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NEW RECORDS FOR THE LIVERWORT AND HORNWORT FLORA OF VIETNAM, 4. – COLLECTIONS OF TRẦN NINH IN TAM ĐẢO MOUNTAINS, VIETNAM

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Abstract: Professor Trần Ninh in 1967 started to deal with the bryoflora of the Tam Đảo Mountains and published two papers in 1980 and 1981 and a checklist of its mosses in 1993. He collected also liverworts. After the long time elapsed I identified these and wish to publish the results. The material consists of hundred specimens, divided among 35 taxa. It contained *Schiffneriolejeunea pulopenangensis* as new to Vietnam and also *Plagiochila cuspidata*, if we consider it as an independent species. Further six became known from Tam Đảo, which were only recently discovered by others in Vietnam (*Chistocaulon dendroides*, *Frullania motoyana*, *Lepidozia fauriana*, *Plagiochila assamica*, *P. javanica*, *Radula retroflexa*).

Keywords: *Bazzania*, endemics, Lejeuneaceae, *Plagiochila*, Southeast-Asia

INTRODUCTION

Tam Đảo Mountains are a 80 km long crystalline range 85 km north of Hanoi, with its slopes covered by wet rainforests. The highest summit has 1592 m elevation. For their rich flora and fauna the area was declared to Conservation Forest in 1977 and was given National Park status in 1996. Its bryoflora since 1967 was studied by Trần Ninh, presently retired professor at the Hanoi University of Science. He published from here many records new to the Vietnam moss flora (Ninh 1980, 1981, 1984), some species even new to science (*Calymperopsis vietnamensis*; *Calypstrochaeta pocsii*, now syn. with *Calypstrochaeta ramosa* subsp. *spinosa* (Nog.) P.J. Lin & B.C. Tan; *Distichophyllum duongii*, now syn. of *Distichophyllum maibarae* Besch.). Finally he published the moss checklist of the area, containing 178 species (Ninh 1993), based on his own collection supplemented by literature data. In this publication he gave a description (with map) of the mountain range and outlined the



history of bryological investigations and the environmental conditions of the area. Between 1967 and 1971 he collected also liverworts and handed me over for identification, which are, after a long elapse, published in the present paper. This liverwort collection contained about hundred specimens, of which 81 were identified (the rest being mostly sterile *Frullania* and *Lejeunea* species). They proved to belong to 35 taxa, of which at least *Schiffneriolejeunea pulopenangensis* (Gottsche) Gradstein is new to the country and other six were only recently published (Shu *et al.* 2017) from other parts of Vietnam. In the November of 1998 we made a joint collecting trip together with professor Trần Ninh in Tam Đảo Mountains, supported by our academies of sciences. We expect to publish its records later.

MATERIAL AND METHODS

The nomenclature follows mostly Söderström *et al.* (2016), except for the species of *Porella*, where I use the names of my revision of Vietnamese species (Pócs 1986). In the enumeration below the same abbreviations mark the different localities, which were used by Trần Ninh (1993) in his moss checklist: **B** – Binh Dân, 950 m; **D** – Dong Dieng, 600 m; **K** – Ký Phú, 950–1420 m; **Q** – Quân Chu, 950 m and **Y** – Yên Mỹ, 1100–1300 m. The collected specimens are deposited in the herbaria of VNU and EGR. After the abbreviation of the localities I give Trần Ninh's original collecting numbers, some annotations and distributional data.

RESULTS

Enumeration of species

Bazzania japonica (Sande Lac.) Lindb. (*Figures 1–2*) **Y**: 68134.

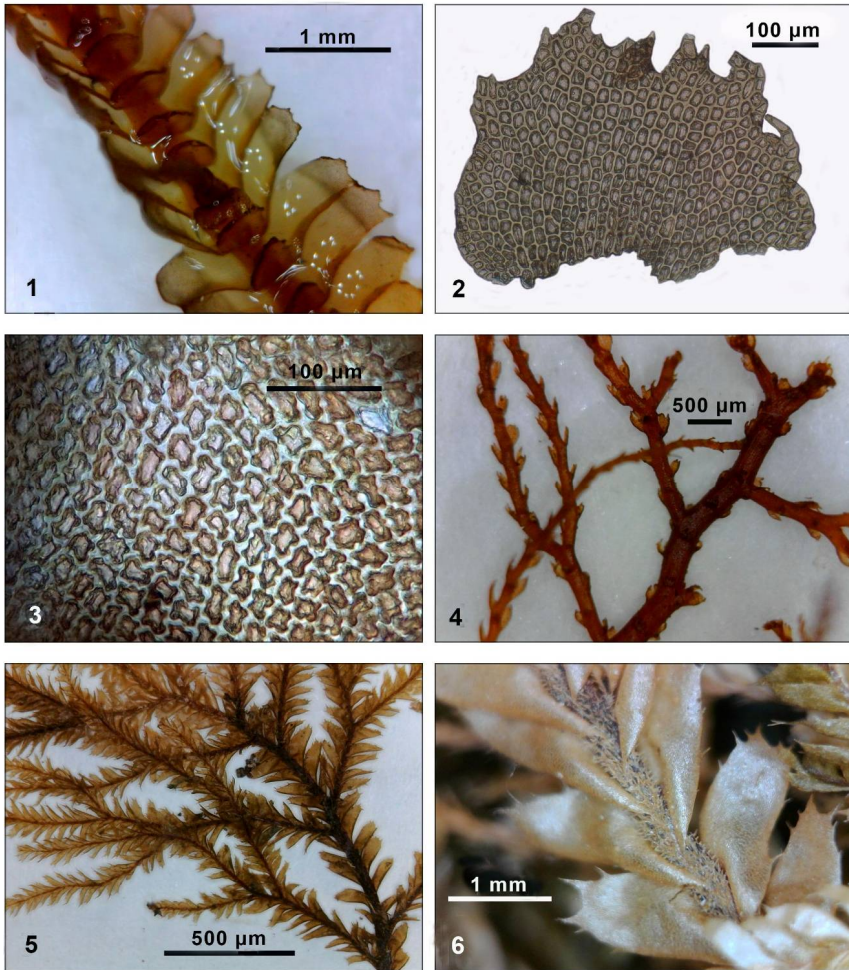
Rare in Vietnam, Southeast Asian species.

Bazzania praerupta (Reinw *et al.*) Trev. **Y**: 68224, 68239, 68241.

Widespread Palaeotropic species.

Bazzania tridens (Reinw. *et al.*) Trev. **Y**: 68183, 68190, 68240C; **B**: 68217, 68236, 68248, 69186; **Q**: 69141; **K**: 69189. The most widespread species in Vietnam and possible also in the whole tropical Asia (Pócs 1969).

Bazzania tridens (Reinw. *et al.*) Trev. var. ***cornistipula*** (Steph.) Pócs **K**: 69102; **B**: 69188. Relatively rare from Japan to Vietnam.

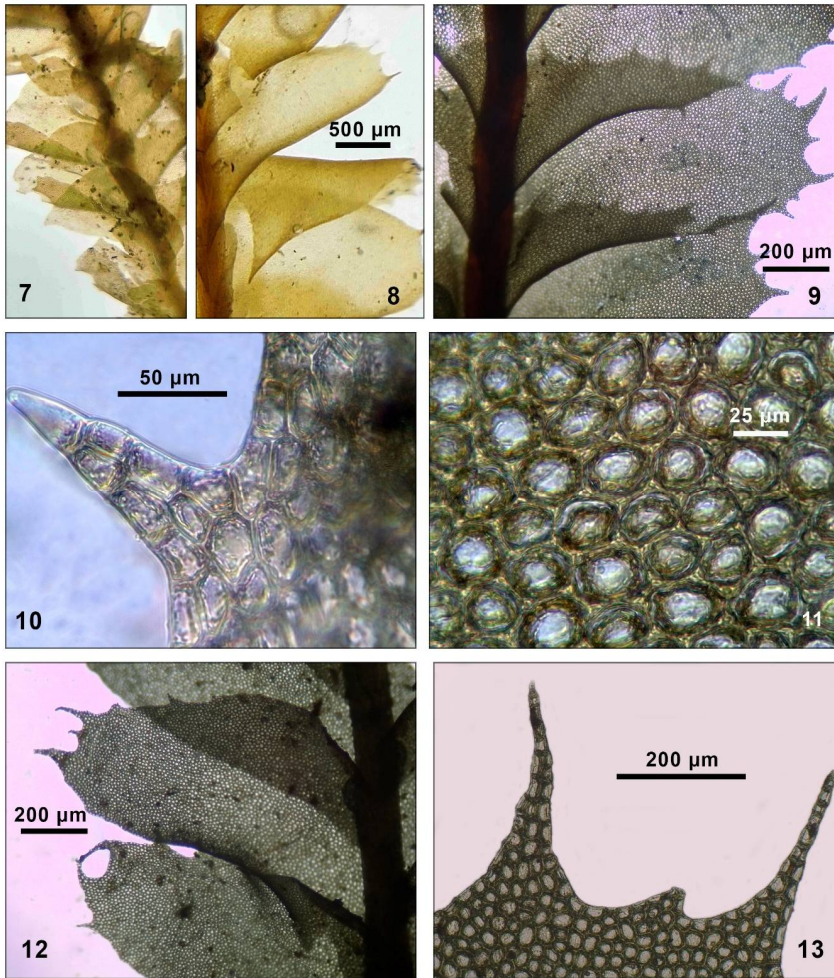


Figures 1–2. *Bazzania japonica* (Sande Lac.) Lindb. Part of shoot, ventral view and underleaf (from 68112). **Figure 3.** *Frullania motoyana* Steph. Lobe cells (from 68192). **Figure 4.** *Lepidozia fauriana* Steph. Habit, ventral view (from 68187). **Figures. 5–6.** *Plagiochila assamica* Steph. Habit, ventral view and part of shoot with paraphyllia, dorsal view (from 67164).

Bazzania vietnamica Pócs Y: 68232. Vietnamese – South Chinese endemic. Closely related to the Neotropical *Bazzania aurescens* Spruce. Pócs (2020) synonymized with it, but probably a different species, being much larger in size, having broader underleaves.

- Cheilolejeunea trapezia*** (Nees) R.M.Schust. & Kachroo **Y:** 68240D. Widespread Palaeotropical species.
- Cheilolejeunea trifaria*** (Reinw. et al.) Mizut. **B:** 69277A. Widespread Pantropical species (Zhu and So 2001).
- Cheilolejeunea ventricosa*** (Schiffn. ex P.Syd.) Xiao L.He **B:** 69170E. Uncommon Malesian species occurring from Mauritius to Australia (Pócs and Streimann 2006).
- Chiastocaulon dendroides*** (Nees) Carl. **Y:** 68211, 68240A. It is known also under the name of *Plagiochila dendroides* (Nees) Lindenb. It was recently found in other parts of Vietnam (Shu *et al.* 2017). Malesian-Pacific species widespread from Japan and Indonesia to Fiji (Inoue 1970).
- Cololejeunea appressa*** (A.Evans) Benedix **K:** 69169C. Widespread Pantropical species.
- Cololejeunea ceatocarpa*** (Aongstr.) Steph. **B:** 69231A. Indomalesian-Pacific species occurring from Réunion to Hawaii (Tixier 1985).
- Cololejeunea sigmoidea*** Ast & Tixier **K:** 69169A Indomalesian species (Zhu and So 2001). Main characteristics are the sigmoid marginal cells gradually passing into normal lobe cells and its strongly reduced lobules, which distinguish it from *Cololejeunea rotundilobula* (P.C.Wu & P.J.Lin) Piippo, with almost always well developed, ovate-orbicular, inflate lobules (Zhu and So 2001).
- Frullania ericoides*** (Nees) Mont. **B:** 69250; **Q:** 69303A, 69312, 69316, 69401A, 69406; **Y:** 69318. Widespread light demanding Pantropical species.
- Frullania hamatiloba*** Steph. **B:** 69278, 69307; **Y:** 68150. Southeast Asian species.
- Frullania motoyana*** Steph. (Figure 3) **Y:** 68192, 68213. Dioicous, with flexuose lobe cell walls and entire perichaetial leaves. Southeast Asian species known from Japan and China. recently found in northern Vietnam (Shu *et al.* 2017).
- Heteroscyphus argutus*** (Reinw. et al.) Schiffn. **Y:** 68204. Widely distributed Indomalesian-Pacific species.
- Jubula hutchinsiae*** (Hook.) Dum. ssp. ***javanica*** (Steph.) Verd. **Y:** 68246A. Indomalesian-Pacific species penetrating in the northern warm temperate zone and occurring also in Madagascar and Transcaucasus (Guerke 1978).
- Lejeunea adpressa*** Nees **Y:** 68122. Widespread Pantropical species (Gradstein 2021).

- Lejeunea parva*** (S.Hatt.) Mizut. **Y:** 68240B. It is known only from Tam Đảo in Vietnam. Southeast Asian species distributed in India and from Japan through southern China, Korea and Thailand to Singapore (Shu *et al.* 2017).
- Lepidozia fauriana*** Steph. **Y:** 68187. It was recently found in other parts of Vietnam. Southeast Asian species (Shu *et al.* 2017).
- Leptolejeunea subacuta*** A. Evans **B:** 69231B, 69274; **K:** 69169D. According to the molecular studies of Shu *et al.* (2021) true *Leptolejeunea elliptica* (Lehm. & Lindenb.) Schiffn. is not Pantropical species but occurs only in the Neotropics. In Tropical Asia the related *Leptolejeunea subacuta* and *L. dapitana* (Spruce) Steph. thrive, and our specimens match the former.
- Lopholejeunea nigricans*** (Lindenb.) Schiffn. **B:** 69277B. Common Pantropical species.
- Mastigolejeunea humilis*** (Gottsche) Schiffn. **Q:** 69401B., According to Sukkharak and Gradstein (2017, as *Thysananthus humilis* (Gottsche) Sukkharak & Gradst., is a species of Palaeotropic distribution).
- Plagiochila assamica*** Steph. (Figures 5–6) **K:** 67164, 67165, 67166, 68385; **Y:** 68305. Characteristics of the species are the abundant setose paraphyllia only on the dorsal side of the stem. It was recently found in other parts of Vietnam too (Shu *et al.* 2017). General distribution: Continental tropical Asia from India to Thailand (So 2001).
- Plagiochila cuspidata*** Steph. (Figures 7–8) **Y:** 68021. According to So (2001) it is an independent species, then is new to Vietnam. According to Rawat and Sriwastava (2007) is a synonym of *Plagiochila parvifolia* Lindenb. As another species is described from the same relationship, as a southern China endemic, *Plagiochila kunmingensis* Piippo (1997), which also differ in size and leaf shape from *P. parvifolia*, I should prefer to keep these species apart, until molecular proofs are available for their synonymy. *Plagiochila cuspidata* is known from India: Darjeeling, Nepal, Bhutan, Myanmar and Thailand (So 2001).



Figures 7–8. *Plagiochila cuspidata* Steph. Shoot, ventral and dorsal views (from 68021). **Figures 9–11.** *Plagiochila javanica* Sande Lac. Part of shoot, dorsal view, apical tooth and lobe cells (from 68288). **Figures 12–13.** *Plagiochila junghuhniana* Sande Lac. Part of shoot, ventral view and apical teeth (from 69051).

***Plagiochila javanica* (Sw.) Nees & Mont. Y: 68315. K: 69147 and Y: 68288** are forms, which were described before as *Plagiochila infirma* Sande Lac., unknown from Vietnam (Figures 9–11). Syn. by So and Grolle (2000). A Malesian-Pacific species (Inoue 1984; Söderström *et al.* 2011).

- Plagiochila junghuhniana*** Sande Lac. (*Figures 12–13*) **K:** 69017, 69051, 69109. A Malesian-Pacific species (So 2001).
- Plicanthus birmensis*** (Steph.) R.M.Schust. **K:** 67133; **Y:** 68225. Widespread Palaeotropic taxon, maybe only a variety of *Plicanthus hirtellus* (F.Weber) R.M.Schust. Sukkharak (2023) discusses their relationship in details.
- Porella acutifolia*** Hampe ex Gottsche & al. (*Figure 14*) **Y:** 68024. Indomalesian species (Pócs 1968a).
- Porella piligera*** (Steph.) Pócs (*Figure 15*). **B:** 69022; **K:** 69010, 69022, 69024, 69157; **Q:** 69347, 69348; **Văn Yên** 1000 m: 74230. Syn.: *Porella caespitans* (Steph.) Hatt. var. *setigera* (Steph.) Hatt. according to Hattori (1978). Vietnam endemic species (Pócs 1968b).
- Porella piligera*** (Steph.) Pócs var. ***grossidentata*** Pócs **B:** 69262; **K:** 67131. Syn.: *Porella caespitans* (Steph.) Hatt. var. *reflexigastria* (Pócs) Hatt. (Hattori 1978). Endemic.
- Ptychanthus striatus*** (Lehm. & Lindenb.) Nees **B:** 69357; **K:** 69036, 69059, 69092; **Y:** 68225A, 68230. Widespread Indomalesian-Pacific species. In Africa replaced by *Ptychanthus africanus* Steph. (Ahonen *et al.* 2005).
- Radula acuminata*** Steph. **K:** 69169B. Widespread Indomalesian-Pacific species. (Yamada 1979).
- Radula retroflexa*** Taylor (*Figures 16–17*) **B:** 69270. Lobules turning away from the stem. Only recently found in Vietnam (Shu *et al.* 2017). Malesian-Pacific species (Yamada 1979).
- Schiffneriolejeunea pulopenangensis*** (Gottsche) Gradstein **Q:** 69303B, 69401C. Characterized by the plane, free lobe margin with 2-4 lobule teeth and by the toothed perichaetial leaves. Widespread Indomalesian species (Wang *et al.* 2016), new to Vietnam.

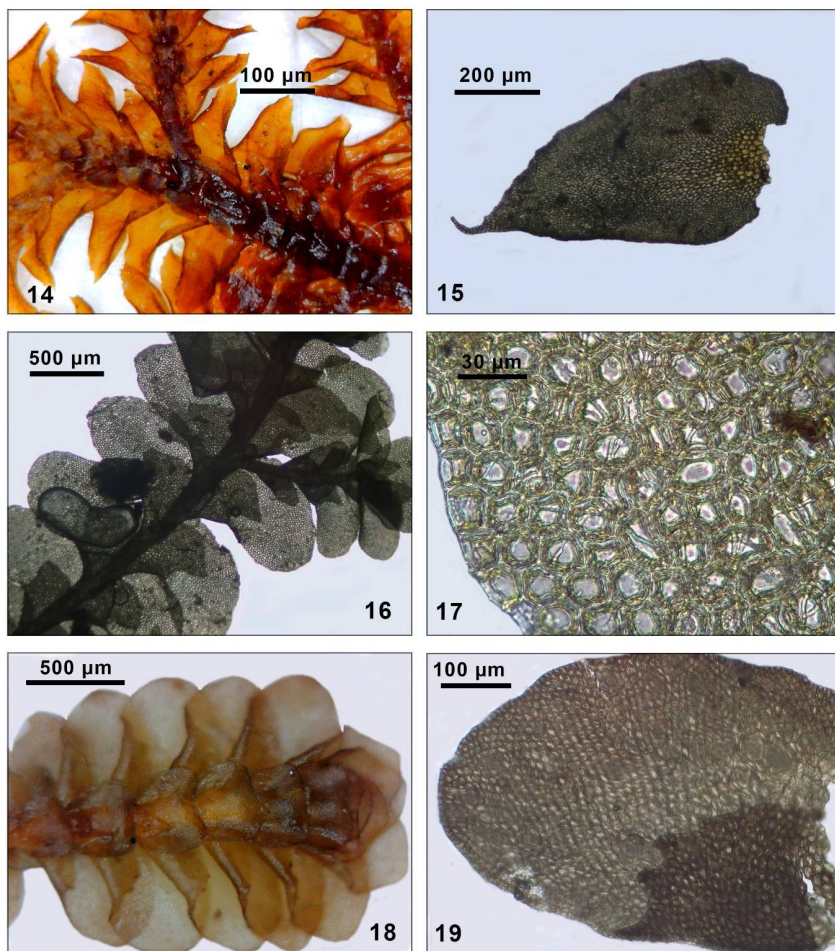


Figure 14. *Porella acutifolia* (Lehm. & Lindenb.) Trev. Part of shoot, ventral view (from 68024). **Figure 15.** *Porella piligera* (Steph.) Pócs. Leaf (from 69022). **Figure 16–17.** *Radula retroflexa* Taylor. Part of shoot, ventral view and lobe cells (from 69270). **Figure 18–19.** *Schiffneriolejeunea pulopenangensis* (Gottsche) Gradstein. Part of shoot, ventral view and leaf, ventral view (from 69303).

DISCUSSION

With these records the known number of liverworts in Tam Đảo National Park, based on Pócs (1969, 2023), Pócs *et al.* (1967), Bakalin and Sinh (2016), Shu *et al.* (2017) and Zhu and Lai (2003) is raised from 56 to 84. As it was already seen (Pócs 1969, 2023), in the northernmost part of Vietnam the number of Indomalaysian

distribution elements is moderate, only about 25% of the bryoflora, along the Sino-Himalayan, Southeast-Asian, Palaeotropic and a few endemic species. Going southwards, the ratio of Indo-Malesian and Malesian-Pacific elements increases and that of the Sino-Himalayan and Southeast Asian decreases (Pócs *et al.* 2019).

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FIRST RECORDS OF *LEPIOTA CYSTOPHORA* (AGARICALES, BASIDIOMYCOTA) IN HUNGARY

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Abstract: *Lepiota cystophora* is reported new to Hungary based on six collections identified by morphological and molecular genetic methods. The species is known from thermophilous oak forests on calcareous soil from the Buda and Vértes Hills. The former generic placement of the species in *Cystolepiota* is not supported by ITS phylogeny.

Keywords: *Cystolepiota*, Central Europe, phylogeny, Agaricaceae

INTRODUCTION

Lepiota (Pers.) Gray is a large genus in the family Agaricaceae with global distribution, characterized by white spore print, a hymenidermal, cutis-like, epithelial or trichodermal pileipellis and with the presence of clamp connections (Candusso and Lanzoni 1990; Vellinga 2003a). The shape of basidiospores and the structure of pileal elements are the most important taxonomic characteristics among *Lepiota* species (Singer 1986; Candusso and Lanzoni 1990; Bon 1993; Vellinga 2003a, b; Qasim *et al.* 2015) and sections were delimited based on these two characters (Singer 1986; Candusso and Lanzoni 1990; Bon 1993). In Hungary, Margit Babos had an outstanding work in the thorough assessing of the lepiotoid genera (Babos 1958, 1961, 1974, 1980).

The genus *Cystolepiota* (Agaricaceae) was described by Rolf Singer based on the type species *Cystolepiota constricta* Singer from Argentina (Singer and Digilio 1952). The genus was originally established for lepiotoid species with neither dextrinoid, nor amyloid basidiospores, and with epithelioid pileipellis. *Cystolepiota*



differs from *Lepiota* in the structure of the universal veil which is made up of loosely arranged globose or inflated elements in the former, while in most *Lepiota* species the veil consists of long, cylindrical or clavate elements (Vellinga 2001a; Noordeloos *et al.* 2001).

Lepiota cystophora Malençon was described from Morocco under *Quercus ilex* (Malençon and Bertault 1970). It is a tiny species with solitary growing basidiomes, appressed, whitish squamules, and pileipellis consisting of large, globose or subglobose cells (Bon 1993; Antonín 1994). Bon (1976) later transferred it to the genus *Cystolepiota*, based on morphological characteristics, such as the spherocysts of the pileipellis. However, recent molecular phylogenetic analyses indicate that the genus *Cystolepiota* as originally circumscribed is polyphyletic, and the taxonomic placement of *C. cystophora* and related species is also unclear (Qu *et al.* 2023; Vellinga 2003b).

The species was originally described from the Mediterranean region, however, later collections confirmed that it occurs in other habitats, too. The first extra-Mediterranean specimens were found in a thermophilous oak forest in the Czech Republic (Antonín 1994). In 2015, 2020 and 2023, fresh basidiomes were discovered in similar habitats in Buda Hills, and in 2022 in Vértes Hills in Hungary. The present work reports the first records of *L. cystophora* from Hungary based on detailed morphological characteristics and phylogenetic analysis.

MATERIALS AND METHODS

Morphology

The dried basidiocarps were deposited in the collection at the Hungarian University of Agriculture and Life Sciences and at the Department of Plant Anatomy of the Eötvös Loránd University. Macromorphological descriptions were based on field notes. Micromorphological data were obtained from dried, we used a Zeiss Axio Imager A2 microscope, and measurements were made with the AxioVision Release 4.8.2 program. Preparations were stained with 1% ammoniacal Congo Red and Melzer's reagents. In describing basidiospores the following abbreviations were used: W = mean spore width, L = mean spore length, Q = variation in the L/W ratios, n = number of spores measured.

Molecular study

We extracted total DNA from dried specimens by using Omega Biotek E.Z.N.A.® SP Plant DNA Kit, according to the manufacturer's instructions. The nrDNA ITS region was amplified by PCR using ITS1/ITS4 primers (White *et al.* 1990; Gardes and Bruns 1993). The successful amplification was confirmed by agarose gel electrophoresis using 1% gel. The amplicons were sequenced at the Biological Research Centre (Szeged, Hungary), with the same primers as used in PCR reactions. The chromatograms were inspected in Chromas 2.6.6 (Technelysium Pty Ltd).

The newly generated sequences were compared to GenBank (<https://www.ncbi.nlm.nih.gov/>). Sequences of *Lepiota cystophora* and closely related taxa based on BLASTn (Altschul *et al.* 1999) as well as additional sequences from various phylogenetic studies in *Lepiota* and *Cystolepiota* (e.g. Qu *et al.* 2023; Vellinga 2003b) were retrieved from GenBank in order to generate a phylogenetic tree. Sequences of *Macrolepiota mastoidea* (Fr.) Singer (AY243604) and *Macrolepiota procera* (Scop.) Singer (HM125514) were chosen as outgroups.

Altogether 37 ITS sequences were included in our phylogenetic analysis (Table 1). The alignment was performed with the online version of MAFFT version 7 (<https://mafft.cbrc.jp/alignment/server/>), and then edited in SeaView version 5.0.5 (Gouy *et al.* 2020). The dataset was subjected to maximum likelihood (ML) analysis in raxmlGUI 2.0 (Edler *et al.* 2021) using 1000 rapid bootstrap searches. The phylogenetic tree was visualised in MEGA 11 (Tamura *et al.* 2021).

RESULTS

Phylogenetic analyses

We generated four new ITS sequences from the Hungarian samples of *Lepiota cystophora* (Table 1). The final alignment consisted of 782 nucleotide positions including gaps. The gaps were treated as missing data. The phylogenetic tree in Figure 1 shows the relationship between the lepiotaceous species included in this study. All sequences of the specimens from Hungary clustered together with a *L. cystophora* sequence (GQ141550) originated from a specimen collected in Italy and published as *Cystolepiota cystophora*

(Vellinga 2010; Osmundson *et al.* 2013) in a strongly supported clade (MLBS = 99%). *Lepiota cystophora* formed a strongly supported sister clade (MLBS = 98%) in our analysis represented by a sequence of *Lepiota cystophoroides* Joss. & Rioussset from France published by Vellinga (2001a). *Lepiota scaberula* Vellinga (2001a) described from the USA, California also belongs to this species complex forming a strongly supported clade (MLBS = 100%) together with *L. cystophora* and *L. cystophoroides*. Based on our ITS phylogeny, *L. cystophora* clearly belongs to the genus *Lepiota*, and shows distant relationship with other *Cystolepiota* sequences (Figure 1).

Table 1. Taxa used in the nrDNA ITS phylogenetic analysis. Species, GenBank accession number, country and specimen voucher are presented, along with references. Sequences produced in this study are indicated in **bold**.

Species	GenBank Accession no.	Locality	Specimen voucher	References
<i>Chamaemyces fracidus</i>	AY176343	Belgium	X-1977	Vellinga (2004)
<i>Cystolepiota pseudofumosifolia</i>	MN810152	China	KUN-HKAS 105918	Hou and Ge (2020)
<i>Cystolepiota pseudofumosifolia</i>	MW447313	Pakistan	FA101	GenBank, unpublished
<i>Cystolepiota pulverulenta</i>	AF391037	Netherlands	E.C. Vellinga 1763 (L)	Vellinga (2001a)
<i>Cystolepiota pulverulenta</i>	AF391036	Great Britain	E.C. Vellinga 1872 (L)	Vellinga (2001a)
<i>Cystolepiota seminuda</i>	AY176350	Netherlands	4-X-1989, H.A. Huijser s.n	Vellinga (2004)
<i>Cystolepiota</i> sp.	U85332	Costa Rica	DUKE-JJ87	Johnson (1999)
<i>Lepiota albogranulosa</i>	LK932285	Pakistan	T 19	Qasim <i>et al.</i> (2015)
<i>Lepiota albogranulosa</i>	LK932284	Pakistan	T 14	Qasim <i>et al.</i> (2015)
<i>Lepiota apatelia</i>	AY176462	Netherlands	26-IX-1990, H.A. Huijser	Vellinga (2003b)
<i>Lepiota castaneidisca</i>	AF391065	USA	E.C. Vellinga 2516 (UC)	Vellinga (2001b)
<i>Lepiota castaneidisca</i>	GQ203818	USA	E.C. Vellinga 3005 (UC)	Vellinga (2010)
<i>Lepiota castaneidisca</i>	AF391055	USA	E.C. Vellinga 2594 (UC)	Vellinga (2001b)
<i>Lepiota clypeolaria</i>	MZ005559	Italy	IZS182	Giusti <i>et al.</i> (2021)
<i>Lepiota clypeolaria</i>	AY176361	Germany	E.C. Vellinga 1683 (L)	Vellinga (2004)
<i>Lepiota coloratipes</i>	KC900376	Spain	SAV F-3212	Vizzini <i>et al.</i> (2014)
<i>Lepiota coloratipes</i>	KC900377	Spain	SAV F-3213	Vizzini <i>et al.</i> (2014)

Species	GenBank Accession no.	Locality	Specimen voucher	References
<i>Lepiota cristata</i>	OL527695	Germany	SeSa55	Sarawi <i>et al.</i> (2022)
<i>Lepiota cystophora</i>	GQ141550	Italy	14107	Osmundson <i>et al.</i> (2013)
<i>Lepiota cystophora</i>	PP776109	Hungary	RA_221006_01	this study
<i>Lepiota cystophora</i>	PP776110	Hungary	DB-2020-11- 12-6	this study
<i>Lepiota cystophora</i>	PP776111	Hungary	AL 20/205	this study
<i>Lepiota cystophora</i>	PP776108	Hungary	DB5896	this study
<i>Lepiota cystophoroides</i>	AF391031	France	E.C. Vellinga 2142 (L)	Vellinga (2001a)
<i>Lepiota erminea</i>	AY176470	Netherlands	E.C. Vellinga 2290 (L)	Vellinga (2003b)
<i>Lepiota lilacea</i>	AY176379	USA	E.C. Vellinga 2451 (UCB)	Vellinga (2004)
<i>Lepiota magnispora</i>	AF391017	USA	AEF1015	Vellinga (2001a)
<i>Lepiota psalion</i>	MG581687	Austria	WU 5152	Vizzini <i>et al.</i> (2019)
<i>Lepiota psalion</i>	MG581688	Italy	CAG P.11_9/7.68a	Vizzini <i>et al.</i> (2019)
<i>Lepiota pseudolilacea</i>	AY176392	Netherlands	E.C. Vellinga 2278 (L)	Vellinga (2004)
<i>Lepiota rufipes</i>	AF391066	Netherlands	9-X-1991, H.A. Huijser	Vellinga (2001b)
<i>Lepiota scaberula</i>	AF391030	USA	E.C. Vellinga 2595 (UC, holotype)	Vellinga (2001a)
<i>Lepiota</i> sp.	OM522666	USA	PUL:00033450	GenBank, unpublished
<i>Lepiota</i> sp.	OM522737	USA	S.D. Russell iNaturalist #91685441	GenBank, unpublished
<i>Lepiota subincarnata</i>	AY176491	Netherlands	E.C. Vellinga 2234 (L) 7-XI-1998, N.J.	Vellinga (2003b)
<i>Macrolepiota mastoidea</i>	AY243604	Netherlands	Dam / E.C. Vellinga 22949 (L)	Vellinga (2004)
<i>Macrolepiota procera</i>	HM125514	China	HKAS8108	Ge <i>et al.</i> (2010)

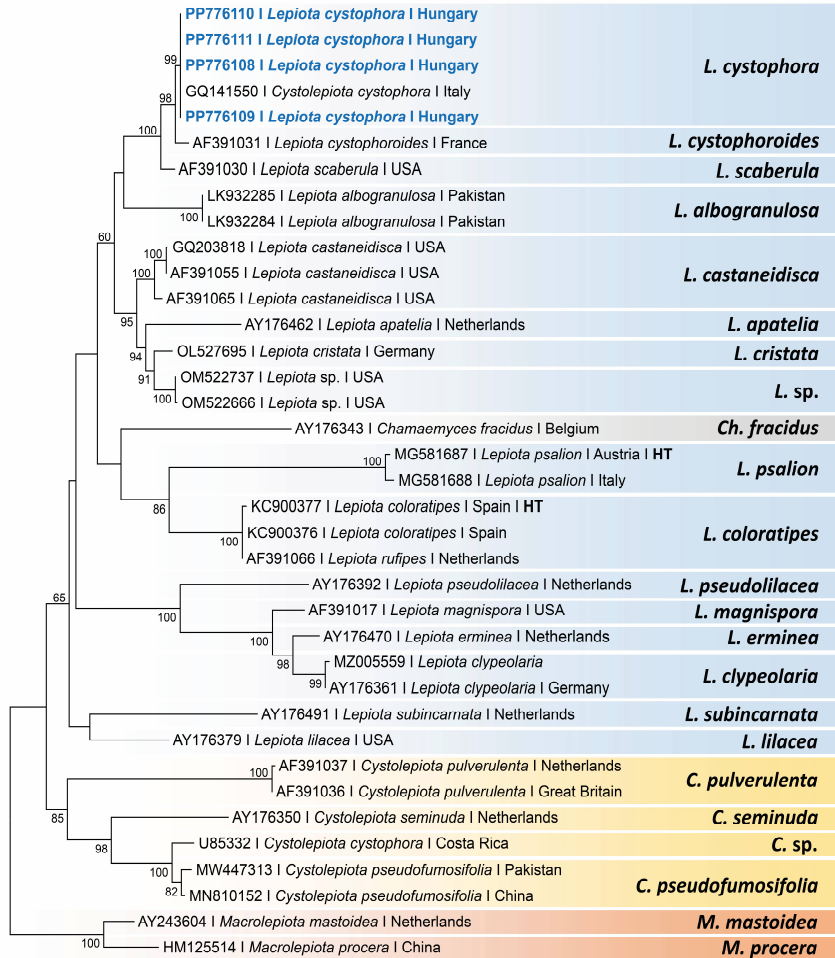


Figure 1. Phylogeny of *Lepiota* with emphasis on *L. cystophora* inferred from Maximum Likelihood and RAXML analyses of the nrDNA ITS sequences. Topology is from the best scoring Maximum Likelihood (ML) tree. *Macrolepiota mastoidea* and *M. procera* served as outgroup. ML bootstrap values >50% are shown above or below the branches. The bar indicates 0.05 expected change per site per branch.

Taxonomy

Lepiota cystophora Malençon, Flore des champignons supérieurs du Maroc 1: 124 (1970)

≡ *Cystolepiota cystophora* (Malençon) Bon, Documents Mycologiques 6 (24): 43 (1976) (Figure 2)



Figure 2. Macromorphology of *Lepiota cystophora*. A–D. Basidiomes A. DB-2020-11-12-6 B. AL 23/333 C. RA_221006_01 D. AL 20/205 (Photos: A: B. Dima, B, D: L. Albert, C: Á. Radnóti).

Pileus 2–4 cm, conical-hemispherical, then convex; squamulose, whitish when young, later ochre, yellow ochre, sometimes with rose tinges with white, floccose veil remnants at the margin. **Lamellae** crowded, free, whitish, cream-coloured when mature, slightly brownish when dry. **Stipe** 2–5 cm long, 0.2–0.4 cm in diameter, whitish with ochre or rosaceous colour towards the base. Ring zone floccose, woolly, fugacious. **Context** thin, whitish, with pinkish tinges (Figure 2). **Basidiospores** (6.2–)7.07–7.72(–8.5) × (3.5–)4.12–4.73(–5.7) μm, average: 7.4 × 4.5 μm, Q = (1.3–)1.52–1.79(–2.1), Qav = 1.7 μm, ellipsoid, thick-walled, hyalin, non-dextrinoid, not metachromatic in Cresyl Blue (Figure 3E). **Cheilocystidia**: 30–50 × 6–10 μm, clavate (Figure 3B–C). **Pileipellis**: composed of large, globose sphaerocysts, 20–30 μm in diameter (Figure 3A).

Habitat: Thermophilous oak forests on calcareous soil. The pH is presumed to be alkaline as a result of the carbonate bedrock. At the Széchenyi-hegy and Csillebérc localities, the vegetation is dominated by *Quercus pubescens* and *Fraxinus ornus* with scattered *Q. cerris* trees, at Szárliget *Q. cerris*, *Q. pubescens* and *Carpinus betulus*, at János-hegy *Q. cerris* with some *C. betulus*, while at Normafa the main species were *Fagus sylvatica*, *Quercus* sp. and *Tilia cordata*.

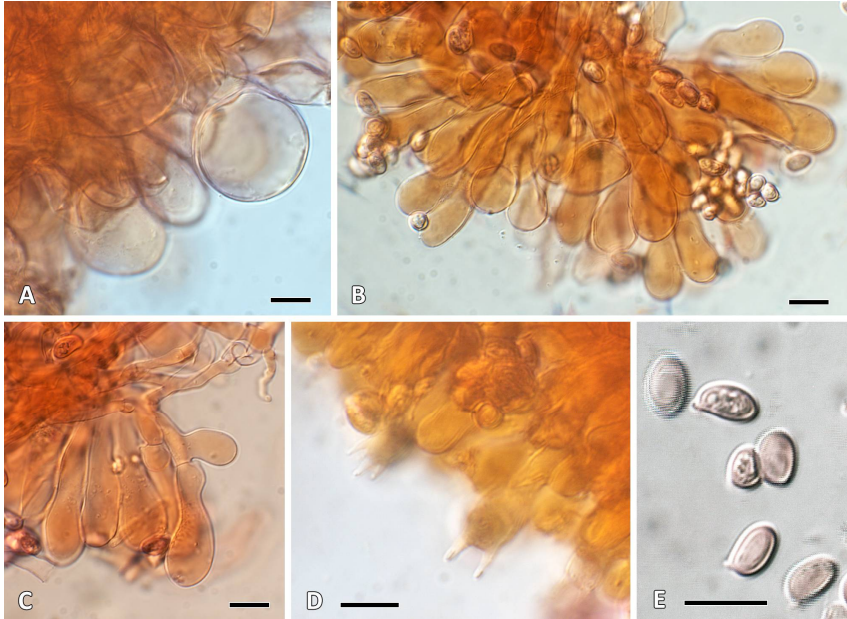


Figure 3. Micromorphology of *Lepiota cystophora*. **A.** Pileipellis. **B–C.** Cheilocystidia. **D.** Basidia. **E.** Basidiospores. Scale bars: A–D = 10 μ m (Photos: Á. Radnóti).

Specimens examined: Hungary, Buda Hills, Budapest, Csillebérc, alt. 450 m, 18 Oct 2015, L. Albert, B. Dima (DB5896); Buda Hills, Budapest, Széchenyi-hegy, alt. 460 m, 15 Oct 2020, L. Albert (AL 20/205); Buda Hills, Budapest, Széchenyi-hegy, alt. 460 m, 12 Nov 2020, L. Albert, B. Dima (DB-2020-11-12-6); Buda Hills, Budapest, János-hegy, alt. 300 m, 9 Nov 2023, B. Dima (RA_231109_09); Buda Hills, Budapest, Normafa, alt. 477 m, 19 Nov 2023, L. Albert (AL 23/333); Vértes Hills, Szárliget, alt. 300 m, 6 Oct 2022, Á. Radnóti (RA_221006_01).

DISCUSSION

Six collection data from five new localities are reported, four from Buda Hills and one from Vértes Hills, representing the first records of *Lepiota cystophora* in Hungary, confirmed by morphological and molecular studies. All samples were collected in thermophilous oak forests on calcareous soil. The species might be overlooked in thermophilous deciduous habitats in Central as well as Western and South Europe.

Our analysis included a sequence from Costa Rica labelled incorrectly as *Cystolepiota cystophora* (U85332), which clustered distantly from the European *L. cystophora* sequences in the phylogenetic tree (Figure 1), representing another species. Qasim *et al.* (2015) suggested that *C. cystophora* might be identical to *L. cystophoroides*, however, the currently available sequence data suggest that these two are sister species (Figure 1). There are slight morphological differences between the two species: *L. cystophoroides* has no purplish or rosaceous tinges in the stipe base and has a rather hymeniform pileipellis with few sphaerocysts, while *L. cystophora* has more sphaerocysts and the stipe base possesses a purplish, rosaceous colour (Josserand and Rioussset 1972). The known sequences of the morphologically identified *Cystolepiota* species did not form a monophyletic group across any phylogeny published to date (e.g. Vellinga 2003b, 2010; Qu *et al.* 2023). One of the key problems to understand the delimitation of *Cystolepiota*, is the lack of any sequence data of *C. constricta*, the type species of the genus. These facts indicate that a global phylogenetic and taxonomic revision is urgently needed to reveal the generic limits of lepiotaceous fungi.

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FRULLANIA RADDI (MARCHANTIOPHYTA) SPECIES NEW TO AUSTRALIA AND TO SCIENCE I.

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Abstract: During the revision of the liverwort genus *Frullania* for Australia, one new species to science is here described, as *Frullania saroltae* sp. nov. (subg. *Frullania* sect. *Microphyllae*). Two new records for the flora are also reported, *Frullania eymae* and *Frullania ornithocephala* var. *intermedia*, (both subg. *Trachycolea*) and previously known only from New Guinea. Descriptions and illustrations of the taxa are provided.

Keywords: Australia, Australasia, endemism, New Guinea, new species

INTRODUCTION

The work on the revision of *Frullania* species for the Flora of Australia started in 1999, encouraged by the late Heinar Streimann. Field collection trips were organised in 1999, 2000 and 2001 financially supported by the Australian Biological Resources Study Participatory Program and in 2004 on the author's own. During these trips thousands of *Frullania* specimens were collected and also borrowed for revision from the Australian National Herbarium in Canberra (CANB). Research has continued sporadically over the past two decades with ongoing progress and publications. Recently the author is endeavouring to complete this project in collaboration with Matt von Konrat (Field Museum) with whom have divided the revision into taxonomic groups, including his own responsibility of revising the subgenera *Fusiorelliogeræ*, *Homotropantha* and *Trachycolea*, of which only the last has numerous (about 36) species.

It is here reported one new species to science as well as two new records for the Australian *Frullania* flora representing ongoing research of this project.



DESCRIPTION OF THE NEW SPECIES

Taxonomic treatment

Frullania saroltae Pócs, sp. nov. (Figures 1–12)

Subgenus *Frullania* sect. *Microphyllae* (R.M.Schust.) Gradst., Ilk.Borg., & E.Lima 2020, Phytotaxa 456:122.

Diagnosis: *Frullania saroltae* is similar in several aspects to other members of sect. *Microphyllae* by its small size, usually evenly incrassate, thick cell walls and the scattered red ocelli throughout the leaf-lobe, subcylindric, ampullaceous lobule on a short stalk standing parallel to the stem or incumbent on branches, with narrow underleaves, only 1.5–2 × of the stem width, with parallel segments. It is different from the morphologically allied widespread Asian *Frullania alstonii* Verd. (Verdoorn 1930) by the lack of moniliate ocelli in most of the leaves and by its relatively larger lobules; from Chinese *Frullania punctata* Reim. (Reimers 1931) by its wide, not constructed lobule, from *Frullania perocellata* Onraedt (Onraedt 1978) from Sri Lanka and from the Japanese *Frullania pseudoalstonii* Tsudo and J.Haseg. (Tsudo and Hasegawa 2006) by its entire female perichaetial leaves, without teeth.

Type: AUSTRALIA, NEW SOUTH WALES: Dorrigo National Park E of Dorrigo town. Along Rosewood Creek track, on bark of trees in subtropical notophyll vine rainforest with many epiphytes, 30°22'S, 152°47.8'E, at 600–730 m elevation. 15 Febr. 2000, S. & T. Pócs and E.A. Brown 009/AC (holotype EGR, isotype: NSW).

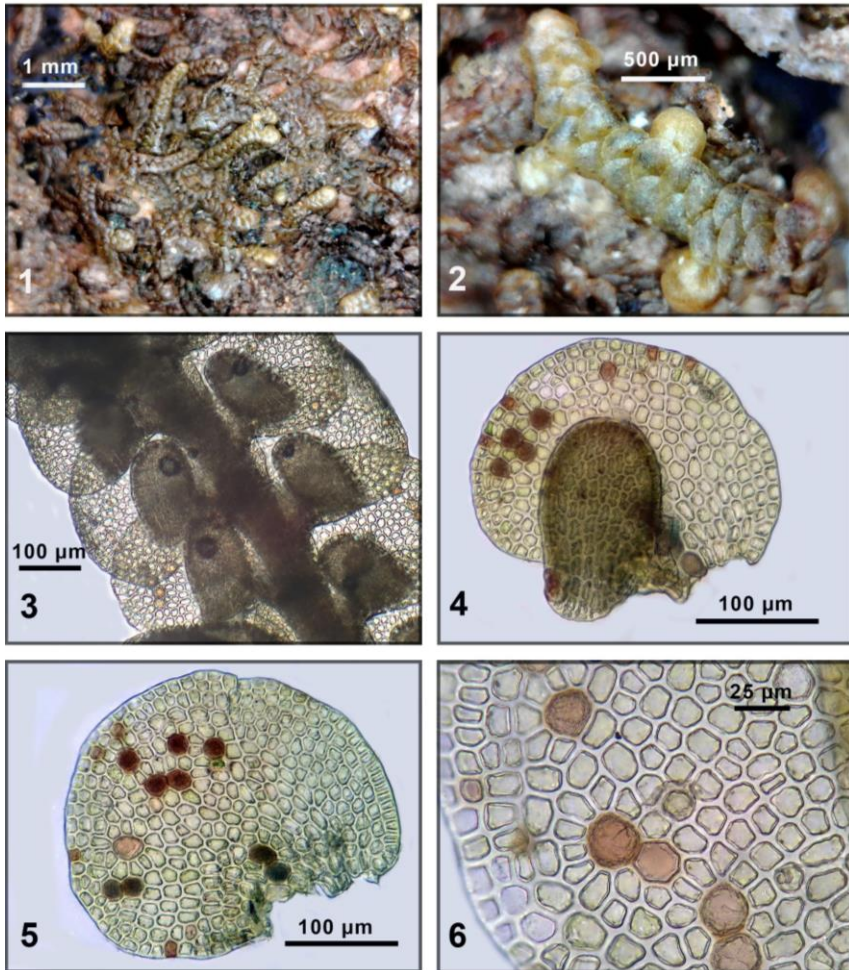
Description: Small, green when alive, in herbarium olive-green plants with a silky shine. Even by a handlens glistening ocelli are visible scattered in the lobe. It forms wefts of 5–8 cm diameter on bark. Dioicous, specimens of both sexes quite fertile. **Shoots** 300–450 µm wide, up to 12 mm long, irregular pinnately branching. Stem 40–80 µm thick, surrounded by 12–18 rows of thick walled, brownish-yellow pigmented, 10–20 × 4.5–6 µm cortical cells. First branch underleaves consist of one unlobed ventral and one unlobed dorsal segment of more or less equal size. Rhizoids short, reddish brown, not commonly develop at the base of underleaves. **Leaf lobes** slightly convex, imbricate, sometimes caducous, asymmetric ovate-reniform, 250–400 × 200–320 µm (20–35 cells long and 16–28 cells wide), with 4–16 scattered, brownish- or pale red ocelli. Moniliate ocelli, composed of 2–4 cells (occur only in very few, in the 1–2 % of the leaves). The dorsal lobe base extending a little beyond the

opposite side of the stem. Cell walls straight, equally (2–3 μm) thickened, at the border square, with a lumen of 6–8 μm , median cells polygonal, 8–12 μm . Basal cells similar. Cuticle smooth, translucent. The ocelli are larger, of 12–20 μm diameter and a little protruding from the leaf surface. **Lobules** subcontiguous, on side branches imbricate, campanulate with slightly widening, open mouth, decorated by 1–4 ocelli in its distal margin, other cells have flexuose walls, 10–15 \times 4–8 μm . Lobule size 150–176 \times 74–100 μm , their length is about 2/3 of the lobe width. The surface of their ventral projection, compared to that of the whole lobe, much larger than in most species of Section *Microphyllae*, covering in average 26,68%. (For example, lobule length in *F. alstonii* is only about 2/5 of lobe width and about 8,5 % of the whole lobe surface). **Stylus** formed by 3–5 uniseriate cells tipped by a hyaline papilla. **Underleaves** narrow, oblong-linear with cuneate base and almost straight incision line, 1,5–2,2 stem width, 350–400 \times 100–115 μm , without ocelli, with narrow, V shaped incision of its 1/5 length. Segments parallel, 4–5 cells wide at base, 6–10 cells long, entire, with obtuse apex. The **androecium** is on short side branches, globular, of 350–400 μm diameter, composed of 4–5 pairs of male bracts. **Gynoeceum** at the end of longer side branches, with or without innovations, with only one or seldom two sets of perichaetial leaves. The female bracts have rounded apex and almost equal sized, but acute lobules, fused by their half length to the lobe. These and the bracteole have entire margins without any dentition and with many scattered ocelli. The perianth is elongated heart shaped, trialate with two sharper side keels and a third blunt ventral one. In the perianth wall there are much more than hundred scattered ocelli. The beak is 90–110 μm long and 50–80 μm wide with truncate and papillose apex. The type specimen had mature **sporophytes** with already open capsules. Seta about 700 μm long. The capsule segments are 800–840 μm long and 240–250 μm wide, most with 8 unispiral elaters, 800–840 μm long and 14–18 μm thick. Spiral thickenings are rust coloured. Spori olive brown, roughly globose of 36–41 μm diameter, each with 28–30 rosettes.

Etymology: The new species is dedicated to my late wife Sarolta, its co-collector, who helped me so much during the Australian trips.

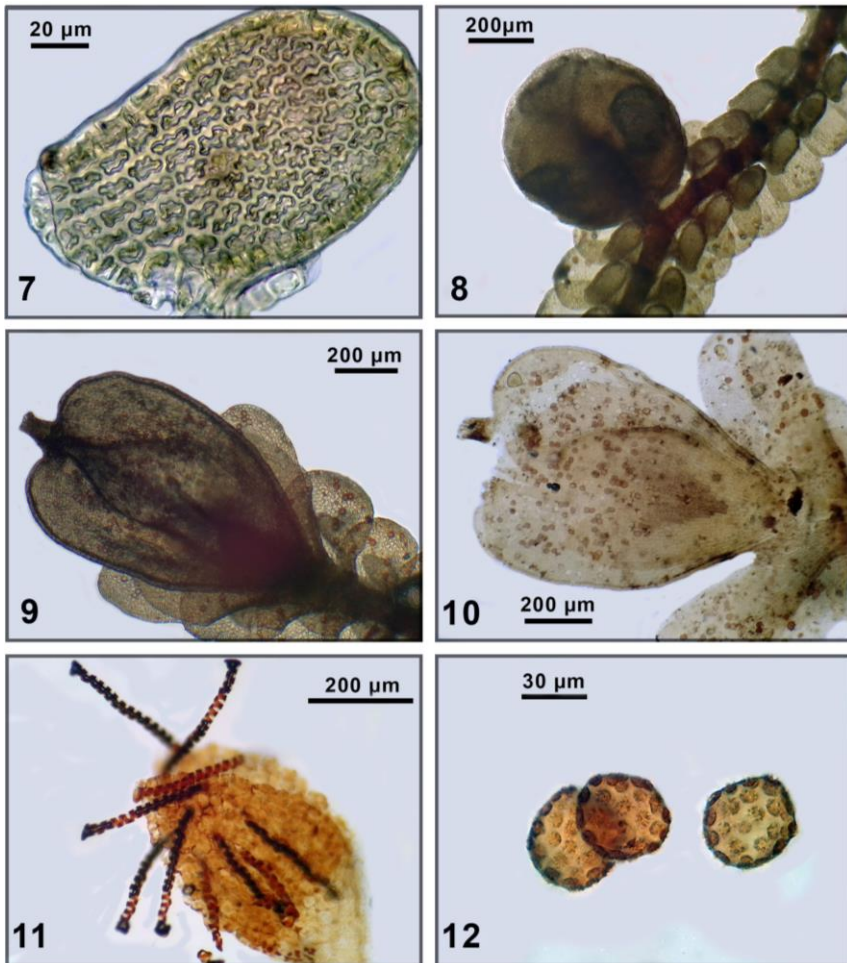
Distribution: The new species is possibly endemic to New South Wales. We came across only one more specimen from the same locality, collected at the same day on a fallen canopy twig: S. & T. Pócs

and *E.A. Brown 009/DH* (CANB paratype, EGR isoparatype on microslide).



Figures 1–6. *Frullania saroltae* Pócs, sp.nov. 1–2: Habit on substrate. Dorsal view. 3: shoot, ventral view. 4: Leaf, ventral view. 5: Lobe after removal of lobule. 6: Marginal and median lobe cells (All photographed from the type).

Taxonomic notes: The *Frullania fragilifolia* complex characterized by their small size, narrow underleaves and many times with caduceus lobes within *Frullania* sect. *Frullania* (“*F. tamarisci* complex” with ocellate species) was distinguished as subsect. *Microphyllae* R.M.Schust, Phytologia 57(5): 370, 1985.



Figures 7–12. *Frullania saroltae* Pócs, sp.nov. **7:** Lobule with style, ventral view. **8:** Male branch, ventral view. **9:** Perianth, ventral view. **10:** Perianth, ventral view, cleared by Hoyer solution to show the numerous ocelli. **11:** two capsule segments covering each other. The front one bears 8 elaters. **12:** spori (All photographed from the type).

Gradstein, Ilkiu-Borges and Lima in *Phytotaxa* 456 (1): 122, 2020, elevated this group into section rank, counting a number of species in Asia and very few in Europe and in the Americas. These species all differ from each other by minor morphological characters. Heinrichs *et al.* (2010) performed a molecular investigation into the *Frullania tamarisci* complex that revealed even smaller morphological

differences reinforced the species rank of these partly sympatric species. This study provides strong support that a similar situation exists for members of section *Microphyllae*, where the above new species belongs. Although some members of this section, like *Frullania alstonii* or *F. microphylla*, are more widespread, the majority of its species are endemic to a smaller area.

TAXA NEW TO THE AUSTRALIAN FLORA

Frullania eymae S.Hatt. J. Hatt. Bot. Lab. 39: 284, 1975.

(*Figures 13–16*).

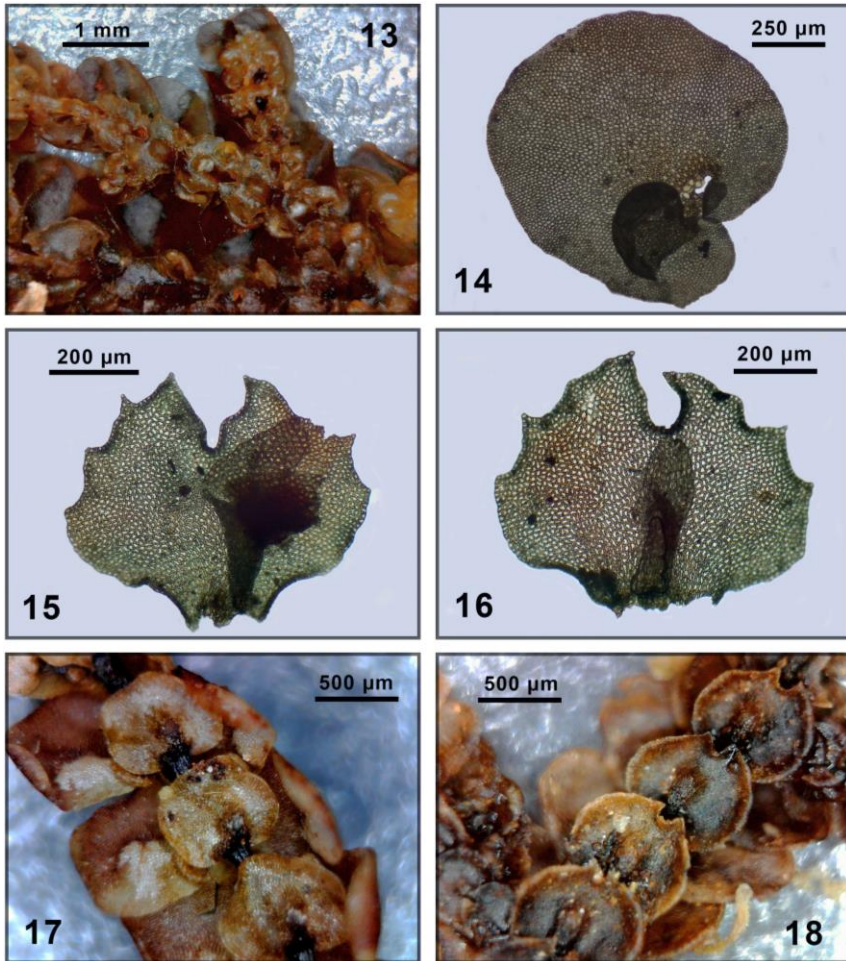
(Subgenus *Trachycolea* Spruce 1884, Hep. Amaz. Et And. 31, 1884)

Frullania eymae var. *dentistipula* S.Hatt. J. Hatt. Bot. Lab. 44: 527, 1978. Synonymized by Hattori (1982: 250).

Frullania dentella S.Hatt. *sp. nov. nom. nud. in sched.*, 1984. (CANB 8302469). Illustrations: Hattori (1975: 285, fig. 142; 1978: 528, fig. 202; Hattori and Streimann 1985: 104, fig. 2)

AUSTRALIA, QUEENSLAND, Atherton Tableland. Yungaburra Road, 2 km SE of Atherton, 17°16'S, 145°29'E, 850 m elevation, Exotic fruit orchard. On *Casuarina* stem. *Streimann 16836* (CANB). Paluma range, Crystal Creek National Park, McClelland's Lookout at the edge of the plateau, at the E side of Paluma village. 19°00'S, 146°12.8'E, at 900 m elev. On bark in montane rainforest opening. *S & T. Pócs, 01109/J*, accompanied by *A. Cairns, E.A. Brown* and *C. Cargill* (CNS, EGR).

The main characters of this medium size plant are the densely imbricate, elliptic lobes with undulate margin and strongly appendiculate dorsal base, helmet shaped lobuli almost covered by the large, imbricate, non appendiculate, reniform to widely cordate underleaves with wavy, toothed margin. Sinus 3/10 deep, V or U shaped, underleaf lobes triangular with acuminate- acute apex. Hitherto it was known only from the mountainous areas of New Guinea where it is widespread in 600–2000 m elevation (Hattori 1975, 1978, 1982; Hattori and Piippo 1986). New to Australia.



Figures 13–18. *Frullania eymae* S.Hatt. **13:** Habit, ventral view. **14:** Leaf, ventral view. **15–16:** Underleaves, ventral view (from Streimann 16836). **17:** *Frullania ornithocephala* Steph. var. *intermedia* S.Hatt. Habit, ventral view (from S. & T. Pócs 01079). **18:** *Frullania ornithocephala* Steph var. *ornithocephala* (Steph.) Hatt. Papua-New Guinea, Chimbu prov., Kegslugi (foot of Mt. Wilhelm), at 2600 m, epiphyte in degraded montane forest. De Sloover 42.722 (sub *Frullania pauciramea* Steph. EGR ex NAM).

***Frullania ornithocephala* Steph. var. *intermedia* S.Hatt.** J. Hattori Bot. Lab. 65: 439, 1988. (Figures 17–18).

(Subgenus *Trachycolea* Spruce 1884, Hep. Amaz. Et And. 31, 1884)

Frullania nobilis Steph. ssp. *nobilis* var. *intermedia* S.Hatt. J. Hattori Bot. Lab. 37: 116, 1973. Synonymized by Hattori (1988: 439).

Illustration: Hattori (1973: 116, fig. 64).

AUSTRALIA: QUEENSLAND, Track to Mt Finigan Range, Cedar Bay National Park, 39 km S of Cooktown. 15°49'S, 145°16'E, at 880 m. Rainforest on steep slope with large rock outcrops. On shaded treelet. *Streimann 57115A, 57120* (CANB); Lamb Range, near Mt. Haig, 20 km SE of Mareeba, 17°05'S, 145°35'E, at 1100 m elevation. Tropical forest on moderate slope. On shaded treelet stem. *Streimann 57656A* (CANB), Atherton Tableland, Hugh Nelson Range. Longlands Gap State Forest. Summit around the telecommunication tower, 22 km S of Atherton. Somewhat disturbed montane rainforest dominated by *Acacia* and *Aphitonia*. On bark and hanging from tree branches. 17°27'S, 145°29'E, at 1235–1240 m elevation. *Pócs & Streimann 99109/AD* (CANB, EGR), *S. & T. Pócs 01079/B*, accompanied by *A. Cairns, E.A. Brown* and *C. Cargill* (BRI, EGR).

In tropical and subtropical Asia and Australasia there are about ten species with entire, unlobed underleaves. But from Australia this is the only one. *Frullania ornithocephala* var. *intermedia* has large (equal to leaf size), reniform, imbricate, nearly flat underleaves, with round or widely retuse apex. *Frullania ornithocephala* is a very variable plant (Hattori 1982; Hattori and Piippo 1986; Hattori 1988). From Queensland there is one record of *Frullania pauciramea* Steph. (Bolin and Henderson 2002: 223, not yet mentioned in Windorf 1987) which finally became the synonym of *F. ornithocephala*. It is a question, whether this record is really that var. *intermedia* or belongs to var. *ornithocephala*.

DISCUSSION

Dr. Sinske Hattori, former leading Japanese hepaticologist decades ago investigated and described a good number of Australian *Frullania* specimens in the possession of Australian National Herbarium (CANB) of the Centre for Australian National Biodiversity Research (Hattori 1979a, 1979b, 1982, 1983, 1984, 1987a, 1987b, 1988a, 1988b; Hattori and Piippo 1986). He pencil marked provisionally a few specimens, what he suspected to be new to science. These still need further investigation. One of those was the case of *Frullania eymae*. Also further fieldwork and the currently unidentified specimens promise novelties, so that the known number of 69 Australian taxa (McCarthy 2003; von Konrat and Braggins 2003 and the present paper) probably will increase, although several taxa could fall in synonymy.

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KEY TO GENERA AND GENERIC GROUPS OF LICHEN-FORMING FUNGI IN HUNGARY

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Abstract: A key to treat 269 genera of lichen-forming fungi including 940 species from Hungary is compiled. The key sorts lichens according to traditionally distinguished morphological groups based on vegetative and reproductive structures, fruticose, foliose and crustose thalli with apothecia and perithecia. Elongated and stalked reproductive structures are also distinguished, as well as the type of photobiont (cyanobacterium, green alga) is considered. Due to recent phylogenetical-taxonomic studies several taxa with recent nomenclature grouped under larger, morphologically related genera following the practice of published national identification keys from Germany and Great Britain.

Keywords: genera, identification, lichens, morphology, taxonomy

INTRODUCTION

The last identification key to lichen-forming fungi in Hungary was published by Klára Verseggy (1994) 30 years ago. Nomenclatural and taxonomic changes took place over the decades in the majority of taxa. An online checklist was compiled in 2009 (Lókös and Farkas 2009) and updated from time to time, resulting the most recent version (Farkas *et al.* 2023). According to our recent knowledge the number of Hungarian lichen-forming fungi reached 940 belonging to 269 genera.

A key to treat these 269 genera of lichen-forming fungi is compiled here to support the identification of recent collections and the revision of herbarium material. The key sorts lichens according to traditionally distinguished morphological groups based on vegetative and reproductive structures, fruticose, foliose and crustose thalli with apothecia and perithecia (*Figure 1–2*). Elongated and stalked reproductive structures are also distinguished, as well as the type of photobiont (cyanobacterium,



green alga) is considered. In several cases, the key is leading to species because of their specific morphological characters or for presenting their relation to similar species. Due to recent phylogenetical-taxonomic studies (e.g., Crespo *et al.* 2010; Divakar *et al.* 2017; Kistenich *et al.* 2018; Zhao *et al.* 2016) several taxa with recent nomenclature are grouped under larger, morphologically related genera following the practice of published national identification keys from Germany (Wirth *et al.* 2013) and Great Britain (Smith *et al.* 2009). In a key to species (currently under preparation) starting from the known genera or generic groups, the identification will result in species named according to the recent nomenclature. For certain species molecular genetic sequencing might be necessary, otherwise the result may lead to the identification of species groups only.

For nomenclatural issues indexfungorum.org and mycobank.org and further recent literature sources were consulted. Stereo and research microscopes are necessary for studying microscopic characters. Spot tests (C, K, Pd) and chromatographic analysis of lichen secondary metabolites must be carried out where it is necessary following the usual methods (Arup *et al.* 1993; Orange *et al.* 2010).

GLOSSARY

acicular – needle-shaped

amyloid – stained blue by Lugol's iodine solution

apothecium (plural: *apothecia*) – a round, nearly flat or often cup-like ascus producing fruiting body, where the → *hymenium* is exposed at maturity (*Figure 1g, i, 2e*)

ascoma (plural: *ascomata*) – ascus producing fruiting body

areolate – consists of islands (areoles) of thallus developing on the hypothallus, resulting a cracked appearance of the thallus

ascospore – reproductive propagule, the product of meiosis, most often 8, formed and arranged in a line within an → *ascus*

ascus (plural: *asci*) – a sac-like fungal cell containing the ascospores (produced by meiotic cell division)

basidioma (plural: *basidiomata*) – basidium producing fruiting body

- basidiospore** – reproductive propagule, the product of meiosis, formed within a basidium (→ *basidioma*), most often 4 are released independently at the apex of a basidium
- biatorine** (of apothecia) – lacking a true exciple when mature, pale or coloured (not black), soft in consistency, often becoming convex
- cephalodium** (plural: *cephalodia*) – delimited region of a lichen thallus containing a photobiont (usually a cyanobacterium) different from that characteristic of the rest of the thallus (mostly a green alga); inside the thallus or a warty, squamulose, or shrubby structure on the surface
- cilia** (singular: *cilium*) – hair-like out-growths of the thallus usually at or near the margin
- clavate** – club-like
- corticolous** – on bark of a tree
- crustose** (of lichen thalli) – crust-like with modified layered or more simple structure (*Figure 1f-i, 2a-d*)
- epiphytic** – on plant / tree bark
- exciple** – tissue forming the margins or walls of an ascoma
- farinose** – flour-like, powdery
- filamentose** (*filamentous*) – thread-like
- foliose** (of lichen thalli) – leaf-like; with layered structure of an upper cortex, photosynthetic layer, medulla and lower cortex separable from the substratum (*Figure 1d, e*)
- fruticose** (of lichen thalli) – shrub-, beard- or worm-like appearance with modified layered structure (*Figure 1b, c*)
- furcate** – forked
- fusiform** – spindle-like; narrowing at both ends
- gyrose** – brain-like, circularly folded
- habitat** – living place
- halo** – → *perispore*
- hymenium** (of ascomata and basidiomata) – the spore-bearing layer of a fruit-body
- hypha** (plural: *hyphae*) – a fungal filament
- hypothecium** – the tissue below the hymenium and generative layer
- isidium** (plural: *isidia*) – a photobiont-containing outgrowth of the cortex (*Figure 2g*)

- lecanorine** (of apothecia) – with a thalline exciple (containing both photobiont cell and fungal hyphae) (*Figure 1f, g*)
- lecideine** (of apothecia) – without a thalline exciple (margin consists of fungal hyphae only → *true exciple*) (*Figure 1h, i*)
- leprose** – thallus surface a granular mass of algal and hyphal cells without any cortex (*Figure 2a, b*)
- lirella** (plural: *lirellae*; adjective: *lirellate*) – long, narrow apothecium (*Figure 2c*)
- mazaedium** – a dry powdery mass of free ascospores and occasionally sterile elements lying on the surface of the ascoma (*Figure 2d*)
- medulla** – the loose layer of hyphae below the cortex and photobiont layer
- muriform** (of spores) – divided by transverse and vertical or oblique cross walls
- mycobiont** – fungal symbiont partner (→ *thallus*) consisting of → *hyphae*
- paraphysis** (plural: *paraphyses*) – a slender, branched or unbranched hypha growing upward and basally in a fruitbody (→ *ascoma*) (*cf.* → *pseudoparaphysis*)
- paraphysoid** – interascal or pre-ascal tissue stretching and coming to resemble → *periphysoid*
- paraplectenchyma** – tissue consisting of cell-like hyphae with ± isodiametric lumina
- periphysoid** – short, interascal filaments growing down from the top of a perithecium, perithecium-like apothecium or pycnidium (e.g. in *Cryptothele*)
- perispore** – a colourless, often gelatinous layer (halo) enveloping a spore outside the main spore wall (epispore).
- perithecium** (plural: *perithecia*) – a subglobose or flask-like ascoma in which the hymenium is not exposed
- photobiont** – a photosynthetic symbiont which may be either a green alga or a cyanobacterium (→ *thallus*)
- podetium** (plural: *podetia*) – the lichenized stem-like part of an apothecium; typically in *Cladonia*, apothecial discs may be missing (→ *secondary thallus*) (*Figure 1a*)
- polarilocular** (of ascospores) – bicellular with the two cells separated by an often thick, centrally perforated septum (e.g., at *Caloplaca*)

- primary thallus** – basal thallus part of fine or coarse granules or squamules (*Figure 1a*)
- prothallus** – initial structure of hyphae without algae from which a lichenized thallus develops; often visible along the edge of the thallus or areoles (by characteristic colour)
- pruina** – a frost-like or flour-like surface layer composed of crystals
- pruinose** – covered with pruina
- pseudocyphella** – dot-like to fusiform or irregular pale areas of the thallus where the medulla is spreading to the surface (and not surrounded by specialized cells) (*Figure 2h*)
- pseudoparaphyses** – very often remotely septate, anastomosing and very narrow *paraplectenchyma* – tissue consisting of cell-like hyphae with \pm isodiametric lumina
- pyrenocarpous** – with perithecioid ascoma
- rhizine** – a root-like hair or thread for attaching mostly the foliose thallus to the substrate (*Figure 2i*)
- saxicolous** – on rock
- secondary thallus** – upright, fruticose part of the thallus (\rightarrow *podetium*) (*Figure 1a*)
- septum** (plural: *septa*) – cell wall or partition
- simple** (of spores) – without septa
- soralium** (plural: *soralia*) – a structure or region of a thallus producing soredia, resulting an appearance of flour-like, castor sugar-like or granular (*Figure 2f*)
- soredium** (plural: *soredia*) – a non-corticate, loose, tiny ball-like structure of photobiont cells and fungal hyphae deliberating from cracks or pores on the thallus surface (*Figure 2b, f*)
- squamule** – a tiny scale, without a cortex on its lower surface
- substrate** – underlying material of the thallus, surface of the habitat
- terricolous** – on soil
- thalline exciple** – margin of apothecium consists of both photobiont cells and fungal hyphae
- thallus** (plural: *thalli*) – the vegetative body of a lichen consisting of fungal hyphae (\rightarrow *mycobiont*) and algal (or cyanobacterial) cells (\rightarrow *photobiont*)
- tholus** – the thickened inner part of the ascus wall in the ascus apex; apical dome

tomentum (adjective *tomentose*) – felt-like mat of hyphae
true exciple – an exciple derived from the ascocarp lacking photobiont cells, usually of a different colour from the thallus (→ *lecideine*)

umbilicus – the central holdfast occurring in certain foliose lichens (e.g., *Lasallia*, *Umbilicaria*) (Figure 1e)

verrucose – covered with warts (verrucae)

ABBREVIATIONS

C – sodium- or calcium-hypochlorite (+ / – spot reaction resulting a characteristic colour or not reacting)

K – potassium-hydroxide (+ / – spot reaction resulting a characteristic colour or not reacting)

Pd – p-phenylene-diamine (+ / – spot reaction resulting a characteristic colour or not reacting)

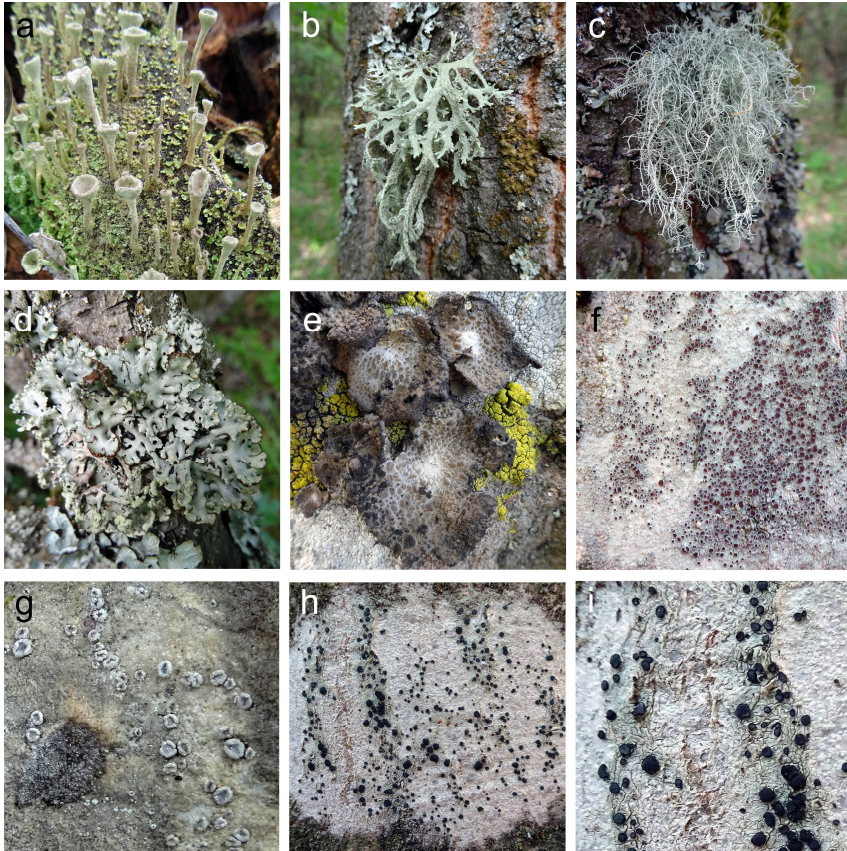


Figure 1. Main morphological characters of lichens: **a)** twofold growth form of foliose primary thallus and fruticose podetium (*Cladonia fimbriata*), **b)** fruticose growth form with flattened strapshaped lobes (*Evernia prunastri*), **c)** fruticose growth form with cylindrical branches (*Usnea* cf. *dasopoga*), **d)** foliose growth form (*Hypogymnia physodes*), **e)** foliose growth form with a central holdfast / umbilicus (*Lasallia pustulata*), **f)** crustose growth form with lecanorine apothecia (*Lecanora chlorotera*), **g)** crustose growth form with lecanorine apothecia (*Lecanora crenulata*), **h)**–**i)** crustose growth form with lecideine apothecia (*Lecidella elaeochroma*).

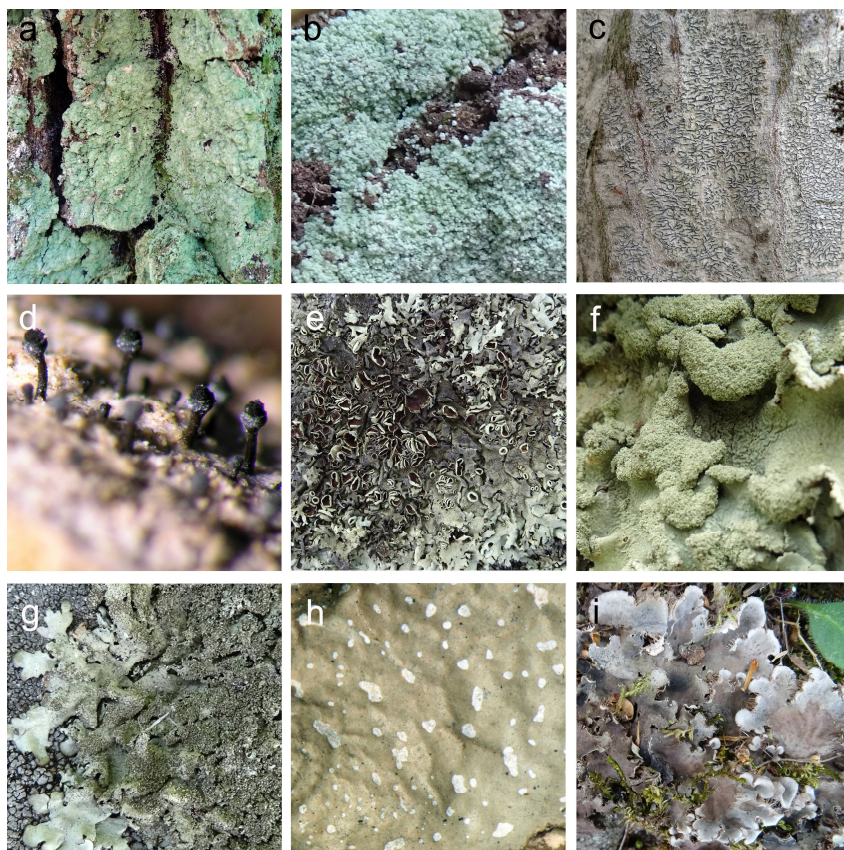


Figure 2. Main morphological characters of lichens: **a)-b)**, entirely sorediate sterile thallus (*Lepraria lobifcans*), **c)**, crustose growth form with elongated apothecia (*Graphis scripta*), **d)**, crustose growth form with stalked ascomata (*Calicium glaucellum*), **e)**, sexual reproductive structures: apothecia (*Xanthoparmelia conspersa*), **f)**, asexual reproductive structures: laminal soralia (*Flavoparmelia caperata*), **g)**, asexual reproductive structures: isidia (*Parmelia saxatilis*), **h)**, pseudocyphellae on the upper surface (*Cetrelia chicitae*), **i)**, rhizines on the lower surface (*Peltigera canina*).

GENERIC KEY TO LICHEN-FORMING FUNGI

- 1 Thallus consists of a basal primary thallus of fine or coarse granules or squamules and an erect secondary thallus part (*Figure 1a*)..... 2
 - Thallus uniformly crustose, foliose or fruticose..... 7
- 2(1) The erect part represents basidiomycete fruitbodies resembling other types of omphalinoid mushrooms or basidiomata club-shaped..... ***Lichenomphalia, Multiclavula***
 - Fruitbody if present, represents ascomycetes..... 3
- 3(2) Primary thallus of granules..... 4
 - Primary thallus of squamules..... 6
- 4(3) Basal thallus fine powdery leprose, erect thallus of short, delicate slender, white, cartilaginous, terete, branching pseudopodetia covered by an almost continuous, sorediate crust; apothecia unknown.....
 - ***Leprocaulon quisquiliare*** (syn. *L. microscopicum*)
 - Basal thallus granulose or small squamulose, erect thallus not sorediate..... 5
- 5(4) Basal thallus granulose or small squamulose, erect thallus solid white podetium (2–10 mm); apothecia (2–3 mm) brown reddish brown or rose, developing on the top of podetia; ascospores simple..... ***Baeomyces, Dibaeis***
 - Basal thallus of 0.2–1 mm non-corticate granules, erect thallus without soredia; podetia hollow (5–15 mm); apothecia rare (1–2 mm), arising on the tips of the podetia, ascospores simple or 1–3-septate..... ***Pycnothelia papillaria***
- 6(3) Erect thallus hollow, often with squamules (at least at the base) and cups; phyllocladia and cephalodia absent; ascospores simple..... ***Cladonia***
 - Erect thallus solid, without cups and squamules; granular, peltate or finger-like phyllocladia and often sessile, wrinkled to convoluted, pale brown to blackish cephalodia present; ascospores septate..... ***Stereocaulon***
- 7(1) Photobiont cyanobacterium (or filamentous green alga *Trentepohlia*), thallus grey, black or brown, often gelatinously swelling when wet..... 8
 - Photobiont green alga..... 25

Lichens with cyanobacterium photobiont

[and a filamentous species with *Trentepohlia* photobiont]

- 8(7) Thallus of fine branching filaments.....***Polychidium*,
Synalissa [*Racodium rupestre*, if photobiont green alga –
a central *Trentepohlia* chain surrounded by elongate,
straight, ± parallel-orientated septate hyphae; filaments
smooth]**
- Thallus foliose or consists of small granules or squamules
or dwarf fruticose.....**9**
- 9(8) Thallus foliose.....**10**
- Thallus consists of small granules or squamules or dwarf
fruticose.....**18**
- 10(9) Thallus swelling strongly when wet, stiff and fragile when
dry, black, grey, brown, can be white pruinose, hairy at
the edge, lower side black, green, bluish grey, sometimes,
yellowish, usually without rhizines, but occasionally with
fine white hairs; apothecia with characteristic margin,
ascospores colourless.....**11**
- Thallus not swelling strongly when wet, brown, grey,
lower side white, beige, or brownish, naked or densely
tomentose, or with rhizines arranged on veins,
photobiont *Nostoc* or other filamentous cyano-
bacterium.....**14**
- 11(10) Photobiont is not filamentous like a string of pearls,
however a single celled cyanobacterium; apothecium
lecanorine, but initially or even longer tiny spotlike,
reminiscent of a perithecium...***Phylliscum* group (*Anema*,
Gonohymenia, *Phylliscum*, *Thallinocarpon*, *Thyrea*)**
- Photobiont is filamentous like a string of pearls,
cyanobacterium *Nostoc*; apothecium with thalline or /
and proper margin, brown, reddish brown, widely
opened.....**12**
- 12(11) Thallus with paraplectenchymatic cortex, consists of a
single row of isodiametric to shortly rectangular cells
well recognizable in section.....***Leptogium* group
(*Leptogium*, *Pseudoleptogium*, *Scytinium*)**

- Thallus without a cortex, however the margin of the apothecium is often surrounded with a paraplectenchymatic cortex (pseudocortex)..... **13**
- 13(12)** Ascospores simple..... ***Lempholemma***
 - Ascospores with parallel septa or submuriform.....
***Collema* group (*Blennothallia*, *Callome*, *Collema*, *Enchylium*, *Lathagrium*, *Scytinium*, *Porocyphus*)**
- 14(10)** Apothecia immersed in the upper surface of the thallus... ***Solorina saccata***..... **8**
 - Apothecia not immersed, sessile or saddle-shaped..... **15**
- 15(14)** Apothecia are at lobe-ends..... **16**
 - Apothecia sessile on the thallus lobes or absent..... **17**
- 16(15)** Apothecia on the often uprising lobe surfaces or in saddle position; the lower surface ecorticate with anastomosing veins where bunches of rhizines arise..... ***Peltigera***
 - Apothecia on the lower surface, but convoluting to the upper surface, not in saddle position; lower surface corticate, without veins, but pubescent or with dense velvety beige, pale brown tomentum..... ***Nephroma***
- 17(15)** Lobes are usually wide, thallus large, stout, lower surface ecorticate, with rhizine carrying anastomosing veins or entirely tomentose..... ***Peltigera***
 - Thallus divided into small lobes, bluish grey or brownish, often forming rosettes; without veins on the lower surface..... ***Pannaria* group (*Fuscopannaria nebulosa*, *Protopannaria pezizoides*, *Vahliella leucophaea*)**
- 18(9)** Terricolous, also on bryophytes and plant debris..... **19**
 - Saxicolous..... **20**
- 19(18)** Thallus black, brownish black, crustose, small granulose or thicker, areolate or coralloid; apothecia, brown, reddish brown, perithecioid at first; photobiont single cells or cells in clusters with common gelatinous sheath *Chroococciopsis*..... ***Psorotichia* group**
 - Thallus of thicker black, greenish, bluish grey, brownish, olive granules or squamules; apothecia reddish brown, immersed in thallus; photobiont filamentous *Scytonema*.....
Heppia

- 20(18) Thallus of minute, grey, olive, umbilicate squamules of sorediate margin; lower surface pale; photobiont unicellular, *Chroococidiopsis*..... ***Peltula euploca***
 - Thallus without sorediate margin..... **21**
- 21(20) Thallus of tiny, hollow squamules, often rosette-shaped forming small cushions attached by a minute umbilicus; photobiont not filamentous, *Gloeocapsa*-type single celled or cells in clusters..... ***Phylliscum***
 - Thallus is not like that..... **22**
- 22(21) Apothecia usually present..... **23**
 - Apothecia can be absent..... **24**
- 23(22) Thallus endolithic in calcareous rocks; apothecia at first closed perithecia-like with white starlike margin, later while opening, the rose, orange disc appear; ascospores acicular, 4-celled, surrounded by halo; photobiont filamentous *Scytonema*..... ***Petractis clausa***
 - Thallus not endolithic; either closely attached, lobes flat or swollen, grey, brown, olive or black; apothecia black, lecideine, extended, not tiny spotlike; ascospores acicular or elliptic, parallel-septate, 2–4(–8)-celled (*Placynthium*) or ascospores simple, broad ellipsoid or globose, thallus dwarf fruticose, photobiont chroococcoid (*Peccania*) ***Placynthium, Peccania***
- 24(22) Lobes are often radiating at thalline edges or surrounded with greenish-, bluish-black hypothallus; photobiont filamentous *Scytonema*..... ***Placynthium***
 - Thallus without radiating lobes and hypothallus; photobiont single celled or cells in clusters, not clearly filamentous, gelatinous sheath is yellow-brown..... ***Psorotichia***

Lichens with green alga photobiont

- 25(7) Thallus fruticose..... **26 (FRUTICOSE LICHENS)**
 - Thallus crustose, squamulose, placodioid or foliose..... **41**

FRUTICOSE LICHENS

- 26(25) Thallus consists of an erect foliose or fruticose part (secondary thallus) and a basal granulose or squamulose part (primary thallus)..... **2**
- Thallus clearly fruticose (consisting of cylindrical or flattened branches, hanging or shrubby)..... **27**
- 27(26) On soil and terricolous bryophytes..... **28**
- On trees, shrubs, wood, rocks..... **34**
- 28(27) Thallus of ± erect, slightly branching flattened, guttered or convoluted lobes..... **29**
- Thallus of terete, cylindrical branches, sometimes angulate or ridged, hanging or erect..... **31**
- 29(28) Thallus olive, brown, dark brown, reddish or blackish brown, sometimes pale..... ***Cetraria*** (incl. ***Flavocetraria***)
- Thallus pale to bright or greenish yellow..... **30**
- 30(29) Thallus bright yellow, medulla yellow, uprising to vertical..... ***Cetraria*** (syn. ***Vulpicida***)
- Thallus pale yellow or pale greenish yellow, pale green, greyish green, flat or convolute to almost tubular..... ***Flavocetraria*** (key with ***Cetraria***)
- 31(28) Thallus section presents a solid structure of lobes..... **32**
- Thallus section presents a hollow, tubular structure of lobes..... **33**
- 32(31) Thallus of slightly branching white solid terete lobes covered richly with white squamules..... ***Stereocaulon***
- Thallus brown, lobes fine, thin, hairlike, slightly branching, mostly pendulous, with or without soralia..... ***Bryoria***
- 33(31) Thallus richly branching, 2-4 terminal branches may turn in the same or different directions..... ***Cladonia***
- Thallus slightly branching, with pointed or scyphose apex, basal part can be squamulose, soredia may occur on podetia..... ***Cladonia***
- 34(27) Thallus of flattened lobes of upper and lower surface different in colour (dorsiventral)..... **35**
- Thallus of flattened lobes or terete, cylindrical branches of ± uniform in colour (lower parts might be somewhat paler)..... **38**

- 35(34) Thallus yellowish green, pale green above, lobes 2–4 mm wide, 2–6(–10) cm long, without isidia, marginal soralia can occur, lower side white.....***Evernia prunastri***
 - Thallus of flattened lobes, greyish above, pale or blackish below, with or without cilia.....**36**
- 36(35) Thallus without cilia, lobes 1–5 mm wide, up to 10 cm long with laminal, cylindrical to coralloid isidia, lower surface white to rose at younger parts, older parts near to the attachment bluish black, black.....***Pseudevernia furfuracea***
 - Thallus with cilia.....**37**
- 37(36) Thallus grey, lobes firmly attached, 1–2 mm wide, forming rosettes (3–8 cm diam.) or more rarely widely spreading, sometimes ascending, with labriform soralia, lower surface white or tan; apothecia, very rare, disc brown-black; ascospores very thick walled, 1-septate, often with 1–3 additional small locules beyond the main locules, brown, ellipsoid-oblong, surface smooth, *Pachysporaria*-type.....***Heterodermia speciosa***
 - Thallus pale grey ± finely tomentose, lobes >2 mm wide, 3–5 cm long, without isidia and soralia, lower surface pale brownish white; apothecia occasional, 2–5 mm diam.; disc brown-black, often blue-grey pruinose; ascospores uniformly thin-walled except at the septum, *Physconia*-type, 34–41 × 17–21 µm, dark brown, wide ellipsoid, with rounded apices, with minute spines or ridges.....***Anaptychia ciliaris***
- 38(34) Thallus of ± erect, slightly branching flattened, guttered, channelled or convoluted lobes.....**39**
 - Thallus of terete, cylindrical branches, sometimes angulate or ridged, hanging, pendulous or erect.....**40**
- 39(38) Thallus yellowish green, greenish grey, lobes flattened, strap-shaped, single to numerous, often with soralia, mostly erect, shrubby.....***Ramalina***
 - Thallus grey, bluish grey, lobes are tubular and ascending with capitate rounded soralia at lobe ends.....***Hypogymnia tubulosa***

- 40(38) Thallus of terete, cylindrical branches, sometimes angulate or ridged, branching irregular, with or without fibrils, tubercles, papillae, pseudocyphellae, isidia, isidiomorphs and soredia, yellowish green, greenish grey, hanging or erect..... ***Usnea***
- Thallus of fine, thin, cylindrical, hair-like branches, brown, reddish brown, pale greyish brown, often with inconspicuous pseudocyphellae and tuberculate or fissure-like soralia, mostly hanging..... ***Bryoria***

Yellow, yellow-green, orange-red lichens of various morphology

- 41(25) Thallus (when dry) bright yellow, bright yellow-green to orange-red (pigments anthraquinones or pulvinic acid derivatives)..... **42**
- Thallus not as above; sometimes dull yellow-green owing to usnic acid..... **49**
- 42(41) Thallus K+ purple-red (pigments anthraquinones)..... **43**
- Thallus K± yellow, not purple-red (pigments pulvinic acid derivatives)..... **45**
- 43(42) On bark, wood or stone; schizidia absent; ascospores polarilocular..... **44**
- On calcareous soil, often spreading to mosses; schizidia often present; ascospores simple or 1-septate, not polarilocular.....
... ***Gyalolechia*** (syn. *Calogaya*, *Fulgensia*) (with ***Caloplaca***)
- 44(43) Thallus closely attached to substratum by entire lower surface, often placodioid, rhizines and other attachment organs absent..... ***Caloplaca*** (incl. ***Klauderuiella***)
- Thallus ± loosely attached to substratum by a basal sheath, attachment discs or rhizines.....
Xanthoria (incl. ***Galowayella***, ***Massjukiella***, ***Oxneria***, ***Rusavskia***, ***Zeroviella***)
- 45(42) Thallus foliose, markedly incised with ± granular or marginal soralia..... **46**
- Thallus placodioid to squamulose..... **47**

- 46(45) Thallus lobes to c. 1 cm wide; soralia marginal.....
.....**Cetraria pinastri** (syn. *Vulpicida pinastri*)
- Thallus lobes very small, 0.3–0.5(–1.5) mm wide; soralia
granular.....**Candelaria concolor**
- 47(45) Marginal lobes elongated, convex. Thallus irregularly
granular-isidiate towards centre with white-pruinose
marginal lobes.....**Candelariella medians**
- Marginal lobes not much different from internal
squamules, flattened.....**48**
- 48(47) Thallus bright greenish yellow to whitish grey, sorediate,
on soil; lobes pruinose; apothecia black; ascospores
septate.....**Arthrorhaphis citrinella**
- Thallus usually on rock, wood or bark; lobes not pruinose;
apothecia yellow; ascospores simple.....
.....**Candelariella vitellina**

Lichens of various morphology

- 49(41) Thallus foliose, squamulose or placodioid (with marginal
lobes differing from the centre), often attached to
substratum by rhizines, hapters or by a central disc
(umbilicus), often with stratified structure.....**50**
- Thallus crustose, continuous or rimose-cracked, areolate,
powdery, or evanescent, immersed, at least partially
embedded in the substratum....**114 (CRUSTOSE LICHENS)**
- 50(49) Thallus distinctly foliose, small to large, often rosette-
forming or strap-shaped, attached to substratum by
rhizines, hapters, tomentum, folds or by an
umbilicus.....**51 (FOLILOSE LICHENS)**
- Thallus squamulose, sometimes minutely so, squamules
mostly closely attached to substratum or elevated
towards margins, scattered, contiguous or overlapping, or
thallus placodioid and \pm crustose at the centre with
contiguous lobed margins.....
.....**86 (SQUAMULOSE OR PLACODIOID LICHENS)**

FOLIOSE LICHENS

- 51(50) Thallus attached to substratum at a central point by an umbilicus.....52
 - Thallus not attached to substratum by an umbilicus, attachments various.....54
- 52(51) Thallus dotted with abundant black perithecia on upper surface.....*Dermatocarpon* (incl. *Heteroplacidium*)
 - Thallus with apothecia or sterile.....53
- 53(52) Thallus surface with distinctive, coarse, oval pustules; isidia black, coralloid, very rarely fertile.....
*Lasallia pustulata*
 - Thallus surface without coarse pustules, surface smooth, wrinkled or ridged, sometimes cracked; soredia, phyllidia or thalloconidia sometimes present, or often fertile.....
*Umbilicaria*
- 54(51) Thallus lobes rounded.....55
 - Thallus lobes spreading, apothecia if present not immersed in thallus.....56
- 55(54) Apothecia single urceolate, ± immersed in the thallus, lower surface white or pale brown, tomentose, with rhizines, without lichen secondary metabolites.....
*Solorina saccata*
 - Apothecia at the edge of podetia (very rare), or the thallus sterile, lower surface white or pale yellow, rhizines and lower cortex are missing, usnic acid, fumarprotocetraric acid (Pd+).....*Cladonia foliacea*
- 56(54) Thallus lobes inflated, hollow in section, lower surface without rhizines.....57
 - Thallus lobes solid in section, flat or convex, with distinct lower cortex.....58
- 57(56) Thallus without perforations on the upper or lower surface.....*Hypogymnia*
 - Thallus with scattered round perforations <1 mm diameter on the upper surface.....*Menegazzia terebrata*
- 58(56) Lobes with tomentum on lower surface, wide-spreading. Cyphellae and pseudocyphellae absent. Thallus green-brown when dry, bright green when wet, with distinct depressions with a network of ridges on upper surface.....

- ***Lobaria pulmonaria***
- Lobes without tomentum on lower surface..... **59**
 - 59(58)** Thallus white, grey or brownish grey, lobes up to 5 mm wide, usually matt, often pruinose: spores brown, 1-septate (Physciaceae)..... **60**
 - Thallus yellow-green, grey or brown, lobes from 1 mm to several centimetres wide, usually ± shiny in younger parts, rarely pruinose; spores colourless, simple (Parmeliaceae)..... **66**
 - 60(59)** Lobes strap-like, erect, elongated and narrowly linear, lower cortex absent..... **61**
 - Lobes rounded or if linear, branching regularly, adnate, forming rosette-like thalli..... **62**
 - 61(60)** Lobes grey-green to white; thallus K+ yellow (atranorin).....
 - ***Heterodermia***
 - Lobes pale to dark brown; thallus K-..... ***Anaptychia***
 - 62(60)** Thallus whitish to bluish grey, maculate and sorediate or with isidia or lobules..... **63**
 - Thallus grey brown to brown, emaculate..... **64**
 - 63(62)** Upper cortex and hyphae running parallel to the upper surface (microscope)..... ***Heterodermia***
 - Upper cortex and hyphae with a cellular (pseudo-parenchymatous) structure (microscope)..... ***Physcia*** (with ***Physciella*** – differing in prosoplectenchymatous lower cortex)
 - 64(62)** Thallus ± loosely appressed to substratum, lobes pruinose at tips..... ***Physconia, Poeltonia***
 - Thallus closely appressed to substratum, lobes not pruinose at tips..... **65**
 - 65(64)** Rhizines absent or sparse..... ***Hyperphyscia adglutinata***
 - Rhizines numerous..... ***Phaeophyscia***
 - 66(59)** Thallus lobes brown throughout..... **67**
 - Thallus lobes yellow-green, green to grey-green or whitish..... **71**
 - 67(66)** Thallus erect, ± tufted. Lobes with distinct upper (darker brown) and lower (paler, sparseley rhizinate) surfaces.....
 - ***Nephromopsis chlorophylla*** (syn.: *Tuckermanopsis c.*)

- Thallus adnate to loosely overlapping, sometimes with irregular clusters of isidia or pale-coloured soralia. Rhizines present at least in the centre of the thallus, lobes loosely to closely attached to the substratum; substrata various.....**68**
- 68(67)** Medulla UV+ white, saxicolous.....*Xanthoparmelia*
- Medulla UV-, corticolous or saxicolous.....**69**
- 69(68)** Medulla C+ red (lecanoric acid).....*Melanelixia*
- Medulla C-.....**70**
- 70(69)** Medulla Pd- (usually without lichen secondary metabolites); lobes flat to concave.....*Melanohalea*
- Medulla usually Pd+ yellow or red; if Pd- then with convex lobes and whitish punctiform pseudocyphellae.....
.....*Melanelia, Montanelia*
- 71(66)** Pseudocyphellae present on upper surface, punctiform or effigurate.....**72**
- Pseudocyphellae absent.....**74**
- 72(71)** Pseudocyphellae effigurate.....*Parmelia*
- Pseudocyphellae punctiform.....**73**
- 73(72)** Lobes overlapping with wavy crisped margins and marginal soralia.....*Cetrelia*
- Lobes with soredia developing from laminal and marginal pseudocyphellae.....*Punctelia* (greyish, with atranorin);
.....*Flavopunctelia* (yellowish green, with usnic acid)
- 74(71)** Rhizines branched dichotomous or squarrose (simple rhizines may also be present).....*Hypotrachyna*
- Rhizines simple (or at most forked).....**75**
- 75(74)** Thallus closely appressed to substratum.....**76**
- Thallus not appressed to substratum with loosely overlapping lobes.....**80**
- 76(75)** Lower surface pale tan to white. Thallus grey-white, K+ bright yellow, densely isidiate.....*Imshaugia aleurites*
- Lower surface dark brown to black.....**77**
- 77(76)** Thallus yellow-green or yellow-grey (usnic acid in cortex).....**78**
- Thallus grey or blue-grey (without usnic acid in cortex). Cilia present in axils and/or on the apices of lobes; soralia absent; medulla UV-.....**79**

- 90(89) Ascospores simple..... ***Catapyrenium***
 - Ascospores 1-septate..... ***Placidiopsis***
- 91(87) Thallus of appressed or ascending, individual squamules, overlapping or ± dispersed, not forming radiating and elongated marginal lobes..... **92**
 - Thallus ± forming rosettes or placodioid when ± crustose in centre with distinctly elongated radiating marginal lobes..... **106**
- 92(91) Asci 50- to 300-spored; ascospores minute, 3–6(–13) × 1–3(–6) µm..... ***Acarospora*** (incl. ***Caeruleum***, ***Glypholecia***, ***Myriospora***, ***Polysporina***, ***Sporastatia***)
 - Asci to 8-spored; ascospore size various; or thallus sterile..... **93**
- 93(92) Thallus sorediate or with clusters of coarse, irregular, isidia-like protuberances, squamules wider, than 0.3 mm, generally not green..... **94**
 - Thallus without soredia and isidia..... **97**
- 94(93) Squamules ± ascending, at least at edges, often ± elongate, margins frequently incised; small podetia frequently present..... ***Cladonia***
 - Squamules ± closely appressed to substratum, mostly rounded, with ± entire margin; podetia absent..... **95** (see also ***Baeomyces rufus***)
- 95(94) Squamules C+ red, squamules orientated in one direction, overlapping, the margin and lower surface sorediate; thallus brownish, Pd–..... ***Hypocenomyce scalaris***
 - Squamules C–..... **96**
- 96(95) Squamules peltate, with a conspicuously darker central area; ascospores (1–)3–5-septate..... ***Stereocaulon***
 - Squamules not peltate, without a darker central area; ascospores simple or 1-septate; soredia lip-shaped, ± restricted to marginal soralia; ascospores simple; on peat or wood; cortex Pd–; on peat or rotting wood..... ***Trapeliopsis***
- 97(93) Squamules ± ascending, at least at edges, often ± elongate, margins frequently incised; often green; small podetia frequently present..... ***Cladonia***
 - Squamules ± closely appressed to substratum, mostly rounded, with ± entire margin; podetia absent..... **98**

98(97)	Thallus on wood or bark; squamules mostly >0.5 mm, bullate or flattened, matt or shiny; apothecia black; ascospores simple to 1(-3)-septate.....	
	<i>Hypocenomyce scalaris</i>
-	Thallus on rocks or soil.....	99
99(98)	Thallus with schizidia; squamules minute, forming a ± continuous crust.....	<i>Baeomyces</i>
-	Thallus without schizidia; squamules varied.....	100
100(99)	Thallus C+ red.....	<i>Trapelia glebulosa</i>
-	Thallus C-.....	101
101(100)	Thallus with lip-shaped soralia on lobe ends; on peaty turf or wood.....	<i>Trapeliopsis</i>
-	Soralia absent.....	102
102(101)	Apothecia without a thalline margin.....	103
-	Apothecia with a thalline margin; thalline margin smooth; thallus with appressed, ± overlapping squamules; squamules yellow-grey or grey-green; ascospores simple.....	<i>Squamarina</i>
103(102)	Ascospores simple.....	104
-	Ascospores colourless, 1- to 5-septate.....	105
104(103)	Ascospores nearly globose; on siliceous rock.....	<i>Schaereria</i>
-	Ascospores ellipsoid; on soil or limestone; Apothecia brown-black; on soil amongst limestone rocks.....	<i>Psora, Romjularia</i>
105(103)	Paraphyses free, each with a distinct swollen apical cell, covered by a pigmented cap.....	<i>Thalloidima, Toninia, Toniniopsis, Xylopsora</i>
-	Paraphyses conglutinated, without cap.....	<i>Bilimbia lobulata</i>
106(91)	Thallus sorediate, leprose or with irregular, isidia-like protuberances.....	107
-	Thallus not sorediate, without irregular, isidia-like protuberances.....	111
107(106)	Thallus surface leprose.....	<i>Lepraria, Botryolepraria, Chrysothrix, Leproplaca</i>
-	Thallus sorediate or isidiate.....	108

- 108(107) Thallus C+ red; soralia developing from the sides of areoles or cracks in the thallus.....
..... ***Trapelia placodioides***
- Thallus C-..... **109**
- 109(108) Thallus K+ yellow, thallus pruinose, with distinct lobes closely contiguous for most of their length; apothecia without a thalline exciple.....
..... ***Diploicia canescens***
- Thallus K-; thallus non-pruinose; apothecia with a thalline exciple..... **110**
- 110(109) Thallus lobes corticate; soralia discrete; thallus greenish to brown..... ***Hyperphyscia adglutinata***
- Thallus not corticate; soredia irregular and confluent; thallus whitish..... ***Kuettlingeria teicholyta***
- 111(106) On soil; thallus ± white, or white-pruinose; without papillae; apothecia frequent, sessile; on calcareous soil; apothecia with a thalline exciple; ascospores colourless, simple..... ***Squamarina lentigera***
- On rocks..... **112**
- 112(111) Thallus ± crustose with elongated marginal lobes; apothecia immersed; asci >200-spored; thallus C+ red.....
..... ***Sporastatia***
- Thallus squamulose, apothecia sessile; asci <8-spored, thallus C-..... **113**
- 113(112) Ascospores 1-septate; on limestone and coastal serpentine..... ***Solenopsora***
- Ascospores simple; on a range of substrata, especially nutrient enriched rocks and building materials.....
..... ***Lecanora*** (placodioid species)

CRUSTOSE LICHENS

- 114(49) Thallus fertile (fruits globose, volcano-like, disc-like, elongate, stalked, shelf, or mushroom-like).....
..... **Fertile thalli**
- Crustose thalli with perithecioid ascomata.....
..... Fertile thalli **KEY 1**
- Crustose thalli with stalked apothecia.....
..... Fertile thalli **KEY 2**

- Crustose thalli with lirellate (elongated) apothecia..... Fertile thalli **KEY 3**
- Crustose thalli with rounded apothecia..... Fertile thalli **KEY 4**
- Thallus sterile (often with isidia or ± delimited soredia, or entirely leprose)..... **Sterile thalli** (not treated)

Fertile thalli KEY 1 – Crustose thalli with perithecioid ascomata

- 1** Thallus squamulose of squamules < 1cm or placodioid (central crustose part surrounded by elongated lobes)...**2**
- Thallus crustose, granulose or inconspicuous..... **6**
- 2(1)** Ascospores muriform, colourless or brown..... **3**
- Ascospores simple or septate, colourless..... **4**
- 3(2)** Thallus greenish brown (turning yellowish, greyish brown in herbarium) squamules of 0.3–1 cm, perithecia conical or barrel shaped with verrucose surface, among squamules or slightly immersed in the thallus. Without hymenial algae..... **Agonimia tristicula**
- Thallus brown, greyish or olive brown, squamulose with squamules of 0.5–2.5(–4) mm. With hymenial algae..... **Endocarpon**
- 4(2)** Ascospores 1–3-septate..... **Placidiopsis, Placopyrenium, Thelidium**
- Ascospores simple..... **5**
- 5(4)** Thallus of single or confluent squamules..... **Placidium, Placocarpus, Catapyrenium**
- Thallus of areolae, with larger marginal areolae or rarely of squamules..... **Verrucaria, Verruculopsis**
- 6(1)** Ascospores slightly or clearly muriform, at least with one longitudinal septum..... **7**
- Ascospores simple or transverse septate..... **9**
- 7(6)** With small sphaerical or cylindrical algae in the hymenium; saxicolous..... **Staurothele**
- Without hymenial algae..... **8**
- 8(7)** Perithecia conical or barrel shaped with verrucose surface, paraphyses missing. Thallus granulose or squamulose. On soil, mosses, plant remnants..... **Agonimia tristicula**

- Perithecia different, not conical or barrel shaped, not with verrucose surface, hemisphaerical to entirely immersed paraphyses dissolved while ripening. Thallus obvious to inconspicuous. On calcareous and siliceous rock, also on soil, mosses, plant remnants.....
.....***Polyblastia, Verrucula***
- 9(6) Ascospores various, simple, 1- to multiseptate or muriform..... **10**
- Ascospores septate..... **21**
(with submuriform/muriform at ***Gyalecta***)
- 10(9) On bark.....***Strigula, Thelenella, Chromatochlamys***
- On rock, soil, mosses, plant remnants..... **11**
- 11(10) Ascospores small, in a large number in the asci.....
.....***Thelopsis, Thelocarpon***
- Ascospores 1–8 in the asci..... **12**
- 12(11) On bark, wood..... **13**
- On soil, mosses, rocks..... **15**
- 13(12) Ascospores needle-shaped, thin acicular- fusiform, slightly curved, under 3.5 µm wide.....***Leptorhaphis***
- Ascospores ellipsoid..... **14**
- 14(13) Ascospores thin-walled, 3–11 µm, perithecia black, up to 0.7 mm, ± immersed.....***Bagliettoa, Verrucaria***
- Ascospores thick-walled, mostly at least 30 µm wide.....
.....***Pertusaria***
- 15(12) Ascospores at least partly wider than 40 µm.....
.....***Pertusaria* (incl. *Lepra*)**
- Ascospores up to 40 µm wide..... **16**
- 16(15) On soil, mosses..... **17**
- On rocks..... **19**
- 17(16) Paraphyses obvious, not branching. Thallus inconspicuous when dry, mucous when wet.....***Thrombium***
- Paraphyses missing in developed perithecia. Thallus well developed..... **18**
- 18(17) Thallus of areoles or squamules, grey to brown.....
.....***Catapyrenium***
- Thallus of tiny greyish green goniocytangia-like granules of 0.015–0.04 mm or thin.....***Verrucaria***
- 19(16) Ascospores very thin, threadlike to cylindrical or clavate.....
.....***Sarcopyrenia***

- Ascospores ellipsoid to sphaerica..... **20**
- 20(19)** Photobiont cyanobacterium (*Gloeocapsa* photobiont with a red-brown K+ purple gelatinous sheaths); hamathecium of periphysoids near to the ostiole; interascal filaments either absent or inconspicuous and much shorter than the asci; asci thin-walled, with pointed tips..... ***Cryptothele rhodosticta***
- Photobiont green alga; hamathecium of periphyses and periphysoids, interascal filaments absent; asci clavate, wall thickened above.....
- ***Parabagliettoa, Psoroglaena, Verrucaria***
- 21(9)** Ascospores brown, thallus with *Trentepohlia* photobiont..... ***Pyrenula*** (incl. *Eopyrenula*)
- Ascospores colourless or slightly coloured..... **22**
- 22(22)** Ascospores septate or muriform. Ascomata flat to urceolate apothecia. Thallus thin, smooth, photobiont *Trentepohlia*..... ***Gyalecta***
- Ascospores transversely septate..... **23**
- 23(22)** Ascospores 1(–3)-septate with thick walls, clavate, oblong or fusiform, in single line in asci (uniseriate), paraphyses sparsely branched or anastomosing.....
- ***Acrocordia, Anisomeridium, Arthopyrenia, Naetrocymbe, Naevia***
- Ascospores with several septa..... **24**
- 24(23)** Ascomata perithecioid, exciple pseudoparenchymatous. Ascospores threadlike, acicular-fusiform, 1–3-septate mostly without photobiont or loosely associated with *Trentepohlia*..... ***Leptorhaphis***
- Ascospores acicular-fusiform, 4–8-celled; paraphyses are unbranched or branched, thallus without soredia.....
- ***Porina, Pseudosagedia, Swinscowia***

Fertile thalli KEY 2 – Crustose thalli with stalked apothecia

- 1** Apothecium with a dry spore mass (mazaedium)..... **2**
- Apothecium without a dry spore mass (mazaedium), stalk white..... **7**
- 2(1)** Apothecium with short stalk or without stalk, ascospores with brown wall..... **3**

- Apothecium obviously stalked..... **4**
- 3(2)** Ascospore simple, on bark..... *Sphinctrina*
- Ascospore 2-celled to submuriform, on wood.....
..... *Calicium notarisii*
- 4(2)** On siliceous rock..... **5**
- On bark or wood..... **6**
- 5(4)** Thallus, stalk and lower part of apothecia is yellow,
yellow-green. Ascospore simple, pale.....
..... *Chaenotheca furfuracea*
- Mazaedium brown, ascospore light brown, not clearly 1-3
septate..... *Chaenotheca, Sclerophora*
- 6(4)** Mazaedium brown, ascospore light brown, not clearly 1-3
septate..... *Chaenotheca, Sclerophora*
- Mazaedium black, ascospore dark brown, 2-celled.....
..... *Calicium*
- 7(1)** Hymenium jelly I+ blue; apothecium pink.....
..... *Dibaeis baeomyces*
- Hymenium jelly I-; apothecium reddish brown.....
..... *Baeomyces rufus*

Fertile thalli KEY 3 – Crustose thalli with lirellate (elongated) apothecia

- 1** Apothecia lacking true exciple or rarely rudimentary and
developed only laterally..... **2**
- True exciple present; photobiont various..... **3**
- 2(1)** Ascospores 1- to 7-septate, ovoid to oblong-ovoid.....
..... *Arthonia, Bryostigma, Diarthonis*
- Ascospores submuriform to strongly muriform, ovoid-
ellipsoid..... *Arthothelium*
- 3(1)** Ascospores simple, outer surface of apothecium pale
brown; paraphyses simple or sparingly branched.....
..... *Xylographa*
- Ascospores multiseptate or muriform..... **4**
- 4(3)** Ascospores I+ violet, thick-walled, their cells with lens-
shaped, round or oval lumina; hamathecium of simple
paraphyses. Ascospores not coloured when mature,
occasionally becoming brown when old; apothecia with
surfaces becoming longitudinally grooved or ridged and

- narrow discs..... **Graphis**
- Ascospores 1–, with cylindrical or cuboid lumina, colourless or not; hamathecium variable..... **5**
- 5(4)** Ascospores brown. True exciple not friable; ascospores often becoming rough..... **Opegrapha**
- Ascospores colourless. Lateral exciple in section well-developed, brown to black..... **6**
- 6(5)** Thallus Pd+ yellow; on rock; ascospores 3(–5)-septate.....
..... **Psoronactis (Lecanactis) dilleniana**
- Thallus not Pd+ yellow.....
..... **Opegrapha (incl. Alyxoria, Zwackhia, Gyrographa)**

Fertile thalli KEY 4 – Crustose thalli with rounded apothecia

- 1** Thallus yellow, orange, orange-red, K+ deep red immediately. Ascospores 1(–3)-septate, mostly thick walled..... **Caloplaca (incl. Athallia, Blastenia, Calogaya, Cerothallia, Flavoplaca, Seawardiella, Solitaria, Squamulea)**
- Characters different from the above ones. If thallus is yellow, reaction is not as above..... **2**
- 2(1)** Ascospores multicelled..... **3**
- Ascospores simple..... **42**
- 3(2)** Ascospores muriform (both with longitudinal and transverse septa) **4**
- Ascospores only with transverse septa..... **9**
- 4(3)** Apothecia grey to white, similar to soralia, immersed in the whitish grey thallus. Thallus K+ red, on bark. Ascospores ellipsoid, 1–2 in ascus..... **Phlyctis**
- Apothecia not similar to soralia, not, covered with whitish granules..... **5**
- 5(4)** Apothecia pink, brown, yellowish, orange to almost colourless. Thallus with *Trentepohlia*, or usual green algae..... **Gyalecta, Gyalidea**
- Apothecia black, but also grey pruinose ones..... **6**
- 6(5)** Ascospores soon brown to green coloured..... **7**
- Ascospores remain long colourless or with a pale hue / tone. Apothecia black, pruinose. Paraphyses richly branched and forming a net. Thallus well developed, with

- areoles. On rocks..... ***Rhizocarpon***
- 7(6) Apothecium disc deeply sitting from the beginning, true and thalline exciple turning inwards with narrow opening, at first black, later becoming pruinose. Thallus thick, grey to ochraceous, C+ red. On rock, soil, mosses.....
- ***Diploschistes***
- Apothecium disc not immersed, no thalline margin or white thalline margin. Thallus C-..... **8**
- 8(7) Ascospore with halo at least when young, Paraphyses richly branched and forming a net. Thallus well developed, with areoles. On rocks..... ***Rhizocarpon***
- Ascospore without halo. Paraphyses simple of forking. On various substrates..... ***Diplotomma***
- 9(3) Ascospores colour, greenish, olivaceous, dark brown..... **10**
- Ascospores colourless (slightly coloured when old)..... **13**
- 10(9) Photobiont *Trentepohlia*. Apothecia without a margin, brown to black, occasionally pruinose. Excipulum reduced, thin. Paraphyses netlike and anastomosing. Ascospores 2- or multicelled, brown only with ripening.....
- ***Arthonia, Coniocarpon***
- Photobiont coccoid green alga..... **11**
- 11(10) Paraphyses richly branched and anastomosing. Ascospores halonate at least when young. Thallus usually well developed, areolate. On rocks..... ***Rhizocarpon***
- Paraphyses simple or forked. Ascospore not halonate. On bark and rock..... **12**
- 12(11) Apothecia with thalline margin. Hypothecium mostly colourless or pale brownish. Septa of apothecia unevenly thickened..... ***Rinodina*** (incl. ***Helmutiopsis***)
- Apothecia without thalline margin (no algae in the margin) or immersed in thallus. Hypothecium dark. Septa evenly thickened..... ***Buellia*** (incl. ***Amandinea, Dimelaena, Epilichen, Monerolechia, Tetramelas***)
- 13(9) Ascospore bipolar 2-celled, with small opening in the middle of the wall. The wall can be thin or thick. Apothecium yellow, orange, red or orange-brown, K+ dark red... ***Caloplaca*** (incl. ***Athallia, Kuettlingeria, Laundonia, Olegblumia, Opeltia, Pisutiella, Pyrenodesmia, Rufoplaca, Variospora, Xanthocarpia***)

-	Ascospore not bipolar, septae thin.....	14
14(13)	Photobionta <i>Trentepohlia</i>	15
-	Photobionta coccoid green alga.....	26
15(14)	Ascospores 2-celled.....	16
-	Ascospores with more cells.....	18
16(15)	Paraphyses netlike, anastomosing. Exciple reduced. Apothecia without margin.....	<i>Arthonia</i>
-	Paraphyses simple.....	17
17(16)	Apothecium whitish to orange, with concave to flat disc. Hypothecium and excipulum colourless. Ascospores fusiform or ellipsoid. Thallus thin, grey to green. On bark and mosses.....	<i>Coenogonium</i>
-	Apothecium dark, black or brown, round. Hypothecium and excipulum dark or pale. Ascospore remaining colourless, cells with the same size.....	<i>Catillaria</i>
18(15)	Apothecium not black.....	19
-	Apothecium black, sometimes with pruina.....	21
19(18)	Ascospores 16 or more in the asci.....	<i>Gyalecta</i>
-	Ascospores up to 8 in the asci.....	20
20(19)	Apothecia with margin.....	<i>Gyalecta</i>
-	Apothecium without margin.....	<i>Arthonia</i>
21(18)	Ascospores multicelled, needle-shaped, threadlike, 50–80(–100) × 1.5–3 µm, fragmented into short parts. Apothecia black, thallus whitish, on <i>Quercus</i> bark.....	<i>Bactrospora (Lecanactis) corticola</i>
-	Ascospores shorter, mostly fusiform, not fragmented into smaller parts.....	22
22(21)	Hamathecium of paraphysoids. Asci clavate, apex thickened. Ascospores without a halo. Apothecia grey, with pruina.....	<i>Dirina massiliensis</i>
-	Hamathecium of paraphyses. Ascospores with or without a halo.....	23
23(22)	Paraphyses simple or forked, anastomosing in lower part. Ascospore 4-celled, with halo. Asci thinwalled Apothecia black. On calcareous rock.....	<i>Sagiolechia</i>
-	Paraphyses branched and anastomosing. Asci thickwalled.....	24

- 24(23) Apothecium peritheciumpulike, disc tiny dotlike. Dark exciple develops only sidewards. Thallus dark brown with pale soralia, prothallus represented as a black line.....
 ***Enterographa zonata***
 - The disc not dotlike. Dark exciple develops also below, a conelike sheath develops..... **25**
- 25(24) Apothecium not round, disc often with rim and umbilicus. Sheath is black, often strongly developed below. On rock..... ***Opegrapha***
 - Apothecium round, often pruinose, disc shiny, with proper margin, sheath blackish brown. On rock and bark..... ***Dendrographa (Lecanactis) latebrarum***
- 26(14) Apothecium with a starlike opening, a thalline cover splits radially from a central pore as the structure expands, partly sunked in calcareous rock. Photobiont *Scytonema*.....
 ***Petractis clausa***
 - Apothecium edge not starlike. Not with *Scytonema*, but green algae..... **27**
- 27(26) Apothecium disc yellow, orange, brown orange, K+ deep red, ascospores 2-celled..... ***Caloplaca* (incl. *Athallia*)**
 - If apothecium disc yellow to orange, not immediately K+ deep red..... **28**
- 28(27) Ascospores large of several hundred μm , 2-celled, with wall of several layers. Apothecia immersed by 1–3 in a thalline wart. Thallus thick, white to grey, C+ red. On plant remnants, mosses..... ***Varicellaria***
 - Ascospore smaller, thinwalled..... **29**
- 29(28) Apothecium margin with algae (colour similar to that of thallus – lecanorine) or apothecia immersed in thalline warts. On plant remnants, mosses and wood..... **30**
 - Apothecium margin without algae (colour similar to that of disc – lecideine, biatorine) or without a margin..... **32**
- 30(29) Ascospores at least 30 μm long, fusiform or needleshaped with (1–)3–7 transverse septa. Apothecium red, K+ dark red or blue, or apothecium rarely brown, not pruinose, concave or swollen..... ***Haematomma***
 - Ascospore shorter, ellipsoid, simple to 2–4-celled, up to 8 in asci. Apothecium not red..... **31**

- 31(30) Apothecium disc yellow, dirty yellow, brownish yellow, K- or slightly reddish. Thallus yellow or grey, ascospores simple to 2-celled..... **Candelariella**
 - Apothecia brown, reddish brown to black, white pruinose, c. 1mm, thalline margin soon disappearing, ascospore 2- or 4-celled..... **Lecania**
- 32(29) Thallus of thick, more seldom thinner squamules. Apothecia black, occasionally pruinose. Ascospore fusiform or needleshaped, 2–8-celled, on soil, moss, calcareous, more seldom siliceous rock.....
 **Toninia, Toniniopsis, Thalloidima, Porpidinia**
 - Thallus not squamulose, crustose, even, not differentiated or areolate..... **33**
- 33(32) On thin branches and conifer needles and evergreen deciduous trees..... **34**
 - On other habitat..... **35**
- 34(33) Apothecia pale, whitish, beige, or pink, brown, orange. Excipulum of spherical to ellipsoid or angulate cells. Ascospores 2-celled or 2–4(–6)-celled, very thin (up to 1.5–2 µm)..... **Bacidina**
 - Apothecia dark brown, dark reddish brown to black, soon without margin, excipulum of radiating, branched and anastomosing hyphae..... **Scoliciosporum**
- 35(33) Hypothecium and excipulum missing, hymenial gelatine missing, I- . Apothecia flat or swollen, thallus fine granulose of goniocystangia, ephemeral..... **Vezeadaea**
 - Hypothecium well developed, hymenial gelatine present, I+ blue..... **36**
- 36(35) Ascospores 2-celled, not needleshaped or threadlike..... **37**
 - Ascospores multiseptate, if needleshaped or threadlike or wider up to 4 µm. Apothecia pale to brown, with obvious margin at least when young, exciple well developed, proso- or paraplectenchymatic (consisting of radial or cellular hyphae)..... **Bacidia** (incl. **Aquacidia, Bellicidia, Bibbya, Coppinsidea, Mycobilimbia**)
- 37(36) Apothecia frequent, zeorine, immersed, 0.5–2(–4) mm across, with a brownish black, concave to flat, smooth disc, triangular lobes radiating all around the apothecia. Asci many-spored, narrowly clavate, thin-walled, apically

- thickened, the wall I-. Ascospores 1-septate, colourless, narrowly ellipsoid (7-)11-17 × (2-)3-4 μm. On loess.....
.....***Gyalidea asteriscus***
- Apothecia not with startlike appendages, asci not with many ascospores, substrate different.....**38**
- 38(37)** Apothecium concave, small, up to 0.4 mm, yellowish to pale brownish, asci thickened in upper part, I+ reddish brown. Paraphyses simple. On wood.....***Absconditella***
- Apothecia flat or swollen.....**39**
- 39(38)** Apothecia black.....**40**
- Apothecia pale to black.....**41**
- 40(39)** Apothecium margin persistent. Thallus effuse, thin, ± evanescent; soralia large, grey-white to glaucous, granular, hypothecium colourless or pale brown, K-. Ascospores broadly to fusiform-ellipsoid, the walls rather thick; not with halo (perispore absent). On bark.....
.....***Megalaria pulverea***
- Apothecium margin disappearing. Hypothecium black to brown. Ascospores with halo at least when young. On rock.....***Rhizocarpon***
- 41(39)** Paraphyses simple or forked. Apothecium pale to black, margin obvious at least when young.....***Catillaria***
(key with ***Biatora***, ***Catinaria***, ***Cliostomum***)
- Paraphyses branching. Apothecium not brownish black or black, without or with inconspicuous margin.....
.....***Micarea***, ***Leimonis***
- 42(2)** Asci with many (above 50) tiny ascospores.....**43**
- Asci with 1-16 (sometimes 32) ascospores, mostly 8.....**47**
- 43(42)** Apothecia / thallus warts with apothecia with small, dotlike opening, peritheciump-like, pale or greenish yellow. Thallus inconspicuous.....***Thelocarpon***
- Apothecium with widely opening disc, if less wide, then thallus well developed.....**44**
- 44(43)** On rock.....**45**
- On bark, wood, plant remnants.....**46**
- 45(44)** Apothecia mostly with brown, reddish brown disc (sometimes black, dotlike), with thalline margin or immersed in thallus.....***Acarospora***

- Apothecia black to reddish brown, white pruinose, with proper margin, no thalline margin. Thallus obvious or inconspicuous.....**Sarcogyne**
- 46(44) Apothecium blackish, ±flat, with obvious thalline margin. Hypothecium pale, epihymenium dark brown. Ascospores 5–6.5 × 2.5–3.5 µm. Thallus pale greyish, olive-green to olive-brown, warty, granulose.....**Maronea constans**
- Apothecium without thalline margin, with or without proper margin, whitish, yellowish, ochraceous, brown, reddish brown or blackish, soon swollen. Hypothecium pale to brownish.....
.....**Biatorrella, Biatoridium, Piccolia, Sarcosagium**
- 47(42) Apothecia yellow, orange or rust coloured, immediatly strongly swollen, without margin and fast K+ deep red.....
.....**Protoblastenia**
- With other characters, if thallus or apothecium yellow, then not K+ deep red.....**48**
- 48(47) Ascospores very large, mostly above 30 µm, widely ellipsoid or spherical.....**49**
- Ascospores less large, mostly up to 30 µm.....**51**
- 49(48) Apothecia mostly flat, with thick thalline margin, disc wide, rose-brownish, white pruinose. Asospores ± thickwalled.....**Ochrolechia** (key with **Pertusaria**)
- Apothecium different.....**50**
- 50(49) Ascospores ±thickwalled, mostly ovoid, 1–8 in ascus. Apothecium peritheciumlike, disc narrow, dotlike, immersed in thallus warts, rarely black, widely opened, not immersed in thallus warts, often rich in lichen secondary metabolites.....**Pertusaria**
- Ascospores ± thinwalled (up to 2.5 µm), 4–8 in ascus, widely ellipsoid 30–60 × 20–40 µm. Apothecia deeply immersed in the 1.5 mm wide thallus warts, disc black, dotlike at first, then opens more widely (to 0.5 mm). Thalline margin somewhat black inside. Thallus uneven, whitish, pale grey. On mosses, plant rests.....
.....**Megaspora verrucosa**
- 51(48) Apothecium yellow, lemon yellow, ochraceous, brownish or olive yellow, often also thallus is yellow.....**52**
- Apothecium not yellow.....**54**

- 52(51) Ascospore 4–7 × 1–2 µm, up to 8 in ascus. Thallus fine farinose, lemon yellow, greenish yellow. Apothecium swollen, without margin. Paraphyses ±simple, thin. Usually on vertical position on siliceous rock.....
..... ***Psilolechia lucida***
- Ascospores larger..... **53**
- 53(52) Ascospores to 8 or 12–32, mostly cylindric. Apothecia flat to swollen, mostly with margin, lecanorin, seldom biatorince, yellow. Thallus fine granulose or sometimes areolate, sometimes finely sorediate. Tholus amyloid only at lower part – usually with pulvinic acid derivatives.....
..... ***Candelariella***
- Ascospores up to 8. elliptic. Apothecium lecanorine, pale yellow, pale greenish yellow, brown yellow. Thallus areolate or sorediate, the entire tholus is amyloid, rich in lichen secondary metabolites, mostly without pulvinic acid derivatives..... ***Lecanora, Bryonora***
- 54(51) Photobiont *Trentepohlia*..... ***Hymenelia epulotica***
- Photobiont not *Trentepohlia*, usually coccoid green alga....
..... **55**
- 55(54) Apothecium lecanorine, with thaline margin..... **56**
- Apothecium biatorine or lecideine, with thaline margin....
..... **57**
- 56(55) Apothecium disc concave, deeply immersed in thallus. On rock..... ***Aspicilia, Circinaria, Ionaspis, Lecaimmeria, Lobothallia***
- Apothecium disc flat to swollen. Apothecium normally sessile, only rarely immersed.....
Lecanora (incl. ***Eiglera, Glaucomaria, Myriolecis, Olegblumia, Placynthiella, Polyozosia, Rhizoplaca, Rimularia, Straminella, Tephromela***)
- 57(55) Hypothecium and excipulum missing, hymenial gelatine missing, I–. Apothecia flat or swollen, thallus fine granulose of goniocystangia, ephemeral..... ***Veздаea***
- Apothecia flat to swollen, immersed or sessile. Paraphyses simple or branched and anastomosing. Ascospores elongated, narrow or wide elliptic or spherical. Hypothecium colourless or brown black. Asci obvious, K+I reaction is strong + blue, hymenium I+ blue..

.....**Lecidea**
(incl. **Ainoa**, **Bryobilimbia**, **Carbonea**, **Clauzadea**,
Farnoldia, **Fuscidea**, **Lambiella**, **Porpidia**, **Ropalospora**,
Schadonia, **Steinia**, **Strangospora**)

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SUPPLEMENT TO GENERIC KEY TO LICHEN-FORMING FUNGI IN HUNGARY

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Checklist of genera (269) with the number of species numbers included (940) are in brackets [some dubious species are not recorded].

Absconditella (1)	Bryobilimbia (1)	Dermatocarpon (4)
Acarospora (16)	Bryonora (1)	Diarthonis (1)
Acrocordia (3)	Bryoria (3)	Dibaeis (1)
Agonimia (6)	Bryostigma (2)	Dimelaena (1)
Ainoa (1)	Buellia (7)	Diploicia (1)
Alyxoria (3)	Caeruleum (1)	Diploschistes (6)
Amandinea (1)	Calicium (8)	Diplotomma (8)
Anaptychia (1)	Callome (1)	Dirina (1)
Anema (3)	Calogaya (4)	Eiglera (1)
Anisomeridium (4)	Caloplaca (16)	Enchylium (4)
Aquacidia (1)	Candelaria (1)	Endocarpon (4)
Arthonia (10)	Candelariella (8)	Enterographa (1)
Arthopyrenia (6)	Carbonea (1)	Eopyrenula (1)
Arthohelium (1)	Catapyrenium (4)	Epilichen (1)
Arthrorhaphis (1)	Catillaria (4)	Evernia (2)
Aspicilia (7)	Catinaria (1)	Farnoldia (1)
Athallia (4)	Cerothallia (1)	Flavoparmelia (2)
Bacidia (8)	Cetraria (4)	Flavoplaca (7)
Bacidina (6)	Cetrelia (4)	Flavopunctelia (1)
Bactrospora (1)	Chaenotheca (10)	Fuscidea (3)
Baeomyces (1)	Chrysothrix (2)	Fuscopannaria (1)
Bagliettoa (6)	Circinaria (6)	Gallowayella (2)
Bellicidia (1)	Cladonia (52)	Glaucumarina (6)
Biatora (1)	Clauzadea (3)	Glypholecia (1)
Biatorella (1)	Cliostomum (2)	Graphis (2)
Biatoridium (1)	Coenogonium (1)	Gyalecta (10)
Bibbya (1)	Collema (1)	Gyalidea (1)
Bilimbia (2)	Coniocarpon (1)	Gyalolechia (1)
Blastenia (3)	Coppinsidea (1)	Gyroglypha (1)
Blennothallia (1)	Cryptothele (1)	Haematomma (1)
Botryolepraria (1)	Dendrographa (1)	Helmutiopsis (2)



Heppia (2)	Mycobilimbia (3)	Porocyphus (2)
Heterodermia (1)	Myriolecis (1)	Porpidia (7)
Heteroplacidium (1)	→Polyozosia	Porpidinia (1)
Hymenelia (1)	Myriospora (1)	Protoblastenia (3)
Hyperphyscia (1)	Naetrocymbe (5)	Protopannaria (1)
Hypocenomyce (1)	Naevia (1)	Protoparmelia (2)
Hypogymnia (3)	Nephroma (2)	Protoparmeliopsis (5)
Hypotrachyna (1)	Nephromopsis (1)	Pseudevernia (1)
Imshaugia (1)	Normandina (1)	Pseudoleptogium (1)
Ionaspis (2)	Ochrolechia (4)	Pseudosagedia (3)
Klauderuiella (2)	Olegblumia (1)	Pseudoschismatomma (1)
Kuettlingeria (4)	Opegrapha (4)	Pseudothelomma (1)
Lambiella (2)	Opeltia (1)	Psilolechia (1)
Lasallia (1)	Oxneria (3)	Psora (3)
Lathagrium (4)	Parabagliettoa (2)	Psoroglaena (1)
Laundonia (1)	Parmelia (5)	Psoronactis (1)
Lecaimmeria (1)	Parmeliella (1)	Psorotichia (3)
(Lecanactis)	Parmelina (4)	Punctelia (3)
→Dendrographa,	Parmeliopsis (1)	Pycnothelia (1)
Psoronactis	Parmotrema (2)	Pyrenodesmia (4)
Lecania (9)	Peccania (1)	Pyrenula (3)
Lecanora (33)	Peltigera (16)	Racodium (1)
Lecidea (10)	Peltula (1)	Ramalina (9)
Lecidella (10)	Pertusaria (10)	Rhizocarpon (18)
Leimonis (1)	Petractis (1)	Rhizoplaca (3)
Lempholemma (2)	Phaeophyscia (8)	Rimularia (1)
Lepra (4)	Phlyctis (2)	Rinodina (25)
Lepraria (13)	Phylliscum (1)	Rinodinella (2)
Leprocaulon (1)	Physcia (12)	Romjularia (1)
Leproplaca (5)	Physciella (2)	Ropalospora (2)
Leptogium (2)	Physconia (3)	Rufoplaca (2)
Leptorhaphis (4)	Piccolia (1)	Rusavskia (2)
Lichenomphalia (1)	Pisutiella (2)	Sagiolechia (1)
Lichinella (1)	Placidiopsis (1)	Sarcogyne (2)
Lobaria (1)	Placidium (5)	Sarcopyrenia (1)
Lobothallia (5)	Placocarpus (1)	Sarcosagium (1)
Maronea (1)	Placopyrenium (5)	Schadonia (1)
Massjukiella (2)	Placynthiella (4)	Schaereria (1)
Megalara (1)	Placynthium (7)	Sclerophora (1)
Megasporea (1)	Platismatia (1)	Scoliciosporum (4)
Melanelixia (5)	Pleurosticta (1)	Scytinium (16)
Melanohalea (3)	Poeltonia (2)	Seawardiella (1)
Menegazzia (1)	Polyblastia (5)	Solenopsora (2)
Micarea (10)	Polychidium (1)	Solitaria (1)
Monerolechia (1)	Polyozosia (8)	Solorina (1)
Montanelia (3)	Polysporina (1)	Sphinctrina (4)
Multiclavula (1)	Porina (1)	Sporastatia (1)

Squamarina (4)
Squamulea (1)
Staurothele (10)
Steinia (1)
Stereocaulon (2)
Straminella (2)
Strangospora (2)
Swinscowia (1)
Synalissa (1)
Tephromela (2)
Tetramelas (1)
Thallinocarpon (1)
Thalloidima (3)
Thelenella (2)
Thelidium (10)
Thelocarpon (2)
Thelopsis (1)
Thrombium (2)
Thyrea (1)
Toninia (8)
Toniniopsis (4)
Trapelia (4)
Trapeliopsis (5)
Umbilicaria (4)
Usnea (10)
Vahliella (1)
Varicellaria (2)
Variospora (2)
Verrucaria (29)
Verrucula (1)
Verruculopsis (1)
Vezdaea (5)
Xanthocarpia (1)
Xanthoparmelia (15)
Xanthoria (2)
Xylographa (1)
Xylopsora (1)
Zeroviella (1)
Zwackhia (1)

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ANTIOXIDANT CAPACITY AND MEMBRANE PERMEABILITY UNDER REWETTING RECOVERY OF *PORELLA PLATYPHYLLA* (L.) PFEIFF., A DESICCATION-TOLERANT LIVERWORT

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Abstract: Relatively constant photosynthetic pigment content (chlorophyll *a*, chlorophyll *b*, and carotenoids) in *Porella platyphylla* (L.) Pfeiff. when experiencing frequent desiccation/rehydration cycles support photosynthetic efficiency, directly influencing its growth and survival and essential for resilience against oxidative stress and its ability to maintain cellular integrity. When actual pigment composition reports healthy physiological condition and functionality the homoiochlorophyllous desiccation tolerance 'machinery' plays a crucial role in the antioxidant defense mechanisms of the liverwort. After a short period of desiccation (1 week), an approximately 200% increase in DPPH inhibition was shown during the first hour of rehydration, indicating a substantial enhancement in antioxidant activity. Folin-Ciocalteu (F-C) assay revealed exceptionally high polyphenol content for *P. platyphylla*, whose values were significantly higher than those typically reported for a wide range of plants. There is a need for further research to explore the specific types of polyphenols present in *P. platyphylla* and their bioactive properties or determine the potentially overestimated polyphenol content by F-C assay. The antioxidant capacity of *P. platyphylla* in mg FeSO₄ equivalent appears to be relatively low. However, it shows a remarkable increase in the antioxidant capacity after 48 hours of rehydration, with an increase of almost 900% compared to the initial 24 hours. The difference in the results from the two antioxidant capacity assays (DPPH, FRAP) for the same sample can be attributed to the specific types of antioxidant molecules present in *P. platyphylla*. GC-MS analysis has revealed the presence of several metabolic compounds, those amounts decreased during rehydration. D-turanose was identified as a substantial component. Membrane permeability after a short period of desiccation in *P. platyphylla* recovered at 90% during the 48 hours of rewetting, while after a medium-long period of desiccation, the recovery was 50%, and only 5-10% after a very prolonged desiccation.

Keywords: *Porella platyphylla*, antioxidant capacity, desiccation tolerance, rehydration, membrane permeability, photosynthetic pigments



INTRODUCTION

Bryophytes represent a very diverse and non-homogeneous group of plants (Shaw *et al.* 2011). Most species tested inhabit shady and moist environments, such as damp trees and rocks, or alongside streams and pools. However, the bryophytes from other ecological groups, and also from open habitats (Wolski *et al.* 2021), are often overlooked. Bryophytes are recognized for their high content of biologically active compounds (Asakawa and Ludwiczuk 2018), extensively used in ethnopharmacology for treating wounds and burns. Specifically, bryophytes exhibit antibacterial, antifungal, antiviral, antioxidant, antiplatelet, antithrombin, insecticidal, neuroprotective properties, and antiproliferative activity against cancer cells (Cheng *et al.* 2012; Vollár *et al.* 2018). These cryptogamic plants are utilized for various purposes across many cultures because of the diverse compounds they contain, such as terpenoids, simple benzoic, cinnamic, and phthalic acid derivatives, coumarins, and nitrogen-containing aromatic compounds like benzonaphthol-xanthenones. The chemical composition of bryophytes can vary significantly depending on their taxonomic group, leading to a wide range of properties. Factors contributing to this diversity include habitat, seasonal changes, water and moisture exposure levels, and materials absorbed from the environment (Heinrichs 2000).

Porella platyphylla, a leafy liverwort (Özenoğlu Kiremit and Keçeli 2009), thrives in shady and moist environments like many bryophytes. A key feature of *Porella platyphylla*, as with many bryophytes, is its desiccation tolerance (DT) the ability to withstand extreme drying and resume normal metabolic activity upon rehydration (Marschall and Proctor 1999). Desiccation tolerance is considered an ancient evolutionary trait, present across all kingdoms of life, from cyanobacteria to angiosperms. Even desiccation-intolerant species may harbor the genes necessary for this trait, though they may not always express them. Desiccation tolerance (DT) is common in poikilohydric bryophytes, but not universal. Among vascular plants (homoiohydric), true desiccation-tolerant (abbreviated also as DT) plants are rare, occurring in only 0.15%. As DT bryophytes dry out, they pass through the water stress levels at which DT vascular plants succeed but only briefly face these water conditions, i.e. they only temporarily face the problem of continuing their metabolism under water stress (Proctor *et al.*

2007). DT bryophyte cells have 2 stable states: one is the full turgor state and the other is the desiccated state. *P. platyphylla*, alternate between active photosynthesis and metabolic dormancy, drying out when water is unavailable and resuming activity upon rehydration. DT bryophyte's tolerance to desiccation is greatly influenced by the relative length of time spent in the two states, the intensity of desiccation, and the associated temperature and light conditions. Vascular DTs do not survive desiccation if it is too rapid (less than 12 h), as 'time-consuming' inductive cell defense mechanisms (e.g. protein synthesis) are required for the development of their desiccation tolerance. DT bryophytes can tolerate dehydration for short periods (up to 1 h, but typically 3 h) and fully recover their metabolism after rewetting. A fundamental difference in their strategy compared to vascular DTs is the existence of an active rehydration-induced repair and regeneration mechanism, which is not based on the transcriptional activity of well-defined tolerance genes, but on a much faster translational control. It is an ABA-independent constitutive defense system that incorporates some key elements of the inducible system characterized in vascular plants. The meaning of this constitutive system is that it pre-expresses protective proteins, pre-forms mRNA-protein complexes, and maintains their levels during desiccation. Furthermore, a constitutive trait is that the amount of osmotically active carbohydrates (mainly sucrose) is significant and their levels are virtually unchanged during desiccation and rewetting.

Upon rehydration, recovery of respiration, photosynthesis, and protein synthesis takes place within minutes or an hour or two; recovery of the cell cycle, food transport, and the cytoskeleton may take 20 hours or more. Sustaining a positive carbon balance is essential for enduring recurrent cycles of desiccation and rewetting. Significant growth requires continuously wet periods of a few days or more. The mechanisms of DT in bryophytes, including expression of LEA proteins, high content of non-reducing sugars, and effective antioxidant and photo-protection, are at least partly constitutive, allowing survival of rapid drying, and employing an active rehydration-induced repair and recovery mechanism (Proctor *et al.* 2007; Marschall 2010).

Studies on *Porella platyphylla* have demonstrated its ability to recover rapidly from desiccation. Modulated chlorophyll fluorometry has been used in research to investigate *P. platyphylla*'s

photosynthetic response to desiccation and subsequent rehydration, providing insights into the plant's physiological systems under stress. According to a study (Marschall and Proctor 1999), *P. platyphylla* quickly regains its ability to photosynthesize after being desiccated for seven days. Whether in low light or complete darkness, the maximal quantum yield (F_v/F_m) and effective quantum yield (Φ_{PSII}) both revert to normal after two hours of rehydration. Relative water content (RWC) decreased dramatically during drying, with both parameters falling below 0.5; however, photochemical quenching was substantially unaffected. Under stress, non-photochemical quenching (*NPQ*) rose and peaked after recovery in light. However, within 24 hours, *NPQ* reverted to normal levels, suggesting that photosynthetic performance was temporarily impacted.

Its carbohydrate metabolism is another crucial factor contributing to *P. platyphylla*'s desiccation tolerance. Starch and reducing sugars are present at relatively low concentrations in *P. platyphylla*. Sucrose and fructan are the main soluble carbohydrates. Sucrose and fructans have shapes suitable for association with the polar head groups of phospholipids in place of water and for preventing damaging phase transitions in membranes during desiccation. Additionally, these sugars maintain a vitreous phase in the cytoplasm of desiccated cells, which minimizes protein denaturation. Fructans can be inserted between the head groups of different kinds of phospholipids with some preference for phosphatidylethanolamine. They are the key regulators of adaptation to various environmental stresses, act as antioxidants, scavenging ROS, and prevent cell damage under abiotic stress conditions. Fructan-accumulating species contain only traces (~1%) of starch, which means that fructan is a real alternative to starch. Fructans accumulate in the vacuole, where they play an important role in turgor regulation. More molecules mean these cells are more resistant to osmotic pressure or cold. The size of fructan polymers can be altered quickly; this could be an explanation for their role in osmotic adjustment. Fructans likely protect plants from various environmental stresses such as frost and drought by stabilizing membranes. Starch synthesis drops dramatically when the temperature decreases below 10°C, but photosynthesis and fructan production are much less sensitive to low temperatures, suggesting that fructan production benefits those plants, which actively

photosynthesize during the winter and early spring. The soluble carbohydrate pool is well-balanced in *P. platyphylla* (sugar feeding, dark starvation, desiccation, and low temperature have little effect), and fructans are conserved at the expense of a substantial sucrose stock. In higher plants, a negative correlation between sucrose concentration and soluble acid invertase activity was observed. The high sucrose concentration coupled with high acid invertase activity in *Porella platyphylla*. Dark starvation significantly increased the activity of acid invertase, i.e. acid invertase is active in the dark. Glucose and fructose treatment in the dark significantly decreased the activity of the enzyme. When applied in the light, it increased, a role associated with fructan accumulation, consistent with an increase in the concentration of low molecular weight carbohydrates. Desiccation reduced the glucose, fructose, and sucrose content of the leaves. One hour after rehydration, glucose, and fructose contents were 5 times higher than before dehydration. Sucrose and fructan contents were at pre-desiccation levels 1 hour after rehydration. During desiccation, the amount of low molecular weight fructans decreased while that of high molecular weight fructans increased, suggesting that fructan polymerization occurred during desiccation. The concentration of fructans increased under low water potential, doubling at $\Psi = -0.62$ MPa, while specific acid invertase showed only a small increase at $\Psi = -4.5$ MPa. According to this regulation, invertase is crucial for maintaining a balance between energy consumption and sucrose preservation throughout the plant's desiccation recovery. These adaptation strategies highlight *P. platyphylla*'s resilience to severe dehydration (Marschall *et al.* 1998).

In addition to its carbohydrate metabolism, *P. platyphylla* produces a wide range of secondary metabolites, including sesquiterpenoids and diterpenoids. Along with three known pinguianes and the sacculatane perrottetianal B, studies have identified a new sesquiterpenoid derivative of pinguisanoic acid, methyl 2 α -hydroxy-6-oxo-11-pinguisanoate, and a new hemiacetal sacculatane diterpenoid, (13S)-15 ξ -hydroxysacculaporellin, from the liverwort. These findings further highlight the chemical diversity of *P. platyphylla* and contribute to our understanding of its secondary metabolism (Buchanan *et al.* 1996).

While the desiccation tolerance of *P. platyphylla* has been well studied (Marschall *et al.* 1995; Marschall 1998; Marschall *et al.* 1998; Marschall and Proctor 1999; Marschall 2010; Marschall and Sütő 2022) but its antioxidant capacity remains relatively unexplored compared to other bryophytes. In a recent study (Aydin 2020), the objective was to determine the free radical scavenging activities, fatty acid, and vitamin contents of *Dicranum scoparium* and *Porella platyphylla*. *Dicranum scoparium* exhibited a significantly higher DPPH radical scavenging effect compared to *P. platyphylla*. A strong correlation exists between the phenolic compound content in methanol extracts of the plants and their DPPH radical scavenging efficiency. *Dicranum scoparium* had higher amounts of D-3 α -tocopherol, stigmasterol, and betasterol. *P. platyphylla* contained higher levels of all unsaturated fatty acids except for α -linolenic acid when compared to *Dicranum scoparium* (Aydin 2020). Although, *P. platyphylla* showed lower DPPH scavenging activity in this study, its high unsaturated fatty acid content indicates it could still be a potent antioxidant.

Additionally, *P. platyphylla* has demonstrated significant antioxidant capacity (AC) and high total phenolic compound (TPC) content, particularly in the n-butanol fraction. A prominent phenolic acid, p-hydroxybenzoic acid (p-HBA), is found in free form and likely contributes to the strong antioxidant activity observed in the species (Yildirim Akatin *et al.* 2024). These findings suggest that *P. platyphylla* could serve as a valuable source of bioactive compounds with potential applications in biotechnology and ecological adaptation.

In light of its demonstrated desiccation tolerance and its potential for antioxidant activity, this study aims to further investigate the antioxidant properties, and membrane integrity of *P. platyphylla*, focusing on its unique chemical composition, and its capacity to withstand desiccation and recover quickly upon rehydration. Understanding how desiccation tolerance and antioxidant capacity are interrelated could provide valuable insights into the plant's resilience mechanisms and its potential biotechnological applications.

MATERIALS AND METHODS

Plant material

Porella platyphylla was collected from an area of limestone woodland in the Bükk Mountains, north-east Hungary, near Felsőtárkány village in the late spring season. The field history of the collected material was unknown; therefore, field-collected plants were allowed a state of uninterrupted hydration under nonstressful conditions, to lose any physiological hardening to DT that has been gained by experiencing, under field conditions, wetting and drying events (Marschall and Sütő 2022). The plants were acclimatized at 100% relative humidity (r.h.) in glass desiccators, at 20°C, at a photosynthetic photon flux density (PPFD) of 100 $\mu\text{mol m}^{-2} \text{s}^{-1}$, and a photoperiod of 12/12 h light/dark for 3 days before desiccation treatment. So-called 'natural drying' meant drying in laboratory air (at 20°C, ~35% RH, in natural light, infiltrating through the laboratory windows) was applied for 1 week. When testing membrane permeability during rewetting recovery three periods of desiccation were applied before rehydration: a short period (1 week), a medium-long period (1 month), and a very prolonged period (6 months) of desiccation. The air-dried weight of only the green parts of the liverwort samples was measured with analytical accuracy (0.02 g). Samples with known air-dried weight are used for extraction or set up for rehydration. Rewetting was carried out with distilled water. Rehydrated samples were monitored for various physiological parameters after 1, 24, and 48 hours, and kept in a desiccator with 100% RH between measurements, in natural light (outdoor, natural light infiltrating through the laboratory window) (n=3).

Determination of Pigment Content

Pigment analyses followed Lichtenthaler and Wellburn (1983). The air-dry bryophyte samples were extracted in 96% (v/v) ethanol and absorbance at 470, 649, and 665 nm read on a spectrophotometer (Varian Cary 3E).

Determination of Antioxidant Capacity – DPPH Test

The DPPH (1,1-diphenyl-2-picrylhydrazyl) assay is a well-established method for evaluating the antioxidant activity in plant extracts, leveraging the ability of antioxidants to neutralize DPPH radicals. DPPH, a stable free radical with a distinct purple color, undergoes a color change from purple to yellow upon reduction, a shift that can be quantified spectrophotometrically ($\lambda = 517 \text{ nm}$) (Blois 1958; Frankel and Meyer 2000). DPPH was added to the different volumes (100, 200, 300, 400, and 500 μL) of 96% (v/v) ethanolic extracts of *P. platyphylla* to test inhibition.

Determination of Antioxidant Capacity – Folin-Ciocalteu Assay

The determination of phenol content was based on the reduction of phenolic compounds by the Folin-Ciocalteu reagent under alkaline conditions resulting in a blue-colored complex whose intensity is proportional to the total phenol content in the sample (Pérez *et al.* 2023). Ethanolic extracts (96% (v/v)) of *P. platyphylla* were used in the Folin-Ciocalteu assay. Absorbance was measured at 765 nm. The polyphenol content of the samples was calculated in terms of gallic acid equivalents.

Determination of Antioxidant Capacity – FRAP Assay

The ferric-reducing antioxidant power (FRAP) assay is based on the reducing ability of antioxidants, which involves the reduction in Fe^{3+} -2,4,6-tripyridyl-s-triazine (TPTZ) complex while taking absorbance at 593 nm (Benzie and Strain 1996). Ethanolic extracts (96% (v/v)) of *P. platyphylla* were used in the FRAP assay.

Analytical Measurements – GC-MS Analysis

For the GC-MS measurement (Perkin Elmer Autosystem XL GC and Turbomass MS), the 96% (v/v) ethanolic extracts of *P. platyphylla* underwent a derivatization procedure as follows: the extracts were first dried under a stream of N_2 (5.0) gas at room temperature. The dried samples were then redissolved in 50 μL of pyridine to prepare them for derivatization. Following this, 50 μL of BSTFA + TMCS (N, O-Bis(trimethylsilyl)trifluoroacetamide with 1% Trimethylchloro-

silane) was added, and the mixture was heated at 70°C for 1 hour to complete the derivatization reaction. After cooling, 1 µL of the derivatized samples was injected into the GC-MS for analysis, enabling precise measurement of the target compounds.

The Perkin Elmer Autosystem XL GC was equipped with a Turbomass MS series mass spectrometer. The GC-MS was installed with an HP-5 ms capillary column ((5%-Phenyl)-methylpolysiloxane phase, 30 m × 0.25 mm × 0.5 µm) (Agilent). Helium 6.0 was the carrier gas with a constant flow rate of 1.0 ml min⁻¹. The injector temperature was 270°C. Injected volume: 1 µl. Chromatographic conditions were as follows: the oven was held for 2 min at 70°C, and heated to 150°C at 25°C min⁻¹, heated to 200°C at 6°C min⁻¹, heated to 280°C at 10°C min⁻¹ and held 5.2 min at this temperature. The mass spectrometer operated in the EI+ mode with a source temperature of 200°C, an ionizing voltage of 70 eV, and a transfer line temperature of 270°C. The mass spectrometer scanned masses from 30 to 650 m/z at a rate of 0.1 scan s⁻¹ the detection time windows: 3 to 25.27 min. Peak identification was carried out by comparison of the derivatization sample mass spectra with spectra in the NIST/EPA/NIH Mass Spectral Database.

Measurement of ion leakage / Membrane permeability

A modified Szalai's method (Marschall and Sütő 2022) was used to determine the changes in membrane permeability. 0.1 g (fresh weight) of plant material was put in centrifuge tubes containing 20 ml ultrapure distilled water (Milli-Q 50). The samples were shaken for an hour and the electrolytes leaked from the plant cells in the water were measured using an electrical conductivity meter (C1) (Hach, HQ40D conductometer). Then the samples were boiled (at 100 °C) in the same tube containing the distilled water to complete membrane disruption. After cooling down to room temperature samples were shaken for an hour again, and then the membrane permeability of the samples was measured by a conductivity meter as well (C2). The final conductivity was calculated as percent ion leakage, (C1 / C2) x 100.

Statistical analysis

Statistical analysis was performed with the SigmaPlot 11.0 software (Systat Software Inc., San Jose, CA, U.S.A.) using one-way analysis of variance. The presence or absence of significant differences between each treatment was detected by the Holm-Sidak test. Where probabilities are quoted the data have been subject to analysis of variance. Errors, where indicated, are standard deviations. Measurements were performed in three separate replicates ($n=3$) and also 3 replicates were treated in each condition. Replicates for analysis were from three separate samples; they were not homogenized. Replicates represent a single branch. There were not necessary any transformations during the statistical analysis.

RESULTS AND DISCUSSION

Photosynthetic pigment composition

Studying photosynthetic pigments, including chlorophyll *a*, chlorophyll *b*, and carotenoids, in *Porella platyphylla* is essential for understanding its physiological condition and functionality. These pigments are integral to the liverwort's photosynthetic efficiency, directly influencing its growth and survival. In terms of antioxidant activity, chlorophylls and carotenoids can indicate the plant's capacity to mitigate oxidative stress. Both pigment types are recognized for their antioxidant properties, which protect plant cells from oxidative damage caused by reactive oxygen species (ROS). Therefore, quantifying the photosynthetic pigment content can provide valuable insights into the plant's resilience against oxidative stress and its ability to maintain cellular integrity. This assessment aligns with our DPPH inhibition findings, suggesting a link between pigment concentration and antioxidant activity. Based on preserving photosynthetic pigments during desiccation *P. platyphylla* belongs to the homoiochlorophyllous type. Our study's pigment results align with those reported by Marschall and Proctor (2004), confirming that *P. platyphylla* exhibits typical pigment concentrations associated with healthy plant samples (*Figure 1*). This accordance with the literature data not only confirms our results but also supports the hypothesis that photosynthetic pigments and the homoio-chlorophyllous desiccation tolerance 'machinery' play a crucial role in the antioxidant defense mechanisms of *P. platyphylla*.

Bryophytes can achieve photoprotection during dehydration through non-photochemical quenching processes in which xanthophylls harmlessly dissipate the excess absorbed energy as heat (Deltoro *et al.* 1998; Csintalan *et al.* 1999). Such protection seems particularly beneficial during the rehydration phase when the gametophytes experience considerable photooxidative damage.

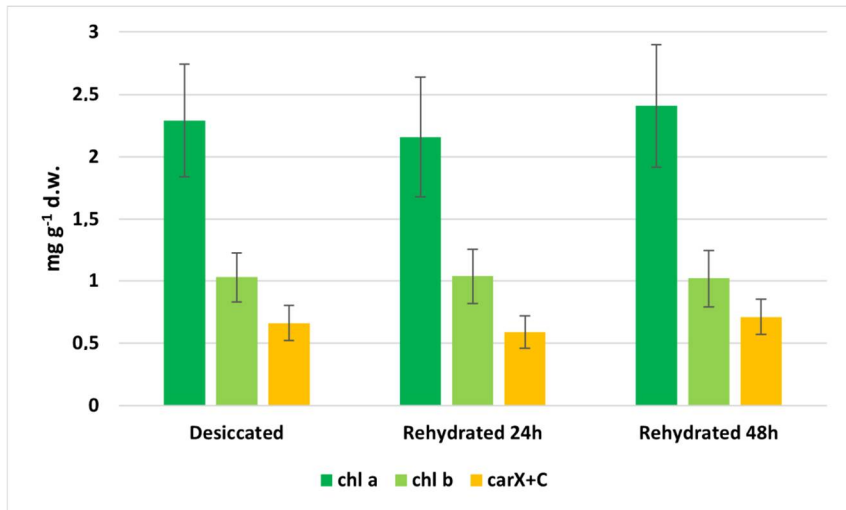


Figure 1. Photosynthetic pigment content (chlorophyll *a*, *b*, and total carotenoids in mg g⁻¹ d.w.) of *Porella platyphylla* after a short period of desiccation, 24 hours and 48 hours following rehydration. Error bars are STDs, where n=3.

Antioxidant capacity

The rehydration of *P. platyphylla* resulted in an approximate 200% increase in **DPPH inhibition**, indicating a substantial enhancement in antioxidant activity over time. Initial DPPH inhibition values demonstrated significant increases upon rehydration, with the most pronounced effect observed after 1 hour following rehydration (*Figure 2*). These results suggest that rehydration may activate or amplify the liverwort's inherent antioxidant mechanisms. This increase in DPPH inhibition could be attributed to multiple factors. Rehydration may activate specific metabolic pathways that promote the synthesis of antioxidant compounds, or it may facilitate the release of antioxidants that were previously bound within the cellular structure, making them more available for free radical

scavenging. The levels of oxidative damage increase directly upon rehydration, especially in light conditions. Since electrons keep reaching the photosystems but photosynthesis is not fully recovered, ROS are highly produced upon rehydration (Minibayeva and Beckett 2001; Beckett *et al.* 2004), increasing damage during the initial phase, especially at the chloroplast level. Additionally, this ROS burst can also act, to a certain level, as a defense against pathogenic fungi and bacteria that can attack cells upon rehydration (Minibayeva and Beckett 2001). Detailed metabolic profiling and molecular studies are necessary to elucidate the precise mechanisms driving this marked increase in antioxidant activity. Additionally, environmental factors during rehydration, such as temperature and light exposure, deserve attention, as they may influence the synthesis and release of antioxidant compounds. The greatest value recorded for *P. platyphylla* in the literature for the DPPH test was 62.39% (Aydin 2020), and the results here (Figure 2) show that during the first hour of rehydration, it can go higher than that. Comparative studies with other bryophyte species could further illuminate the unique properties of *P. platyphylla* that contribute to its high antioxidant capacity when rehydrated.

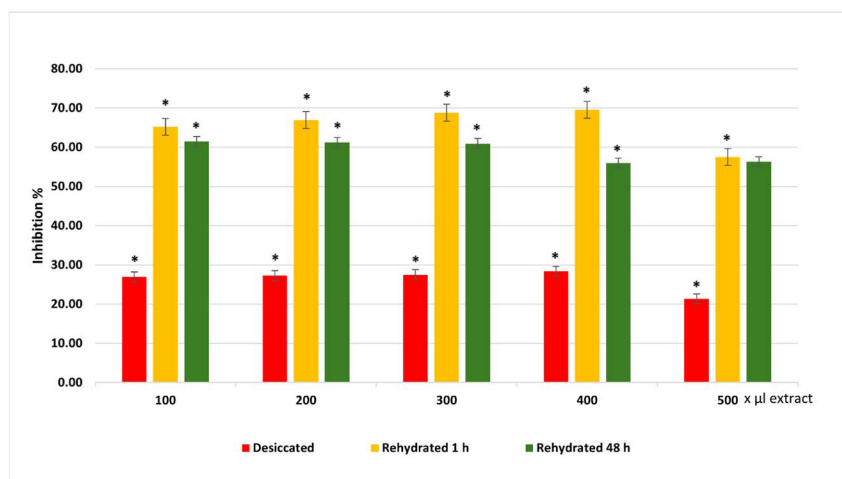


Figure 2. The antioxidant capacity of *Porella platyphylla*, as inhibition %, was determined by the DPPH test after a short period of desiccation, 24 hours and 48 hours following rehydration. DPPH was added to the different volumes (100, 200, 300, 400, and 500 µl) of 96% (v/v) ethanolic extracts of *P. platyphylla* to test inhibition. * = Statistically significant ($p < 0.05$). Error bars are STDs, where $n = 3$.

Our pilot **Folin-Ciocalteu assay** results for *P. platyphylla* reveal exceptionally high polyphenol content, with values of 44960 mg, 31700 mg, and 45130 mg of gallic acid equivalents (GAE) per gram of dry matter. These values are significantly higher than those typically reported for a wide range of plants. For context, many fruits and vegetables exhibit polyphenol content in the range of 10-100 mg GAE/g dry weight (Basu and Maier 2016), while even plants known for their high polyphenol content, such as certain berries or tea leaves, rarely exceed 300-400 mg GAE/g dry weight (Ankolekar *et al.* 2011). However, it is important to consider the Folin-Ciocalteu test is not specifically designed for phenolic compounds, as the reagent could be reduced by other nonphenolic compounds also present in the sample, with the risk of content overestimation. Our results support the need for further research to explore the specific types of polyphenols present in *P. platyphylla* and their bioactive properties. Additionally, comparative studies with other high-polyphenol plants could provide further insights into the unique attributes of *P. platyphylla* that contribute to its elevated polyphenol levels.

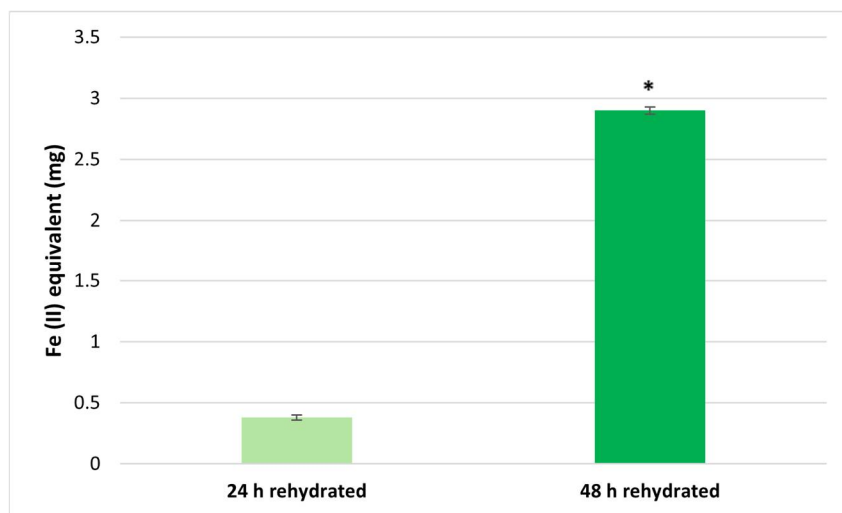


Figure 3. The antioxidant capacity of *Porella platyphylla*, as the ferric-reducing antioxidant power (FRAP) is based on the reducing ability of antioxidants, which involves the reduction in Fe^{3+} -2,4,6-tripyridyl-s-triazine (TPTZ) complex after a short period of desiccation, 24 hours and 48 hours following rehydration. Ethanolic extracts (96% (v/v)) of *P. platyphylla* were used in the FRAP assay. * = Statistically significant ($p < 0.05$). Error bars are STDs, where $n = 3$.

Our **FRAP assay** findings (*Figure 3*) indicate a remarkable increase in the antioxidant capacity of *P. platyphylla* after 48 hours of rehydration, with an increase of almost 900% compared to the initial 24 hours. When compared to the literature values for mosses such as *Ceratodon purpureus* and *Tortula muralis* (Wolski *et al.* 2021), where antioxidant activities range between 60-120 mg FeSO₄ equivalent, the antioxidant capacity of *P. platyphylla* appears to be relatively low. However, this significant enhancement in antioxidant activity during 48 hours of rehydration has not been previously observed in the literature, marking a novel discovery in the study of this liverwort.

The difference in the results from these two assays (DPPH, FRAP) for the same sample can be attributed to the specific types of antioxidant molecules present in *P. platyphylla*. Some antioxidants may have stronger reducing power, thus showing higher activity in the FRAP assay, while others may be more effective in scavenging free radicals, showing higher activity in the DPPH assay (*Figure 2, 3*).

Result of the GC-MS analysis

GC-MS analysis has revealed the presence of several metabolic compounds, those amounts decreased during rehydration (*Figure 4*). The most prominent peak identified was D-turanose, a compound typically found in bryophytes. The identification of D-turanose in *P. platyphylla* is noteworthy as it provides insights into the metabolic profile of the liverwort. This sugar, along with other detected compounds, could play a role in the liverwort's response to desiccation and the subsequent rehydration and its recovery capacity. The detailed analysis of these metabolites can further our understanding of the biochemical pathways active in *P. platyphylla*, potentially linking specific metabolites to enhanced antioxidant activity observed after rehydration.

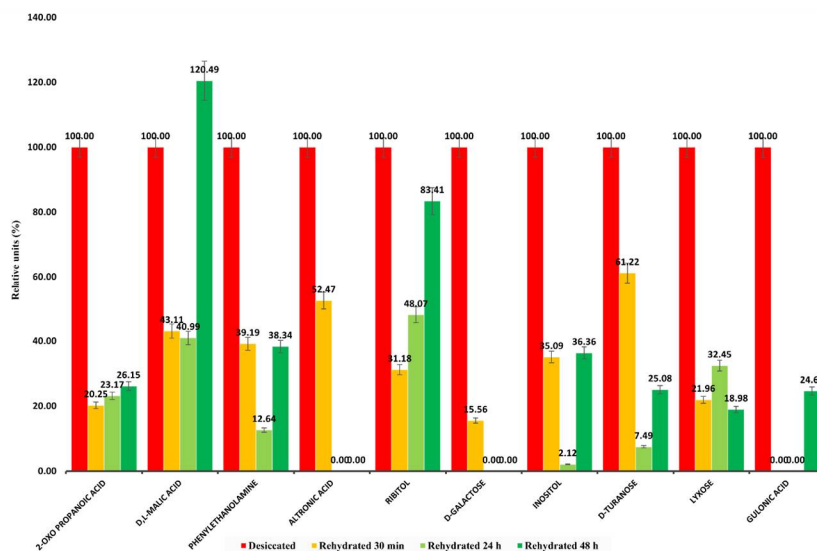


Figure 4. Metabolic components of *Porella platyphylla*, determined by GC-MS after a short period of desiccation, 30 minutes, 24 hours, and 48 hours following rehydration. Ethanol extracts (96% (v/v)) of *P. platyphylla* underwent a derivatization procedure as written in Materials and Methods. Each component is potted in the graph in relative units (%). Error bars are STDs, where n=3.

Membrane permeability measurements

Desiccation disrupts cellular membranes in bryophytes due to mechanical stress, oxidative damage, and loss of structural integrity. This results in the leakage of ions such as K^+ , and Ca^{2+} from the cell interior, and other solutes upon rehydration. Electrolyte leakage, measured as ion conductivity or solute release into rehydration water, is a critical physiological parameter used to assess cell membrane integrity and damage during these stress events. Studies report significant electrolyte leakage immediately after rehydration, indicative of membrane damage. However, many bryophytes, especially desiccation-tolerant species, show rapid repair of membranes, minimizing long-term leakage. Desiccation-tolerant bryophytes, such as *Syntrichia ruralis* and *Syntrichia caninervis*, exhibit lower initial leakage and faster recovery compared to desiccation-sensitive species. Electrolyte leakage peaks during the first few minutes to hours post-rehydration and stabilizes over time as membranes repair. Cells recover their form very rapidly after

rehydration (from 30 seconds to one minute) (Glime 2017). Desiccation-tolerant bryophytes employ protective mechanisms, including the accumulation of late embryogenesis abundant (LEA) proteins, sugars (e.g., trehalose), and antioxidant systems, which mitigate damage and leakage (Glime 2017). In the more DT species, this leakage is transient, probably due to lipid-phase transitions occurring in the plasma membrane (Crowe *et al.* 1992). In the more sensitive species, this leakage is more substantial (Crowe *et al.* 1992), eventually leading to complete loss of all intracellular content and cell death. Membrane permeability after a short period of desiccation in *P. platyphylla* recovered at 90% during the 48 hours of rewetting (Figure 5A). While after a medium-long period of desiccation, the recovery was 50%, and only 5-10% after a very prolonged desiccation (Figure 5B, C). Oliver *et al.* (1993) found that electrolyte leakage alone was not a reliable measure of desiccation tolerance in *Syntrichia ruralis*. Factors such as the rate of desiccation, exposure to light during drying, and temperature influence the extent of membrane damage and leakage. Repeated cycles of desiccation and rehydration may lead to cumulative damage, increasing electrolyte leakage in sensitive species. Bryophytes from arid environments show lower electrolyte leakage compared to species from mesic habitats, underscoring their evolutionary adaptations. Electrolyte leakage can be a more reliable measure of the effect of desiccation if the field physiological history of the collected bryophytes is known. Electrolyte leakage studies provide insights into the resilience and stress physiology of bryophytes. These findings are pivotal in understanding desiccation tolerance mechanisms, which have implications for ecosystem resilience and biotechnological applications. Further research should focus on the molecular mechanisms underpinning membrane repair in desiccation-tolerant bryophytes, and the long-term effects of repeated desiccation/rehydration cycles on membrane integrity. Comparative studies should provide data across broader bryophyte taxa to identify universal and species-specific tolerance strategies.

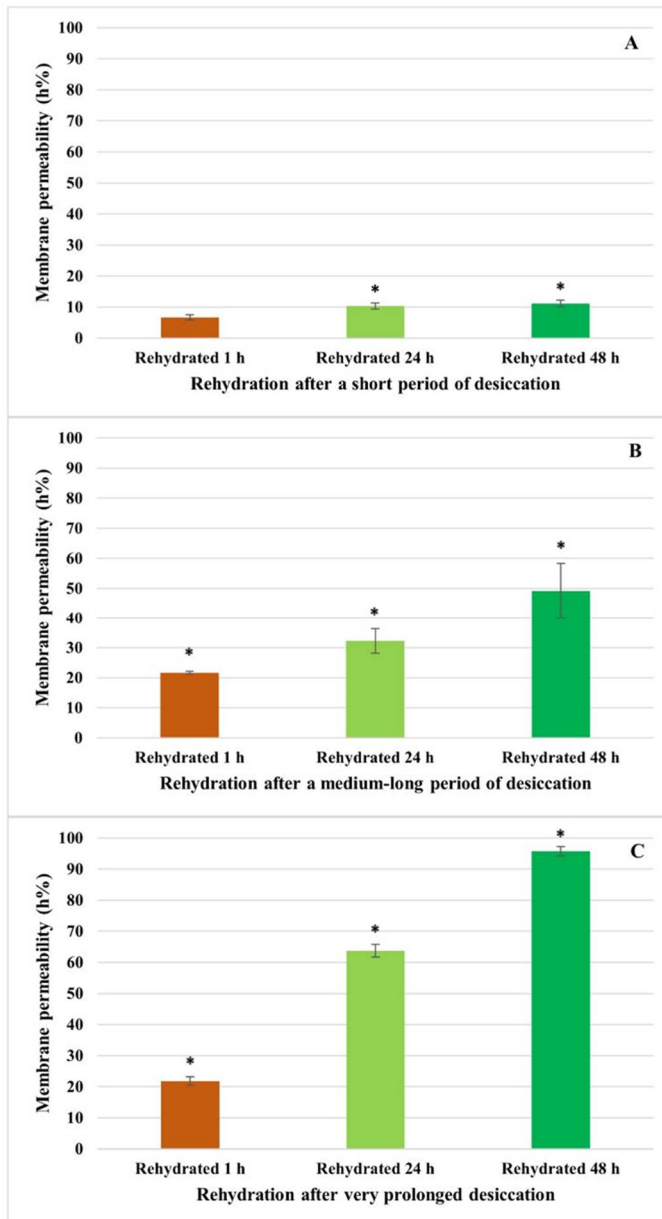


Figure 5. Membrane permeability during rewetting recovery in *Porella platyphylla*. Ion leakage (%) was measured in *P. platyphylla* **A)** after a short period (1 week) of desiccation and after 1, 24, and 48 hours of subsequent rehydration; **B)** after a medium-long period (1 month) of desiccation and after 1, 24 and 48 hours of subsequent rehydration; **C)** after a very prolonged period (6 months) of desiccation and after 1, 24 and 48 hours of subsequent rehydration. * = Statistically significant ($p < 0.05$). Error bars are STDs, where $n = 3$.

CONCLUSIONS

Porella platyphylla a desiccation-tolerant liverwort can achieve photoprotection during the frequent desiccation/ rehydration cycles through non-photochemical quenching processes in which xanthophylls harmlessly dissipate the excess absorbed energy as heat and with a help of effective antioxidant capacity especially under the rehydration phase when the gametophytes exposed to considerable photooxidative stress. *P. platyphylla* has a significant antioxidant capacity and a high content of total phenolic compounds. Further studies should focus on quantifying these metabolites and exploring their biological roles, which may reveal new aspects of how rehydration enhances the liverwort's antioxidant properties. To further understand the mechanisms behind this significant increase in antioxidant capacity, additional studies are needed. Conducting detailed metabolic profiling before and after rehydration could identify specific compounds responsible for the increased antioxidant activity. Extending the rehydration period beyond 48 hours and monitoring antioxidant capacity at various intervals could provide a clearer picture of the dynamics of antioxidant synthesis and release. Investigating the impact of other environmental stress factors, such as light, temperature, and nutrient availability, in combination with rehydration could offer insights into optimal conditions for maximizing antioxidant production. Exploring the genetic and molecular basis of the observed increase in antioxidant activity could help identify key genes and regulatory mechanisms involved in this process. Further research is needed to understand in more detail the rehydration process and to fully characterize the liverwort's antioxidant potential.

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BRYOFLORESTICAL DATA FROM THE TĂTARULUI GORGE (IGNIȘ MOUNTAINS, ROMANIAN EASTERN CARPATHIAN, TRANSYLVANIA)

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Abstract: The main aim of this study was to explore the bryophyte diversity and distribution patterns in the Igniș Mountains. From our collections hitherto 54 bryophyte species were identified. Among them the endangered (EN) *Grimmia trichophylla*, vulnerable (VU) *Plagiomnium ellipticum*, *Dicranella rufescens* the near threatened (NT) *Andreaea alpestris*, *Plagiomnium elatum* and the not evaluated (NE) *Racomitrium elongatum* are worth to be mentioned.

Keywords: bryoflora, vulnerable species, Igniș Mountains, Romania

INTRODUCTION

In spite of other previous investigations, like those of Boros and Vajda (1967), and Codoreanu (1972) and Béres (2011) who compiled the list of bryophyte collection made by A. Coman and A. Boros we only have some limited information on this bryologically neglected area. The following 30 species were known from Tătarului gorge, based on these publications, are the following: *Calypogeia neesiana*, *Lepidozia reptans*, *Marsupella emarginata*, *Metzgeria conjugata*, *Porella arboris-vitae*, *Riccardia palmata*, *Scapania nemorea*, *Abietinella abietina*, *Andreaea alpestris*, *Dicranella rufescens*, *Dicranum montanum*, *Hylocomium splendens*, *Paraleucobryum longifolium*, *Plagiomnium affine*, *P. rostratum*, *Plagiothecium undulatum*, *Pleurozium schreberi*, *Polytrichastrum formosum*, *Polytrichum juniperinum*, *P. strictum*, *Pseudanomodon attenuatus*, *Pseudoleskeella nervosa*, *Pseudotaxiphyllum elegans*, *Pteryginandrum filiforme*, *Ptilium crista-castrensis*, *Racomitrium heterostichum* var. *gracilescens*, *Sanionia uncinata*, *Schistidium rivulare*, *Schistostega pennata* and *Sphagnum capillifolium*.



The investigated area

The Gutâi Mountains are a mountain range within the Vihorlat – Gutâi area of the inner Eastern Carpathians. Igniş and Gutâi mountains are situated at the western and southern limit of Maramureş Land, they are the oldest sector of the volcanic range in Eastern Carpathians. There are two units distinguished by geofoms originating from different types of volcanic activity. They are separated by mountain passes from the neighbouring units (Huta 587 m, Gutâi 984 m, Neteda 1039 m): Igniş mountains is an andesitic plateau, mostly stratified, with small depressions, cliffs and residual forms, named 'rocks' (Piatra Săpânței, Piatra Goală, Piatra Rea, etc.). The climate of the SE Carpathians is colder and more continental than that of the NW Carpathians (Hajdú-Moharos 1996).

Cheile Tătarului is a nature reserve in the Gutâi Mountains, located on the territory of Mara village, Desești commune, Maramureş county. This tourist objective has been included in category III of the National Union for Nature Conservation. The conservation area covers an 15 hectares and the gorges themselves are 700 metres long. They are formed by the Igniş-Mara, in pyroxene andesite bedrock. Their two slopes are different, one is covered by forests and the other is a vertical cliff. The natural reserve Cheile Tătarului is declared a protected area by Law no.5/6 March 2000 (Lege 2000). Runcu stream flows through the gorge. A reservoir with a dam is under construction since 1987 on the same stream. The finalization of this dam will lead to the covering of the area of the gorges.

MATERIAL AND METHODS

In May 2024 we organised a collecting trip to record bryophytes from different substrates growing on soil, rocks, decaying wood, trunks and tree branches. The collected material was compared with the records published by Boros and Vajda (1967), Rațiu and Moldovan (1972) and Béres (2011). The collection was made in various vegetation types: meadows in front of the gorge entrance and the spruce forest which is present along the gorge (*Figure 1*). Igniş Mountain is covered otherwise by beech forests, except for

some spruce inclusions on the northern slopes, enclaves preserved in areas with a cold and humid microclimate. The spontaneous existence of spruce forests is evidenced by studies in the Tătarului Gorge (Moldovan 1970; Rațiu and Moldovan 1974).

The Romanian distribution of mosses was established from Plămadă (1998) and Mohan (1998), while that of the liverworts from Ștefănuț (2008). The nomenclature follows Hodgetts *et al.* (2020). Species names are followed by the collecting site number, and by the substrate on which they were grown. The collected specimens are deposited in the Herbarium of Eger (EGR).



Figure 1. Entrance at Tătarului Gorge (photo: Andrea Sass-Gyarmati).

RESULTS

List of species

During the field study 54 bryophyte species were found in the investigated area, of which 7 belong to Marchantiophyta and 47 to Bryophyta species.

Marchantiophyta

- Apopellia endiviifolia* (Dicks.) Nebel & D.Quandt – 2401/BB, on irrigated rocks
Calypogeia neesiana (C.Massal. & Carestia) Müll.Frib. – 2401/L, on soil
Lophozia longidens (Lindb.) Konstant. & Vilnet – 2401/BG, on rocks
Marchantia polymorpha L. – 2401/BF, on irrigated soil
Ptilidium pulcherrimum (Weber) Vain. – 2401/I, on decaying wood
Riccardia palmata (Hedw.) Carruth. – 2401/Y, on decaying wood
Scapania nemorea (L.) Grolle – 2401/BC, on irrigated rocks

Bryophyta

- Abietinella abietina* (Hedw.) M.Fleisch. – 2401/M, on rocks
Andreaea alpestris (Thed.) Schimp. – 2401/AZ, on rocks
Atrichum undulatum (Hedw.) P.Beauv. – 2401/AD, on soil
Brachythecium glareosum (Bruch ex Spruce) Schimp. – 2401/AZ, on grassland
Bryum argenteum Hedw. – 2401/BE, on rocks
Calliergon cordifolium (Hedw.) Kindb. – 2401/U, on irrigated rocks
Calliergonella cuspidata (Hedw.) Loeske – 2401/AS, on marshy meadow
Climacium dendroides (Hedw.) F.Weber & D.Mohr – 2401/H, on soil
Dicranella rufescens (Dicks.) Schimp. – 2401/BI, on soil
Dicranum scoparium Hedw. – 2401/A, on rocks
Dicranum montanum Hedw. – 2401/AA, on decaying wood
Fontinalis antipyretica Hedw. ssp. *antipyretica* – 2401/S, on irrigated rocks
Grimmia trichophylla Grev. – 2401/BH, on rocks
Hylocomium splendens (Hedw.) Schimp. – 2401/AM, on rocks
Hymenostylium recurvirostrum (Hedw.) Dixon – 2401/V, on rocks
Hypnum cupressiforme Hedw. – 2401/N, on tree buttress
Isopterygiopsis elegans (Brid.) Lindb. – 2401/AT, on decaying wood
Lewinskia speciosa (Nees) F.Lara, Garilleti & Goffinet – 2401/AU, on bark
Oxyrrhynchium hians (Hedw.) Loeske – 2401/AL, on soil
Paraleucobryum longifolium – 2401/AV, on rocks

- Plagiomnium affine*** (Blandow ex Funck) T.J.Kop. – 2401/D, X, on soil
Plagiomnium elatum (Bruch & Schimp.) T.J.Kop. – 2401/AK, on earth covered rocks
Plagiomnium ellipticum (Brid.) T.J.Kop. – 2401/AR, on decaying wood
Plagiomnium undulatum (Hedw.) T.J.Kop. – 2401/E, on *Picea* roots
Plagiothecium curvifolium Schlieph. ex Limpr. – 2401/AH, on tree roots
Pleurozium schreberi (Willd. ex Brid.) Mitt. – 2401/K, on soil
Pogonatum urnigerum (Hedw.) P.Beauv. – 2401/AB, on soil
Polytrichastrum formosum (Hedw.) G. L. Sm. – 2401/AW, on soil
Polytrichum juniperinum Hedw. – 2401/BK, on rocks
Polytrichum piliferum (Sw.) Schimp. – 2401/AC, on soil
Polytrichum strictum Menzies ex Brid. – 2401/BL, on soil
Polytrichum commune Hedw. – 2401/AQ, on soil
Pseudotaxiphyllum elegans (Brid.) Z.Iwats. – 2401/O, on earth covered rocks
Ptychostomum moravicum (Podp.) Ros & Mazimpaka – 2401/AO, on rocks
Ptychostomum pseudotriquetrum (Hedw.) J.R.Spence & H.P. Ramsay ex Holyoak & N. Pedersen – 2401/G, marshy bog on soil
Racomitrium aciculare (Hedw.) Brid. – 2401/T, on irrigated rocks
Racomitrium canescens (Hedw.) Brid. – 2401/BD, on stone bridge over the river
Racomitrium elongatum Ehrh. ex Frisvoll – 2401/P, on rocks
Rhizomnium puntatum (Hedw.) T.J.Kop. – 2401/AF, on rocks
Rhytidiadelphus squarrosus (Hedw.) Warnst. – 2401/F; 2401/AG, on soil
Sanionia uncinata (Hedw.) Loeske – 2401/AJ, on bog
Schistidium apocarpum (Hedw.) Bruch & Schimp. – 2401/AN, on rocks
Schistidium rivulare (Brid.) Podp. – 2401/R, on irrigated rocks
Schistostega pennata (Hedw.) F.Weber & D.Mohr – 2401/B, on *Picea* roots
Sphagnum capillifolium (Ehrh.) Hedw. – 2401/C, on bog; 2401/AP, on soil
Sphagnum girgensohnii Russow – 2401/BA, on marshy meadow
Tetraphis pellucida Hedw. – 2401/Z, on decaying wood

DISCUSSION

The results of this study contributes to the knowledge of the biodiversity of Igriş Mountains. The main reason for relatively high biodiversity is the variety of habitat types that can be found in this area.

Andreaea alpestris (Thed.) Schimp. – subarctic-subalpine element, NT in Romanian bryoflora (Ştefănuţ and Goia 2012). The sporadic occurrence of this species is explained by the fact that it is usually noted in the field as *A. rupestris*, without microscopic analysis.

Dicranella rufescens (Dicks.) Schimp. – is VU in Romania. Not redlisted in the neighbouring countries. A tiny species of wet muddy soils is overlooked and undercollected. We can confirm its presence in the gorge.

Grimmia trichophylla Grev. – is treated as vulnerable (VU) in Romania (Ştefănuţ and Goia 2012), it is known just from few localities in the country: jud. Alba: Mţii Apuseni-Detunata; jud. Hunedoara: Mţii Retezat-VI. Zlătuia; jud. Sibiu: Mţii Cibinului Dl. Ursului (Mohan 1998).

Lophozia longidens (Lindb.) Konstant. & Vilnet – is a circumboreal, subarctic, montane-subalpine in Central Europe, associated with acidic substrates in spruce forests, like siliceous rocks, bases of old tree trunks, decaying wood and peat bogs. Quite rare in the Romanian Carpathians: Ţibleş, Ciucaş, Suceava (Mohan 1998) and Metaliferi Mountains (Sass-Gyarmati *et al.* 2005).

Riccardia palmata (Hedw.) Carruth. – is a circumboreal, mountain species, it is reported only from one locality in Gutâi Mountains: Cheile Tătaru at Mara (Boros and Vajda 1967) and we could confirm its presence here. Also found in Gutai Mountains at Creasta Cocoşului are (Sass-Gyarmati 2019). Other reports from surroundings are from Borşa, Secului Valley, Sighet, Poiana Şarampoiului Forest, Mara, Runc Valley, Puzdra Mountain, (Boros and Vajda 1967); between Tocila Valley and Băiuţ (Jakab 1999), well distributed in the Romanian Carpathians (Mohan 1998).

Plagiomnium elatum (Bruch & Schimp.) T.J.Kop. – is a boreal moss (Düll 1985), NT in Romania. Not red listed in the neighbouring countries, is a characteristic *Plagiomnium* species of wetlands.

Plagiomnium ellipticum (Brid.) T.J.Kop. – is a boreal species, (Düll 1985), VU in Romania (Stefănuț and Goia 2012). It is a rare wetland species.

Racomitrium elongatum Ehrh. ex Frisvoll – The species is a member of the *Racomitrium canescens* (Hedw.) Brid. group with a boreal distribution (Düll 1994). Typically grows on acid rocks in mountain areas with an oceanic climate, NE (not evaluated) status in Romania (Stefănuț and Goia 2012).

Tetraphis pellucida Hedw. – is a montane species, frequent in Romania in montane and subalpine habitats.

These findings should enhance the knowledge of bryoflora, the results emphasizes the importance of conservation in its pristine condition this highly valuable area. It is imperative to maintain this natural habitat for future ecological research and sustainability.

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