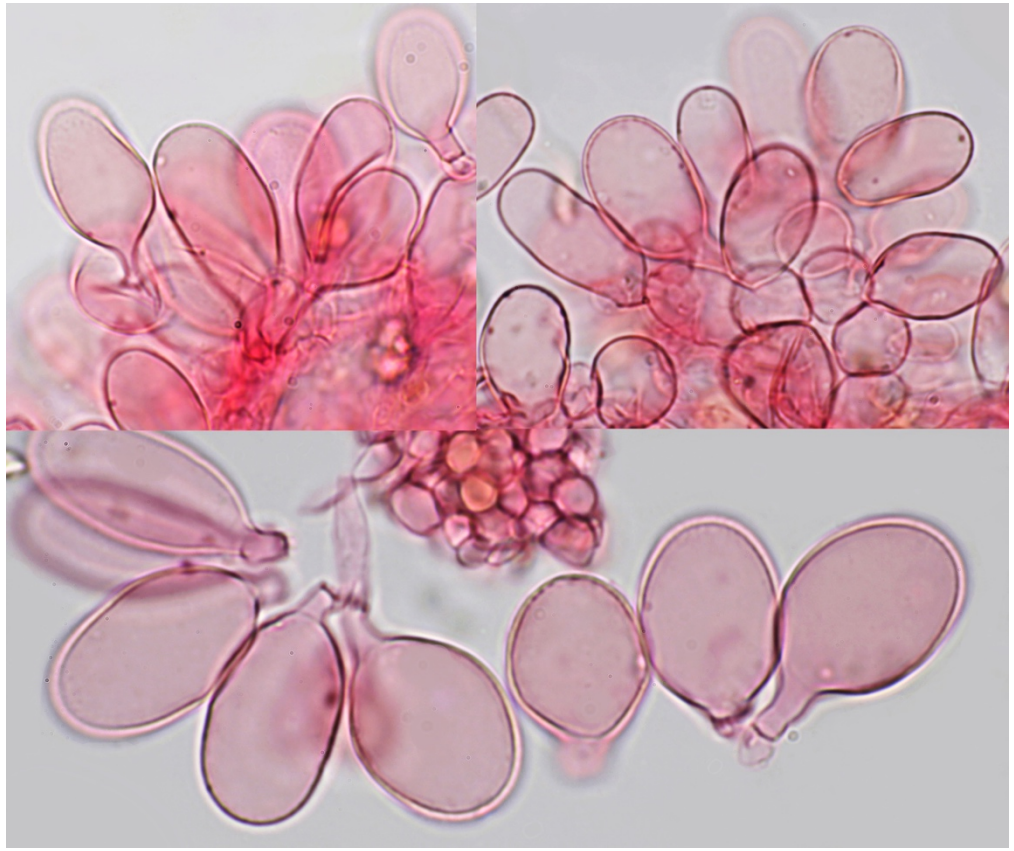


Mesures et diagrammes réalisés avec piximètre
Spores:
(6,2)7,1-8,3 (8,9) × (4)4,4-5,1(5,5) µm
Q = (1,4)1,5-1,7(2) ; N = 100
Me = 7,6 × 4,8 µm ; Qe = 1,6

Toutes les photos des caractères microscopiques ont été réalisées par le premier auteur.

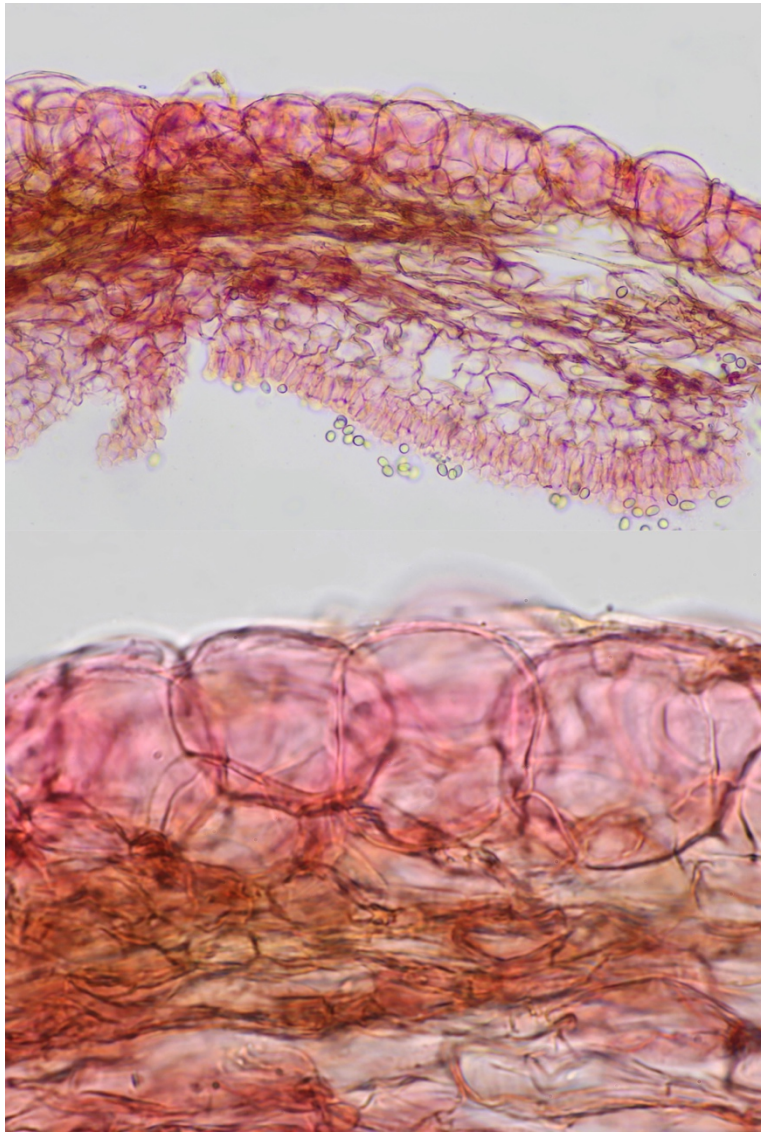
Arête non surlignée, densément couverte d'une palissade de **cellules marginales (= paracystides) clavées et sphéropédonculées,** dont la paroi fine est parfois légèrement teintée, mesurant 20-36 x 11-20 µm (N=20).
Les **cheilocystides** sont rares, voire inexistantes, et semblent correspondre à de simples variations de forme des cellules marginales.
Pleurocystides absentes.
Médiostrate légèrement teintée de beige.



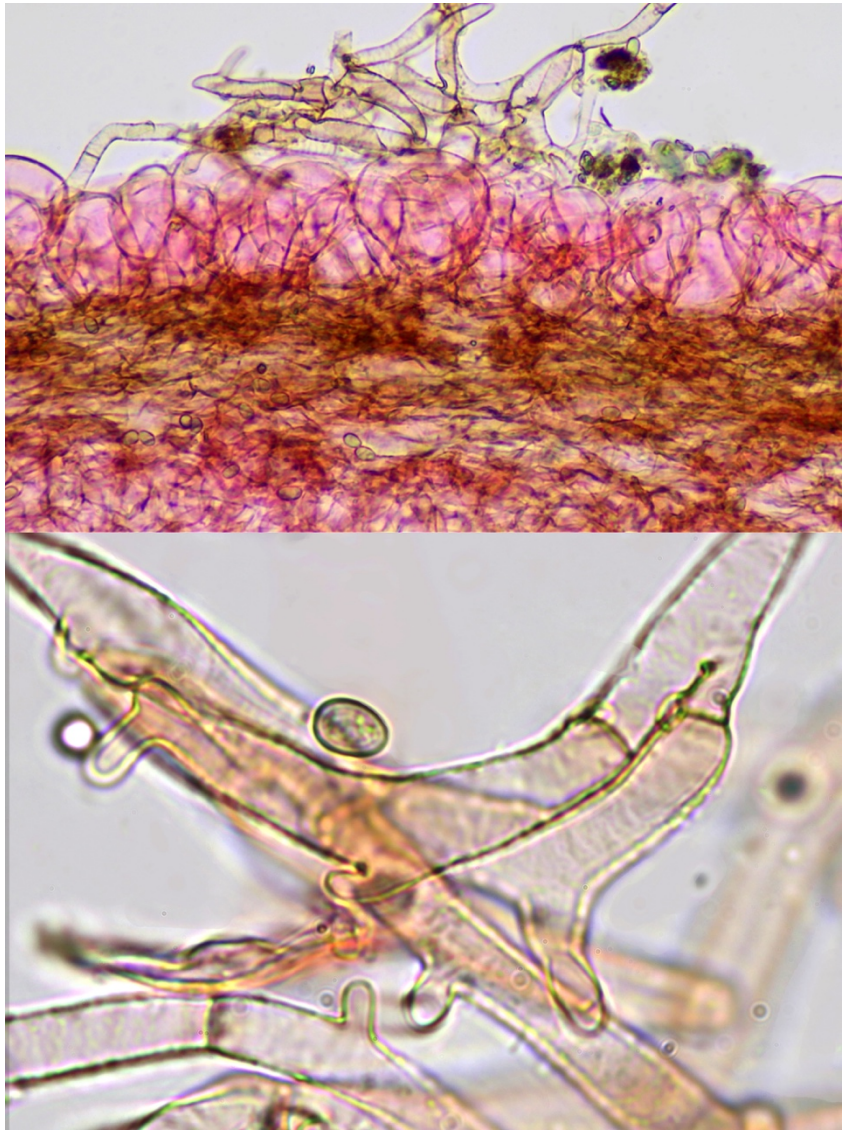


Photos de l'arête

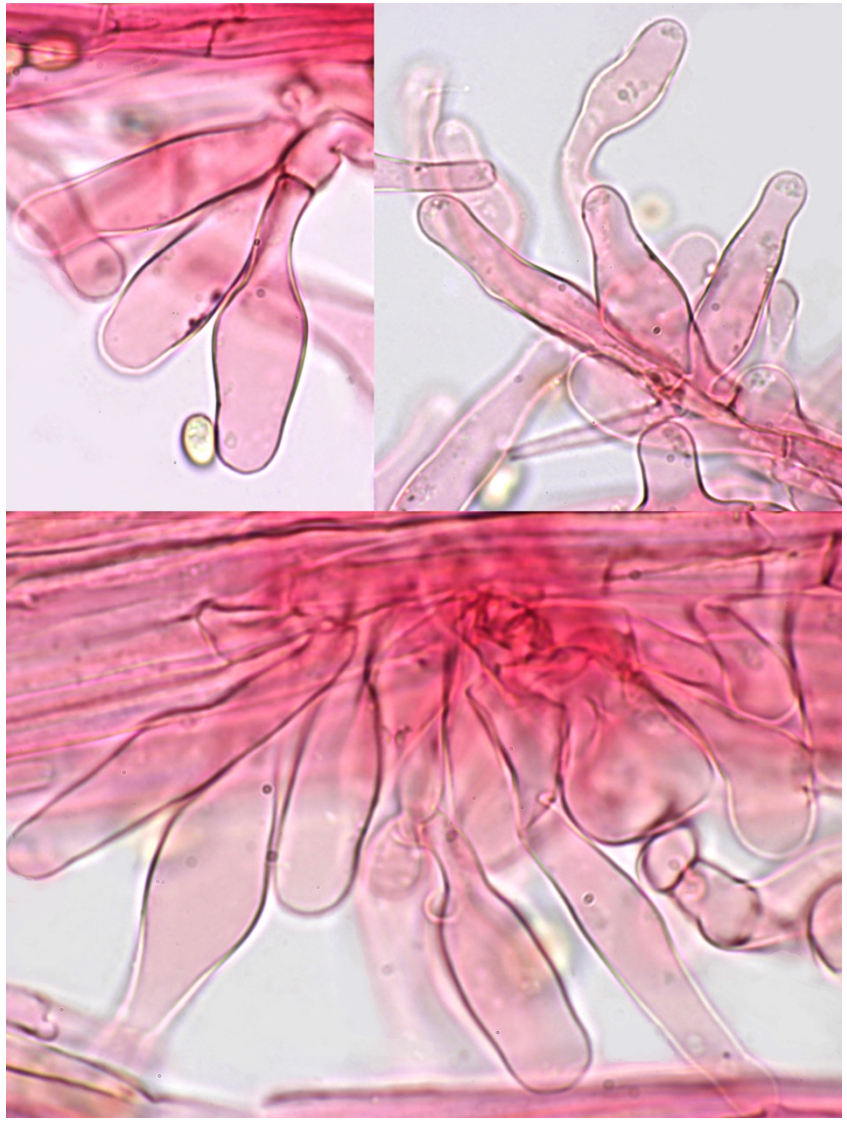
Pileipellis : un hyméniderme constitué d'une couche de cellules globuleuses clavées et sphéropédonculées.
Pileitrame : hyphes cylindriques fortement pigmentées.



Voile : hyphes cylindriques ramifiées, à pigmentation intracellulaire brun-jaune, incrustées et partiellement bouclées.



Caulocystides abondantes, d'aspect variable, clavées, lagéniformes, utriformes, longuement subcylindriques.



Etude biomléculaire :

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 fruiting bodies. Amplification of the ITS region was performed with the ITS4 primer [1], amplification of the LSU region was performed with the LR5 primer [2], amplification of the ef-1α region was performed with the EF1-1567R primer [3]. The initial base calling was done with FinchTV [4]. 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 /*candolleana* s.l.-clade by divergence matrix and corrected if necessary. In the present case only a trimming of the trails and som minor corrections were 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 [5] and Unite [6] (essential ones of the /*candolleana* s.l.-clade see Table 1). Region boundaries for the ITS- and LSU-region were carried out with ITSx [7] and HMMER [8] including the databases. As outgroup, the sequence sets of the most closely related clades of the ingroup were used, i.e. from other /*Psathyrella* taxa the /*Cystoagaricus*, /*Typhrasa* and /*Kauffmania*-clades. 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 [9] using the FFT-NS-2 method. The initial alignments of the LSU-, β-tub and ef-1α genes was carried out using E-INS-i method. The indel matrices for the ITS and LSU regions were each coded with SeqState [10] using the SIC = "Simple Indel coding" [11] method. After each alignment step, an ML analysis with RAxML [12] (model: GTRCAT, refining under GTR+G for DNA, GTR2+G with acquisition bias correction according to Lewis [13] 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 [14], 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 [15]. For the final partitioning, the guide tree of the last iteration step was used. As information criterion the Bayesian Information Criterion (BIC) [16] used was after comparison with the Corrected Akaike Information Criterion (AICc) [17] and evaluation with respect to over- or under-partitioning. The partitioning scheme of the final phylogeny was:

- 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) 1: ITS1 + ITS2
- Binary partition (gap matrices) 2: LSU

The final maximum likelihood analysis was done with RAxML 8.2.10 [12]. For all DNA partitions, the GTR substitution matrix [18] under the CAT model [12] was used. The final optimization took place under gamma distribution [12]. For the binary partitions, the "Two State Time-Reversible Model" with acquisition bias correction [13] 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 [19]. The Outgroup and the upper /*candolleana* s.l. clades have been collapsed for a better view.

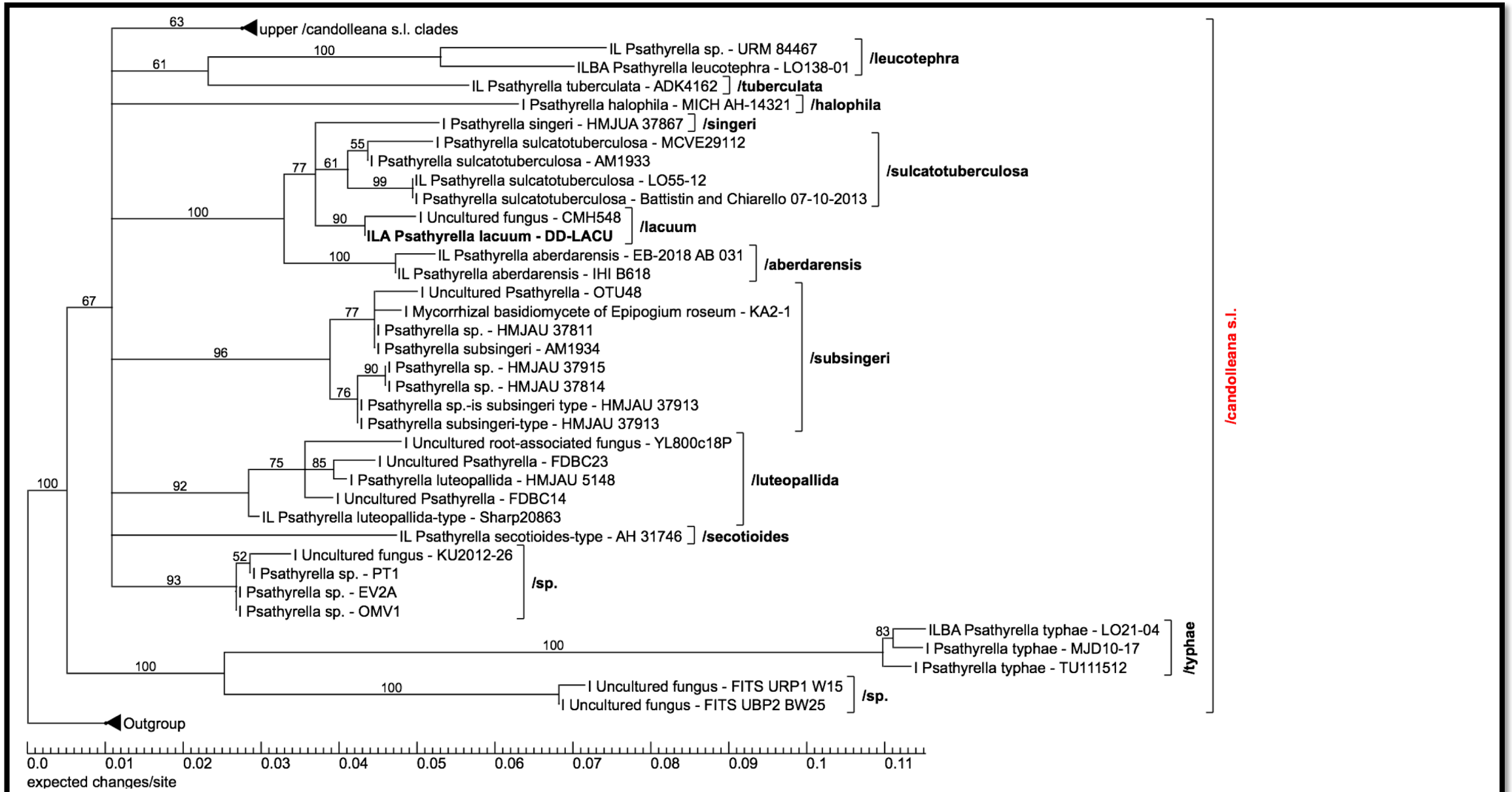


Fig 1 50% collapsed maximum likelihood consensus phylogram. The values on the branches are ML bootstrap values. Abbreviations: I: ITS region, L: LSU region, B: β-tubulin region, A: ef-1α region.

Table 1 List of relevant sequences used in this publication

Taxon	Voucher	ITS+5.8S+ITS 2	LSU	β-Tub	ef-1α
Psathyrella aberdarensis	EB-2018 AB 031	MH880928.1	MH880928.1		
Psathyrella aberdarensis	IHI B618	MK421517.1	MK421517.1		
Psathyrella aberdarensis	AM1934	follows			
Psathyrella halophila	MICH AH-14321	MG825900.1			
Psathyrella leucotephra	LO138-01	KC992885.1	KC992885.1	KJ664865.1	KJ732775.1
Psathyrella luteopallida	HMJAU 5148	MG734736.1			
Psathyrella luteopallida	Sharp20863	KC992884.1	KC992884.1		
Psathyrella secotioides-type	AH 31746	NR_158908.1	NG_060148.1		
Psathyrella singeri	HMJUA 37867	MG734718.1			
Psathyrella sp.	URM 84467	KC348454.1	KC348448.1		
Psathyrella sp.	PT1	KU847439.1			
Psathyrella sp.	OMV1	KU847442.1			
Psathyrella sp.	EV2A	KU847440.1			
Psathyrella subsingeri-type	HMJAU 37913	NR_160505.1			
Psathyrella sulcatotuberculosa	Battistin and Chiarello 07-10-2013	KJ138423.1			
Psathyrella sulcatotuberculosa	LO55-12	KJ138422.1	KJ138422.1		
Psathyrella sulcatotuberculosa	MCVE29112	MF326002.1			
Psathyrella sulcatotuberculosa	AM1933	follows			
Psathyrella tuberculata	ADK4162	KC992886.1	KC992886.1		
Psathyrella typhae	LO21-04	DQ389721.1	DQ389721.1	KJ664866.1	KJ732776.1
Psathyrella typhae	MJD10-17	JX077004.1			
Psathyrella typhae	TU111512	TU111512			
Uncultured fungus	KU2012-26	AB828223.1			
Uncultured fungus	CMH548	KF800637.1			
Uncultured fungus	FITS URP1 W15	HQ436090.1			
Uncultured fungus	FITS UBP2 BW25	HQ436078.1			
Uncultured Psathyrella	FDBC23	JQ247354.1			
Uncultured Psathyrella	FDBC14	JQ247345.1			
Uncultured root-associated fungus	YL800c18P	FJ362326.1			

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Unite: Kõljalg U, Nilsson RH, Abarenkov K, Tedersoo L, Taylor AFS, Bahram M, Bates ST, Bruns TD, Bengtsson-Palme J, Callaghan TM, Douglas B, Drenkhan T, Eberhardt U, Dueñas M, Grebenc T, Griffith GW, Hartmann M, Kirk PM, Kohout P, Larsson E, Lindahl BD, Lücking R, Martín MP, Matheny PB, Nguyen NH, Niskanen T, Oja J, Peay KG, Peintner U, Peterson M, Põldmaa K, Saag L, Saar I, Schüßler A, Scott JA, Senés C, Smith ME, Suija A, Taylor DL, Telleria MT, Weiß M, Larsson K-H. 2013. Towards a unified paradigm for sequence-based identification of Fungi. Molecular Ecology, DOI: 10.1111/mec.12481
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