ACTA
BIOLOGICA PLANTARUM
AGRIENSIS

TOMUS 8.

REDIGIT
PÉTER SZŰCS

EGER 1774
ESZTERHÁZY KÁROLY UNIVERSITY

EGER, 2020

ACTA BIOLOGICA PLANTARUM AGRIENSIS

(ABPA)

from Acta Academiae Paedagogicae Agriensis Sectio Biologiae

a *Journal of Plant Biology*

TOMUS 8.

EGER 1774
ESZTERHÁZY KÁROLY UNIVERSITY

Eger
2020

HU ISSN 2061-6716 (Print)
HU ISSN 2063-6725 (Online)
Papers of this volume are available: http://abpa.ektf.hu/

A kiadásért felelős az Eszterházy Károly Egyetem rektora
Megjelent az EKE Líceum Kiadó gondozásában/Published by Líceum Publisher EKE
Kiadóvezető/Head of publisher: Nagy Andor
Tördelőszerkesztő/Layout editor: Szűcs Péter
Megjelent/Year of publication: 2020
Nyomdai munkák: Eszterházy Károly Egyetem nyomdája
/Printed by Károly Eszterházy University Press
Felelős vezető/Responsible of printing: Kérészy László

Acta Biologica Plantarum Agriensis **8**(1): 3–16 (2020)
ISSN 2061-6716 (Print), 2063-6725 (Online)
http://abpa.ektf.hu/

DOI:10.21406/abpa.2020.8.1.3
Research article

THE BRYOPHYTE DIVERSITY OF CENTRAL PARK (ARCHBISHOP'S GARDEN) OF EGER TOWN (HUNGARY)

Péter Szűcs[1]*, Gabriella Fintha[1] & Gergő Fazekas[2]

*[1]Eszterházy Károly University, Institute of Biology, Department of Botany and Plant Physiology, H-3300 Eger, Leányka u. 6, Hungary; [2]Eszterházy Károly University, H-3300 Eger, Leányka u. 6, Hungary; *E-mail: szucs.peter@uni-eszterhazy.hu*

Abstract: The objective of the present work was the evaluation of the bryophyte diversity of the central park of Eger town. Altogether 59 taxa (4 liverworts and 55 mosses) were recorded. Nearly half of the identified species (49%) belong to three families: Orthotrichaceae, Pottiaceae, and Brachytheciaceae. *Brachythecium glareosum*, *Cirriphyllum piliferum*, *Eucladium verticillatum*, *Orthotrichum obtusifolium* and *Orthotrichum pumilum* are rated near threatened (NT) according to the Hungarian Red List. Some of the taxa found in Eger were not known from other central east european urban parks (*Ctenidium molluscum*, *Hygroamblystegium tenax*, *Pohlia melanodon*, *Cirriphyllum crassinervium*, *Hypnum cupressiforme* var. *lacunosum*, *Orthotrichum stramineum* and *Orthotrichum striatum*). There are remarkable differences between central park of Eger and other Central and Eastern European parks regarding species composition and the percentage of species in each of the life strategy categories.

Keywords: downtown, urban area, Sørensen index, comparison

INTRODUCTION

There is growing recognition of urban areas as hosts for innovative ways to conserve and promote biodiversity. Parks, as one specific type of urban green space, constitute particularly important biodiversity hotspots in the cityscape (Nielsen *et al.* 2014).

In Central and Eastern Europe several publications address the bryophyte flora or diversity of parks and gardens in urban areas, for example Warsaw, Łódź and other Polish cities (Fudali 2006, Wolski *et al.* 2012), Lviv (Mamchur *el al.* 2018), Bucharest (Gomoiu and Ștefănuţ 2008), Veľký Krtíš (Mišíková *et al.* 2007), Sofia (Gospodinov *et al.* 2018) and Bratislava (Godovičová and Mišíková

2017). The aim of this study was the examination of the bryophyte diversity of the central park of Eger town.

Knowledge about the bryophytes of anthropogenic habitats in Hungary is limited, and the following papers focus mostly on the description of floristic data: Budapest city (Szepesfalvi 1940, 1941, 1942) Barcs (Szűcs *et al.* 2014), the towns of Sopron (Szűcs 2015) and Gödöllő (Király *et al.* 2019). The bryophyte flora of Almásfüzitő (Szűcs *et al.* 2017a), Balaton village (Zsólyom and Szűcs 2018), and of the manor park of Martonvásár village (Nagy *et al.* 2016) are well documented in Hungary. However, there are no publications aiming at completeness with respect to the bryophyte diversity of parks of downtown areas in Hungary.

MATERIALS AND METHODS

Site details descriptions include data in the following order in the Appendix: habitat, GPS-coordinates, date of collection. Based on the Central European Flora Mapping System (Király *et al.* 2003), each collection point belongs to the 8188.1 square. The nomenclature follows Király (2009) for vascular plants, Söderström *et al.* (2016) for liverworts, and Hill *et al.* (2006) for mosses. In order to characterise the conservation and indicator status of taxa the Hungarian Red List was used (Papp *et al.* 2010). We used the Sørensen index (Sørensen 1948) for the comparison of the species composition of different localities. Collected specimens are deposited at the Cryptogamic Herbarium of the Department of Botany and Plant Physiology at the Eszterházy Károly University, Eger (EGR).

Study area

The town of Eger belongs to the Eger-Bükkalja micro-region, which is a colline area at an elevation of 126 to 420 m above sea level, slightly sloping to the south-east. The settlement is situated on the terraced valley of the Eger Creek, and to a smaller extent it covers the hillside accompanying the valley of the Tárkány Creek (Sugár 1983).

It is a region with a moderately warm to moderately dry climate. The average annual temperature is 9.0–10.0 °C at the highest points. The average annual precipitation is approximately 600 mm, of which 340–380 mm is produced during the vegetation period. The likelihood of rainfall is the highest in early summer and late autumn (Dövényi 2010, Sugár 1983).

The dominance of the North-West winds is evident in every season, which is particularly characteristic during the summer months. In terms of wind flow speed, Eger is classified into the moderately windy areas of Hungary, which is also indicated by the relatively high frequency of wind silences (Dövényi 2010, Sugár 1983). The Eger Creek forms the boundary of the park in the north-east. The groundwater of the Eger valley has particularly hard water rich in sodium-calcium-hydrogencarbonate and sulfate (Sugár 1983).

The historical descriptions from the 15th and 16th century refer to this part of our town as a rich forested area where the wildlife park was located which was probably established by a bishop of Eger from the Renaissance. The former wildlife park was much larger than today's Archbishop's Garden. It included the area of today's Thermal bath, the present Archbishop's Garden, and also the Csákó district, which extends to the present railway station.

In the park there is an ornamental garden, on the left side of the creek there is a flower garden up to the mills, and between the mills and today's Csákány street there is a vegetable garden which together formed the old bishop's garden. The episcopal ornamental garden was established during the time of Ferenc Barkóczy (1710–1765), in the style of French gardens. In 1769 Bishop Károly Eszterházy (1725–1799) initiated the construction of a stone fence and ornate baroque gates to replace the old wooden fence. The oldest trees in the park are sycamore trees, estimated to be between 300 and 400 years old. Lajos Szmrecsányi (1851–1943) opened the gates of the 14-hectare Archbishop's Garden to the citizens of the town in 1919, and since then the area has been under significant human influence. (Herzegné Székely 2010). As for the park's maintenance, lawn mowing and leaf collection is performed regularly.

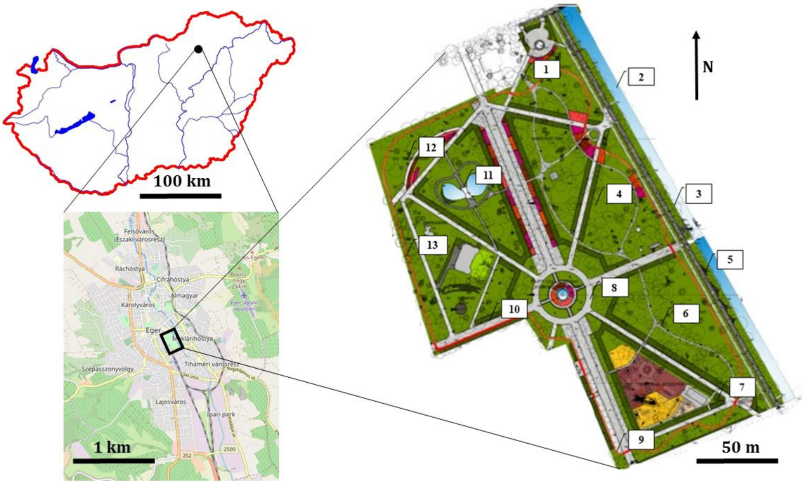


Figure 1. The situation of Eger town, and the map and collecting points of the central park of Eger.

RESULTS AND DISCUSSION

List of species

Numbers refer to sites (*Figure 1*) listed in the Appendix. The substrates given after a colon refer to all listed sites.

Marchantiophyta

Frullania dilatata (L.) Dumort. – LC – 1: bark of *Fraxinus*
Marchantia polymorpha subsp. ***ruderalis*** Bischl. & Boisselier – LC – 3: rock in stream water
Pellia endiviifolia (Dicks.) Dumort. – LC - 2: rock in stream water
Radula complanata (L.) Dumort. – LC – 1: bark of *Sophora japonica*; 6, 7: bark of *Acer negundo*; 5: bark of *Ailanthus altissima*

Bryophyta

Amblystegium serpens (Hedw.) Schimp. – LC – 1: artifical stone; 3: wall of the stream; 4, 5: soil; 7, 12: concrete and mown lawn
Atrichum undulatum (Hedw.) P. Beauv. – LC – 7, 9: shaded soil

Barbula unguiculata Hedw. – LC – 1, 3, 13: shaded soil

Brachytheciastrum velutinum (Hedw.) Ignatov & Huttenen – LC – 12: concrete

Brachythecium glareosum (Bruch ex Spruce) Schimp. – NT – 3: wall of the stream bank

Brachythecium rutabulum (Hedw.) Schimp. – LC – 1, 9: soil; 4, 5, 6: mown lawn; 10: stone fence; 11: artifical stone; 12: concrete; 13: andesit stone wall

Brachythecium salebrosum (F. Weber et D. Mohr) Schimp. – LC – 5: bark of *Ailanthus altissima*

Bryoerythrophyllum recurvirostrum (Hedw.) P. C. Chen – LC-att – 3: wall of the stream bank

Bryum argenteum Hedw. – LC – 1, 8, 11: artifical stone; 3: wall of the stream; 7, 8, 12: concrete; 10: stone fence

Bryum caespiticium Hedw. – LC – 1: concrete

Bryum capillare Hedw. – LC – 3: soil bank

Bryum moravicum Podp. – LC – 3: wall of stream bank, bark of *Tilia platyphyllos*; 13: andesit stone wall; 5: bark of *Ailanthus altissima*

Calliergonella cuspidata (Hedw.) Loeske – LC – 11: artifical stone; 13: andesite stone wall

Ceratodon purpureus (Hedw.) Brid. – LC – 3: wall of the stream bank; 7, 8,12: concrete; 8, 10: soil

Cirriphyllum crassinervium (Taylor) Loeske & M. Fleisch – LC – 3: wall of stream bank

Cirriphyllum piliferum (Hedw.) Grout – NT – 1: soil

Cratoneuron filicinum (Hedw.) Spruce – LC – 2: rock in stream water

Ctenidium molluscum (Hedw.) Mitt. – LC – 5: rubble

Didymodon rigidulus Hedw. – LC-att – 7, 8: concrete; 10: stone fence

Eucladium verticillatum (With.) Bruch & Schimp. – NT – 2: artifical stone in thermal water

Fissidens taxifolius Hedw. – LC – 5, 7, 9: shaded soil

Grimmia pulvinata (Hedw.) Sm. – LC – 1, 8: artifical stone; 7, 12: concrete; 13: andesite stone

Homalia trichomanoides (Hedw.) Brid. – LC-att – 5: rubble

Homalothecium lutescens (Hedw.) H. Rob. –LC – 4, 6: soil

Homalothecium sericeum (Hedw.) Schimp. LC – 3: wall of the stream bank, 13: andesite stone wall

Homomallium incurvatum (Schrad. ex Brid.) Loeske – LC – 3: wall of the stream bank

Hygroamblystegium tenax (Hedw.) Jenn. – LC – 2: rock in stream

Hygroamblystegium varium (Hedw.) Mönk. – LC-att – 11: wet soil on lake shore

Hypnum cupressiforme Hedw.- LC – 1: artifical stone, bark of *Fraxinus*, and *Sophora japonica* 3: wall of the stream bank; 4, 5, 6, 7: soil; 13: andesite stone

Hypnum cupressiforme var. ***lacunosum*** Brid. – LC – 3: wall of the stream bank

Leskea polycarpa Ehrh. ex Hedw. – LC – 1: bark of *Sophora japonica;* 3, 4, 5, 6: bark of *Acer negundo; 5:* bark of *Tilia platyphyllos*

Leucodon sciuroides (Hedw.) Schwägr. – LC – 1: bark of *Fraxinus*

Orthotrichum affine Schrad. ex Brid. – LC – 1: bark of *Fraxinus*; and *Sophora japonica;* 4, 6: bark of *Acer negundo*; 5: bark of *Tilia platyphyllos* and *Ailanthus altissima*

Orthotrichum anomalum Hedw. – LC – 1: artifical stone; 3: wall of the stream bank; 13: andesite stone wall

Orthotrichum cupulatum Hoffm. ex Brid. – LC-att – 1: artifical stone; 3: wall of the stream bank

Orthotrichum diaphanum Schrad. ex Brid. – LC – 1, 4, 6: bark of *Acer negundo*

Orthotrichum obtusifolium Brid. – NT – 1: bark of *Fraxinus*; 4, 6: bark of *Acer negundo;* 10: bark of *Sophoria japonica*

Orthotrichum pallens Bruch ex Brid. – LC – 5, 7: bark of *Tilia platyphyllos;* 10: bark of *Sophoria japonica*

Orthotrichum pumilum Sw. ex anon. – NT – 10: stone fence

Orthotrichum speciosum Nees – LC-att – 5, 7: bark of *Tilia platyphyllos*

Orthotrichum stramineum Hornsch. ex Brid. – LC – 5, 7: bark of *Tilia platyphyllos;* 10: bark of *Sophoria japonica*

Orthotrichum striatum Hedw. – LC-att – 5, 7: bark of *Tilia platyphyllos*

Oxyrrhynchium hians (Hedw.) Loeske – LC – 1, 4, 5, 6, 7, 8, 12: soil

Plagiomnium cuspidatum (Hedw.) T. J. Kop. – LC – 3, 7, 9: soil

Plagiomnium undulatum (Hedw.) T. J. Kop. – LC – 7, 9: soil

Platygyrium repens (Brid.) Schimp. – LC – 5: artifical stone; 7, 8: concrete

Pohlia melanodon (Brid.) A.J. Shaw – LC – 3: soil of stream bank

Pylaisia polyantha (Hedw.) Schimp. – LC – 1: bark of *Fraxinus*; 4, 6: bark of *Acer negundo*

Schistidium crassipilum H. H. Blom – LC – 1: artifical stone; 3: wall of the stream bank

Sciuro-hypnum populeum (Hedw.) Ignatov & Huttunen – LC – 3: wall of the stream bank

Syntrichia papillosa (Wilson) Jur. – LC-att – 1: bark of *Fraxinus*

Syntrichia ruralis (Hedw.) F. Weber & D. Mohr – LC – 3: wall of the stream bank

Syntrichia virescens (De Not.) Ochyra – LC-att – 1: bark of *Fraxinus*; 3: wall of the stream bank; 10: stone fence

Tortula muralis Hedw. – LC – 1, 8, 11: artifical stone; 7, 8, 12: concrete

Tortula truncata (Hedw.) Mitt. – LC – 7: soil

Bryophyte diversity

Altogether 59 bryophytes were detected in the central park of Eger, which include 4 liverworts (7%) and 55 mosses (93%). The liverwort species belong to 4 families and 4 genera, while the mosses belong to 16 families and 32 genera (*Figure 2*).

Nearly half of the species (49.15%) belong to the 3 families Orthotrichaceae (10 taxa), Brachytheciaceae (10 taxa) and Pottiaceae (9 taxa).

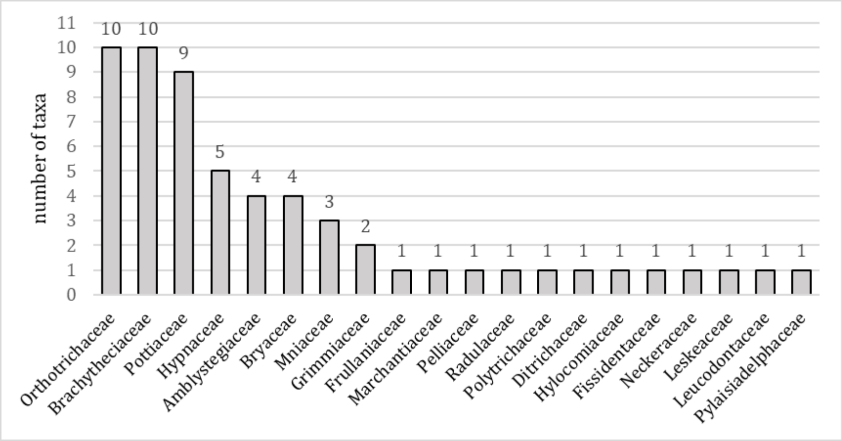


Figure 2. Distribution of bryophyte species found in the central park of the Eger town among families (Taxonomy follows Goffinet and Shaw 2009 and Söderström *et al.* 2016).

Many of the common mosses of the central park of Eger, including *Amblystegium serpens, Barbula unguiculata, Brachythecium rutabulum, B. salebrosum, Bryum argenteum, Ceratodon purpureus, Fissidens taxifolius, Grimmia pulvinata, Hypnum cupressiforme, Leskea polycarpa, Leucodon sciuroides, Orthotrichum affine, O. anomalum, O. diaphanum, O. pumilum, Oxyrrhynchium hians, Plagiomnium undulatum, Platygyrium repens, Pylaisia polyantha, Syntrichia ruralis* and *Tortula muralis* have also been found in some other parks of Central East European settlements (Fudali 2006, Wolski *et al.* 2012, Mamchur *el al.* 2018, Gomoiu and Ştefănuţ 2008, Mišíková *et al*. 2007, Gospodinov *et al*. 2018 and Godovičová and Mišíková 2017).

Atrichum undulatum and *Homalia trichomanoides* occur in some Central East European parks (Fudali 2006, Mamchur *el al.* 2018) and *Eucladium verticillatum* was detected in the forest park of Lviv city (Mamchur *el al.* 2018). These three species are not known from other hungarian settlements (Szűcs *et al.* 2017a, Zsólyom and Szűcs 2018).

There are a few taxa in Eger, which are not known in the Central East European urban parks (Fudali 2006, Wolski *et al.* 2012, Mamchur *et al.* 2018, Gomoiu and Ştefănuţ 2008, Mišíková *et al*. 2007, Gospodinov *et al*. 2018 and Godovičová and Mišíková 2017), for example *Ctenidium molluscum*, *Hygroamblystegium tenax*, *Pohlia melanodon, Cirriphyllum crassinervium*, *Hypnum cupressiforme* var. *lacunosum*, *Orthotrichum stramineum* and *O. striatum*.

Table 1 shows the values of the Sørensen index, which are derived from a comparison of moss species in the region, in Central and Eastern European parks, and the central park of Eger. The values of parks in Central and Eastern Europe are similar in this respect, there is no notable difference between the calculated data (0.415–0.590). Compared to the park of Eger, the discrepancy is most pronounced in the case of the Teaching Garden, Łódź (0.415). The Botanical Garden or EKU is situated closest to the central park of Eger and shows the greates similarity in species composition (highest Sørensen index of 0.590).

Table 1. Comparison of the area, the distance of localities from Eger, the altitude, the number of taxa and Sørensen index of central east european parks with central park of Eger town.

Name of locality	area (hectare)	distance from Central park of Eger (km)	alt (meter a.s.l.)	number of taxa	Sørensen index
Central Park of Eger (present study)	14	0	155-165	59	1
Botanical Garden of EKU, Eger (Hungary) (Szűcs *et al.* 2017b)	1	1.3	230	46	0.590
Mátrai Sanatorium, Mátraháza (Hungary) (Szűcs *et al.* 2018)	14	30.4	650-700	65	0.564
Horka forest park, Velky Krtís, (Slovakia) (Mišíková *et al.* 2007)	1500	83	210	37	0.541
Vrana Park, Sofia (Bulgaria) (Gospodinov *et al.* 2018)	99.3	623	560	68	0.488
Pohulyanka forest park, Lviv, (Ukraine) (Mamchur *et al.* 2018)	129	343	340-370	108	0.455
Exp. and Teaching Garden, Łódź, (Poland) (Wolski *et al.* 2012)	1	435	225	42	0.415

Conservation status

Five taxa belong to the near threatened (NT) category according to the Hungarian Red List (Papp *et al.* 2010): *Brachythecium glareosum, Cirriphyllum piliferum, Eucladium verticillatum, Orthotrichum obtusifolium* and *Orthotrichum pumilum.* Another eight species belong to least concern attention (LC-att), viz. *Bryoerythrophyllum recurvirostrum*, *Didymodon rigidulus, Homalia trichomanoides, Hygroamblystegium varium, Orthotrichum cupulatum, Orthotrichum striatum, Syntrichia papillosa,* and *Syntrichia virescens*.

Some indicator mosses (species which by their presence indicate a higher conservation value of the habitat) also occur in the park, for example *Cirriphyllum piliferum, Eucladium verticillatum, Homalia trichomanoides*, *Hygroamblystegium varium,* *Orthotrichum cupulatum*, *O. obtusifolium*, *O. pumilum*, *O. speciosum*, *O. striatum*, and *Syntrichia papillosa.*

Life strategies

There is a remarkable difference between central park of Eger and other local or regional habitats (*Table 1*) concerning the percentage of species in each of the life strategy categories (Dierßen 2001).

Central park of Eger is more abundant in colonists and pioneer colonists, and less abundant in long-lived shuttle, perennial, and competitive perennial species, compared to the other habitats. None of the bryophytes in the Central park of Eger belong to the ephemeral colonist and fugitive categories (*Figure 3*).

A possible explanation for the above phenomenon is that abundant bare soil surface is available for the bryophytes, but disturbed substrates are very rare in the studied area.

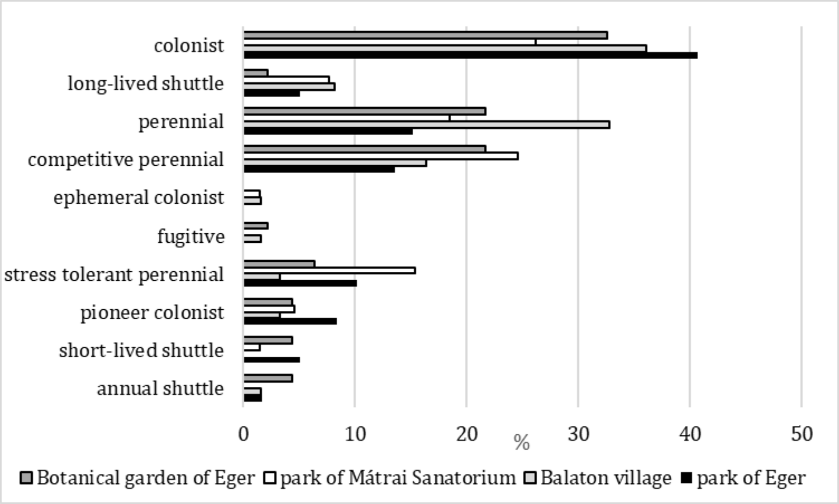


Figure 3. Comparison of the life strategies of bryophytes in Botanical Garden of Eger (Szűcs *et al.* 2017b), park of Mátrai Sanatorium (Mátraháza) (Szűcs *et al.* 2018), Balaton village (Zsólyom and Szűcs 2018) and central park of Eger (present study).

CONCLUSIONS

The central park of Eger has a remarkable bryophyte diversity, which is of comparable magnitude to local, regional places and central east european urban parks in accordance with its size. The high number of indicator mosses shows a high level of conservation value of the park.

The rich bryophyte flora partly can be expained by the history of the different habitats, the abundance of old and varied deciduous trees and the proximity of Eger creek and the municipal thermal spa.

Acknowledgement – The authors would like to express their gratitude to Peter Erzberger and Andrea Sass-Gyarmati for their useful comments. The authors would like to thank Tamás Pócs for the identification of *Eucladium verticillatum*. The first author's research was supported by the grant EFOP-3.6.1-16-2016-00001 ("Complex improvement of research capacities and services at Eszterházy Károly University"). Special thanks to András Vojtkó and András Schmotzer for their help with literature.

REFERENCES

DIERßEN, K. (2001). Distribution, ecological amplitude and phytosociological characterization of European bryophytes. *Bryophytorum Bibliotheca* **56**: 1–289.

DÖVÉNYI, Z. (ed.) (2010). *Magyarország kistájainak katasztere*. MTA Földrajztudományi Kutatóintézet, Budapest, 824 pp.

FUDALI, E. (2006). Influence of city on the floristical and ecological diversity of bryophytes in parks and cemeteries. *Biodiversity. Research and conservation* **1–2**: 131–137.

GODOVIČOVÁ, K. & MIŠÍKOVÁ, K. (2017). Epifytické machorasty urbánneho prostredia Bratislavy (Epiphytic bryophytes in the urban environment of Bratislava). *Bryonora* **59**: 44–57.

GOFFINET, B. & SHAW, A.J. (eds.) (2009). *Bryophyte biology.* Cambridge University Press, Cambridge, 565 pp.

GOMOIU, I. & ŞTEFĂNUŢ, S. (2008). *Lichen and bryophytes as bioindicator of air pollution.* In: CRACIUM, I. (ed.) *Species monitoring in the central parks of Bucharest.* Universitatea din Bucureşti, Bucharest, pp. 14–25.

GOSPODINOV, G., LAMBEVSKA-HRISTOVA, A., NATCHEVA, R. & GYOSHEVA, M. (2018). Vrana Park – a neglected site for bryophyte and fungal diversity in Sofia city. *Phytologia Balcanica* **24**(3): 323–329.

HERZEGNÉ SZÉKELY, A. (2010). *Az Érsekkert története.* In: RENN, O. (ed.): *Egri séták nemcsak Egrieknek I.* Egri Lokálpatrióta Egylet, Eger, pp. 133–147.

HILL, M.O., BELL, N., BRUGGEMAN-NANNAENGA, M.A., BRUGUES, M., CANO, M.J., ENROTH, J., FLATBERG, K.I., FRAHM, J.-P., GALLEGO, M.T., GARILLETI, R., GUERRA, J., HEDENÄS, L., HOLYOAK, D.T., HYVÖNEN, J., IGNATOV, M.S., LARA, F., MAZIMPAKA, V., MUÑOZ, J. & SÖDERSTRÖM, L. (2006). An annotated checklist of the mosses of Europe and Macaronesia. *Journal of Bryology* **28**: 198–267. https://doi.org/10.1179/174328206x119998

KIRÁLY, G., BALOGH, L., BARINA, Z., BARTHA, D., BAUER, N., BODONCZI, L., DANCZA, I., FARKAS, S., GALAMBOS, I., GULYÁS, G., MOLNÁR, V. A., NAGY, J., PIFKÓ, D., SCHMOTZER, A., SOMLYAI, L., SZMORAD, F., VIDÉKI, R., VOJTKÓ, A., & ZÓLYOMI, SZ. (2003). A magyarországi flóratérképezés módszertani alapjai. *Flora Pannonica* **1**: 3–20.

KIRÁLY, G. (ed.) (2009). *Új magyar füvészkönyv* (Magyarország hajtásos növényei, határozókulcsok). Aggteleki Nemzeti Park Igazgatóság, Jósvafő, 616 pp.

KIRÁLY, G., BARÁTH, K., BAUER, N., ERZBERGER, P., PAPP, B., SZŰCS, P., VERES, SZ. & BARINA, Z. (2019). Taxonomical and chorological notes 8 (85–93). *Studia botanica hungarica* **50**(1): 241–252. https://doi.org/10.17110/StudBot.2019.50.1.241

MAMCHUR, Z., DRACH, Y. & DANYLKIV, I. (2018). Bryoflora of the "Pohulyanka" forest park (Lviv city) I. Changes in taxonomic composition under anthropogenic transformation. *Studia Biologica* **12**(1): 99–112. https://doi.org/10.30970/sbi.1201.542

MIŠÍKOVÁ, K., MIŠIK, M. & KUBINSKÁ, A. (2007). Bryophytes of the forest park Hôrka (Veľký Krtíš town, Slovakia). *Acta Botanica Universitatis Comenianae* **43**: 9–13.

NAGY, Z., MAJLÁTH, I., MOLNÁR, M. & ERZBERGER, P. (2016). A martonvásári kastélypark mohaflórája. (Bryofloristical study in the Brunszvik manor park in Martonvásár, Hungary). *Kitaibelia* **21**(2): 198–206. https://doi.org/10.17542/kit.21.198

NIELSEN, A.B., BOSCH, M., MARUTHAVEERAN, S. & BOSCH, C.K. (2014). Species richness in urban parks and its drivers: A review of empirical evidence. *Urban Ecosystems* **17**(1): 305–327. https://doi.org/10.1007/s11252-013-0316-1

PAPP, B., ERZBERGER, P., ÓDOR, P., HOCK, ZS., SZÖVÉNYI, P., SZURDOKI, E. & TÓTH, Z. (2010). Updated checklist and red list of Hungarian bryophytes. *Studia botanica hungarica* **41**: 31–59.

SØRENSEN, T. (1948). A method of establishing groups of equal amplitude in plant sociology based on similarity of species content. Kongelige Dansk e Videnskabernes Selskab*. Biologiske Skrifter* **4**: 1–34.

SÖDERSTRÖM, L., HAGBORG, A., VON KONRAT, M., BARTHOLOMEW-BEGAN, S., BELL, D., BRISCOE, L., BROWN, E., CARGILL, D.C., COSTA, D.P., CRANDALL-STOTLER, B.J., COOPER, E.D., DAUPHIN, G., ENGEL, J.J., FELDBERG, K., GLENNY, D., GRADSTEIN, S.R., HE, X., HEINRICHS, J., HENTSCHEL, J., ILKIU-BORGES, A.L., KATAGIRI, T., KONSTANTINOVA, N.A., LARRAÍN, J., LONG, D.G., NEBEL, M., PÓCS, T., PUCHE, F., REINER-DREHWALD, E., RENNER, M.A.M., SASS-GYARMATI, A., SCHÄFER-VERWIMP, A., MORAGUES, J.G.S., STOTLER, R.E., SUKKHARAK, P., THIERS, B.M., URIBE, J., VÁŇA, J., VILLARREAL, J.C., WIGGINTON, M., ZHANG, L. & ZHU, R.-L. (2016). World checklist of hornworts and liverworts. *PhytoKeys* **59**: 1–828. https://doi.org/10.3897/phytokeys.59.6261

SUGÁR, I. (1983). Eger Gyógyvizei és fürdői. Eger Város Tanácsa V. B. Műszaki Osztálya és a Heves megyei Idegenforgalmi Hivatal kiadása, Eger, 441 pp.

SZEPESFALVI, J. (1940). Die Moosflora der Umgebung von Budapest und des Pilisgebirges I. *Annales historico-naturales Musei nationalis hungarici* **33**: 1–104.

SZEPESFALVI, J. (1941). Die Moosflora der Umgebung von Budapest und des Pilisgebirges II. *Annales historico-naturales Musei nationalis hungarici* **34**: 1–71.

SZEPESFALVI, J. (1942). Die Moosflora der Umgebung von Budapest und des Pilisgebirges III. *Annales historico-naturales Musei nationalis hungarici* **35**: 1–72.

SZŰCS, P. (2015). *Kindbergia praelonga* (Hedw.) Ochyra Sopron város mohaflórájában. *Kitaibelia* **20**(2): 305.

SZŰCS, P., BÖRCSÖK, Z. & KÁMÁN, O. (2014). A *Riccia glauca* L. és a *Riccia sorocarpa* Bisch. előfordulása Barcs belterületén. *Kitaibelia* **19**(2): 365.

SZŰCS, P., PÉNZES-KÓNYA, E. & HOFMANN, T. (2017a). The bryophyte flora of the village of Almásfüzitő, a former industrial settlement in NW-Hungary. *Cryptogamie, Bryologie* **38**(2): 153–170. https://doi.org/10.7872/cryb/v38.iss2.2017.153

SZŰCS, P., TÁBORSKÁ, J., BARANYI, G. & PÉNZES-KÓNYA, E. (2017b). Short-term changes in the bryophyte flora in the Botanical Garden of Eszterházy Károly University (Eger, NE Hungary). *Acta Biologica Plantarum Agriensis* **5**(2): 52–60. https://doi.org/10.21406/abpa.2017.5.2.52

SZŰCS, P., BARANYI, G. & FINTHA, G. (2018). The bryophyte flora of the park of Mátrai Gyógyintézet Sanatorium (NE Hungary). *Acta Biologica Plantarum Agriensis* **6**: 123–132. https://doi.org/10.21406/abpa.2018.6.123

WOLSKI, G.J., STEFANIAK, A. & KOWALKIEWICZ, B. (2012). Bryophytes of the experimental and teaching garden of the faculty of biology and environmental protection, University of Łódź (Poland). *Ukrainian Botanical Journal* **69**(4): 519–529.

ZSÓLYOM, D. & SZŰCS, P. (2018). Balaton település (Heves megye) mohaflórája. (The bryophyte flora of Balaton village (Heves county, Hungary)). *Botanikai Közlemények* **105**(2): 231–242. https://doi.org/10.17716/BotKozlem.2018.105.2.231

(submitted: 10.12.2019, accepted: 27.04.2020)

APPENDIX

Site details

1. soil, artifical stone, bark of trees (04.12.2018, 01.08.2019, 02.03.2020) N47°53’53” E20°22’46”
2. rock in stream water (14.06.2019) N47°53’52” E20°22’52”
3. wall of the stream bank, soil (14.06.2019, 03.07.2019.) N47°53’49” E20°22’54”
4. mown lawn, soil (13.03.2019., 03.07.2019) N47°53’50” E20°22’50”
5. mown lawn, soil, bark of trees (13.03.2019) N47°53’47” E20°22’56”
6. bark of trees, soil, mown lawn (17.04.2019) N47°53’46” E20°22’54”
7. mown lawn, bark of trees, concrete (11.03.2019) N47°53’41” E20°22’54”
8. artifical stone, soil, concrete (11.03.2019) N47°53’45” E20°22’50”
9. soil (11.03.2019) N47°53’40” E20°22’52”
10. stone fence, soil (11.03.2019) N47°53’44” E20°22’44”
11. artifical lake (14.06.2019) N47°53’49” E20°22’45”
12. mown lawn, concrete, soil (13.03.2019) N47°53’51” E20°22’40”
13. artifical stone wall (14.04.2019, 14.06.2019) N47°53’48” E20°22’44”

Acta Biologica Plantarum Agriensis **8**(1): 17–20 (2020)
ISSN 2061-6716 (Print), 2063-6725 (Online)
http://abpa.ektf.hu/

DOI:10.21406/abpa.2020.8.1.17
Short communication

IRIS SIBIRICA HAS UNUSUAL, PRACTICALLY NON-DIFFERENT HEAT TOLERANCE OF PS II AT DIFFERENT GEOGRAPHICAL EXPOSURES ON THE BÜKK-PLATEAU (NORTH-HUNGARY)

Sándor Dulai[1]*, Réka Tarnai[1], Dóra Szopkó[1], Anna E-Vojtkó[2], Dóra Salamon[1], Ammar Allem[3] & András Vojtkó[1]

*[1]Eszterházy Károly University, Department of Botany and Plant Physiology, H-3300 Eger, Leányka u. 6, Hungary; [2]Department of Botany, University of South Bohemia, České Budějovice, Czechia; [3]Biological Doctoral School, St. Stephen University, H-2011 Gödöllő, Hungary; *E-mail: dulai.sandor@uni-eszterhazy.hu*

Abstract: Heat tolerance of photosystem II (PSII) was examined in Siberian flag (*Iris sibirica* L.) living in different microhabitats of a non-forested enclosed depression surface (doline) on the Bükk-plateau. Although the microclimatic parameters of the habitats with different facing sites show sharp contrasts, there was no significant difference between the heat tolerance of PSII in leaves of *I. sibirica* growing in these expositions neither in dark- nor in light-adapted state.

Keywords: *Iris sibirica*, heat tolerance, photosynthesis, microhabitat

INTRODUCTION

Doline vegetation is influenced by a number of factors including altitude and significant differences in microclimate or soil moisture along slopes with given exposures. Due to the zone inversion parallel to climate change, montane species are already present on the bottom and northern slopes, while on southern slopes, species that favor drier and warmer climates are common (Bátori *et al.* 2014). However, some species occur in both aspects of the doline despite significant differences in air temperature, air and soil humidity, which requires special adaptation/acclimation strategies, especially against the temperature. In this paper, we present some preliminary results on the unusual, practically non-different heat tolerance of *I. sibirica* growing in different exposures.

MATERIALS AND METHODS

Leaves of Siberian flag (*Iris sibirica* L.) were collected from five different geographical exposition (bottom, N/S facing slopes, and N/S edge) of the second biggest non-forested doline in the Bükk-plateau (North-Hungary) between six and seven am. The collected leaves were immediately wrapped in wet filter paper and transported to the laboratory within half an hour. The *in vivo* heat induced chlorophyll *a* fluorescence was measured in dark- and light-adapted state of leaves with a pulse amplitude modulated fluorometer (IMAGING PAM M-series, Walz, Effeltrich, Germany). The leaves were dark-adapted for 30 min, and then placed on the thermoelectric module. The minimum chlorophyll fluorescence (F_0) was monitored in leaf discs exposed to elevated temperature from 25°C to 49-55°C at a rate of 1°C min^{-1}. Recording of the steady-state fluorescence (F_s) *v*. T curves started when the photosynthesis was steady at 200 and 1,000 µmol m^{-2} s^{-1} actinic light intensity. Critical temperature (T_c) values were determined as the interception of regression lines fitted to F_0 and F_s data.

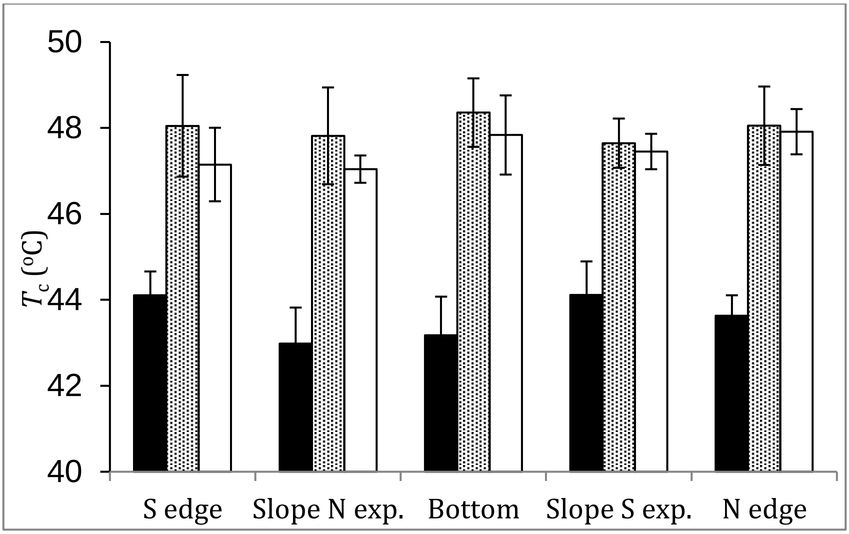


Figure 1. Critical temperature (T_c) values of F_0 (filled bars) and F_s *v. T* curves at 200 (semi closed bars) and 1000 (empty bars) µmol m-2 s^{-1} acinic light (AL) intensities in leaves of *Iris sibirica* growing in different facing parts of the doline.

RESULTS AND DISCUSSION

The heat sensitivity of the photosynthetic apparatus is closely connected to the thermal stability of PSII, which is well characterised by critical values of the temperature dependence of the initial fluorescence (F_0) level in dark-adapted leaves (Dulai *et al*. 1998, Szopkó and Dulai 2018). There was no significant difference in the heat tolerance of PSII determined by the T_c values of the F_0 *v*. *T* curves (recorded practically in darkness) in leaves of *I. sibirica* living in different expositions and it was not sufficient for tolerating such high temperatures that are peculiar to the microhabitat of S facing slope above the surface (*Figure 1*). Similarly to F_0, the breakpoints (T_c) of temperature dependence of steady state fluorescence (F_s) appropriately show the thermal stability of samples at steady-state photosynthesis level in light adapted state (Molnár *et al*. 1998). In connection with this, T_c values of F_s *v*. *T* curves measured at 200 and 1000 μE m^{-2} s^{-1} actinic light intensity are shifted towards significantly higher temperatures (~47-48 ^{o}C), indicating the higher thermal tolerance of PSII (*Figure 1*). This is consistent with those observations that light stimulates the thermal stability of the PSII (Molnár *et al*. 1998, Dulai *et al*. 2006). However, ground level air temperatures on the S-facing slope can often exceed 47-48 ^{o}C with extreme daily fluctuations. So this reduced heat sensitivity is just enough to tolerate the high temperature conditions of the S-facing slope during the daytime period of a summer day. At the same time, the doline micro-habitats with different exposures are characterized by sharp and significant microclimatic variation (Bátori *et al*. 2019). These differences are manifested not only in the average daily means of ground level air temperature but also in daily maxima, daily range and air and soil moisture. As a result, more cool-adapted plants are found on the north-facing slopes and bottoms of dolines (Bátori *et al*. 2017). However, some species, like Siberian flag, occur in both aspects of the depression despite the above mentioned differences in environmental factors, which should require a very plastic phenotypic performance to temperature conditions. This would also require optimizing the heat stability of the photosynthetic apparatus according to the factors specific to the given exposure. Thus, it is very unusual for a plant to occur in all micro-habitats of the doline but not to be able to change its temperature tolerance neither in dark- nor in light-adapted state. Consequently, as the forecasted climatic extremities increase, this plant might disappear completely from the southern slope.

Acknowledgement – The first author's research was supported by the grant EFOP-3.6.1-16-2016-00001 ("Complex improvement of research capacities and services at Eszterházy Károly University").

REFERENCES

BÁTORI, Z., E-VOJTKÓ, A., ERDŐS, L. & VOJTKÓ, A. (2014). Temperature and soil moisture regimes of the forested and non-forested dolines of the Bükk Mountains based on ecological indicator values *(in Hungarian*). *Kitaibelia* **19**: 331–338.

BÁTORI, Z., VOJTKÓ, A., MAÁK, I.E., LŐRINCZI,, G., FARKAS, T., KÁNTOR, N., TANÁCS, E., KISS, P.J., JUHÁSZ, O., MÓDRA, G., TÖLGYESI, CS., ERDŐS, L., AGUILON, D.J. & KEPPEL, G. (2019). Karst dolines provide diverse microhabitats for different functional groups in multiple phyla. *Nature Scientific Reports* **9**: 1–13. 7176. https://doi.org/10.1038/s41598-019-43603-x

BÁTORI, Z., VOJTKÓ, A., FARKAS, T., SZABÓ, A., HAVADTŐI, K., E-VOJTKÓ, A., TÖLGYESI, CS., CSEH, V., ERDŐS, L., MAÁK, I.E. & KEPPEL, G. (2017). Large- and small-scale environmental factors drive distributions of cool-adapted plants in karstic microrefugia. *Annals of Botany* **119**: 301–309. http://dx.doi.org/10.1093/aob/mcw233

DULAI, S., MOLNÁR, I. & LEHOCZKI, E. (1998). Effects of growth temperature of 5 and 25 °C on long-term responses of photosystem II to heat stress in atrazine-resistant and susceptible biotypes of *Erigeron canadensis* (L.). *Australian Journal of Plant Physiology* **25**: 145–153. https://doi.org/10.1071/PP97112

DULAI, S., MOLNÁR, I., PRÓNAY, J., CSERNÁK, Á., TARNAI, R. & MOLNÁR-LÁNG, M. (2006). Effects of drought on photosynthetic parameters and heat stability of PSII in wheat and in *Aegilops* species originating from dry habitats. *Acta Biologica Szegediensis* **50**: 11–17.

MOLNÁR, I., CSIZI, K., DULAI, S., DARKÓ, É. & LEHOCZKI, E. (1998). *Light dependence of thermostability of photosynthetic apparatus.* In: GARAB, G. (ed.): *Photosynthesis: Mechanisms and Effects.* Kluwer Academic Publishers, Dordrecht, pp. 2241–2244. https://doi.org/10.1007/978-94-011-3953-3_524

SZOPKÓ, D. & DULAI, S. (2018). Environmental factors affecting the heat stability of the photosynthetic apparatus. *Acta Biologica Plantarum Agriensis* **6**: 90–107. https://doi.org/10.21406/abpa.2018.6.90

(submitted: 17.03.2020, accepted: 24.04.2020)

Acta Biologica Plantarum Agriensis **8**(1): 21–28 (2020)
ISSN 2061-6716 (Print), 2063-6725 (Online)
http://abpa.ektf.hu/

DOI:10.21406/abpa.2020.8.1.21
Research article

BRYOPHYTE COLLECTION IN THE HERBARIUM OF ESZTERHÁZY KÁROLY UNIVERSITY (EGR): THE DIGITAL DATABASE OF PEAT MOSS (*SPHAGNUM*) SPECIES

***Dávid Kapi*[1]* *& Andrea Sass-Gyarmati*[2]**

*[1]Kozma Andor u. 6, H-3516 Miskolc, Hungary; [2]Eszterházy Károly University, Institute of Biology, Department of Botany and Plant Physiology, H-3300 Eger, Leányka u. 6, Hungary; *E-mail: kapidavid97@gmail.com*

Abstract: The paper describes the *Sphagnum* peatmoss collection in the cryptogamic herbarium of the Eszterházy Károly University (EGR), Eger (Hungary), according to its condition in 2012. All specimens of the peatmosses were documented by digital photographs (2835 specimens), and all data from the labels were entered into MS Excel spreadsheet. 24 % of the specimens were collected in present-day Hungary, the majority of specimens comes mainly from the neighbouring countries, but more distant European lands are represented as well as overseas continents. Hungarian specimens were collected mostly in Vas, Heves, Borsod-Abaúj-Zemplén, Veszprém and Szabolcs-Szatmár-Bereg counties, areas with relatively more humid climate than the average of Hungary. Most of the herbarium sheets originated between 1950-1990's. Apart from these periods the collection has hardly developed. The most prolific Hungarian collectors were: Ádám Boros, László Vajda, Tamás Pócs, Márton Balogh and Ilona Gelencsér. In addition to this, many specimens were collected by foreigners as: Richard E. Andrus, Antoine M. Cleef, Josef Duda, Adam Stebel, etc. The digital photographs and the database are property of the Department of Botany and Ecology of Eszterházy Károly University.

Keywords: biological collections, *Sphagnum* collection, herbarium digitization, natural history collections

INTRODUCTION

The herbarium of the Eszterházy Károly University (Index Herbariorum acronym: EGR, Index Herbariorum 2020) is the second largest herbarium in Hungary (Takács *et al.* 2014), consisting of two main parts. The cryptogam collection, which is one of the largest in Central Europe (200000 mosses and liverworts, and 8000 lichens) is widely known among bryologists. It has gained an international reputation for the collections of Tamás Pócs and his colleagues, who studied and collected bryophytes all over the tropics. This collection comprises mainly lichens from Europe and Africa, but the main part are the bryophytes collected predominantly in the tropical regions of Africa, India, Indonesia, Vietnam, Papua New Guinea, Australia, Fiji, Cuba, Peru and Venezuela (Index Herbariorum 2020). The vascular collection is quite small in number, but not negligible (Sass-Gyarmati and Vojtkó 2010; Pénzesné Kónya *et al.* 2013). It is mainly valued for the age of its specimens, dating back to the early 19th century. It stores the collections of significant Hungarian botanists of that time, providing information about their trips and interests in certain regions or taxa. The vascular collection includes vouchers collected primarily in the territory of present-day Hungary and the neighboring countries (mainly in the Carpathian Basin), but there are specimens from the Alps, Silesia, the Balkan, and from the coastal regions of the Adriatic Sea as well. Only the vascular collection has been digitized up to now (E. Vojtkó *et al.* 2014). Therefore, the digitization and analysis of the cryptogamic herbarium is still an actual challenge. We started this work in 2016 and our preliminary results were presented (Sass-Gyarmati *et al.* 2017). Our aim was to create a database of the *Sphagnum* species belonging to the cryptogamic collection of EGR, including all the main attributes of each herbarium specimen, accompanied by digital photos, which may be useful for floristic research. Currently, some other Hungarian herbaria are being processed similarly, e.g. BP (Hungarian Natural History Museum) (Papp and Rajczy 1998), DE (Szarvas *et al.* 2010).

MATERIAL AND METHODS

The database comprises attributes of the material collected until 2012. Methods of digitization and database building process mostly followed Molnár V. *et al.* (2012) and Takács *et al.* (2014). The nomenclature of the species follows Michaelis (2019). Firstly, digital photographs were taken from each sheet, presenting the specimen and the label. The digital photos were taken using ʻ.jpg extension, and their average size is 2–2.5 MB. The information on the labels was entered into Ms Excel spreadsheet, where rows correspond to individual records and columns represent attributes of the collected specimens. The following attributes were recorded: (1) taxon name on the label, (2) collector, (3) reviewer, (4) locality, (5) altitude of locality, (6) year of gathering, (7) month of gathering, (8) day of gathering, and (9) filename of digital photo. One record of the database represents specimen(s) of the same taxon collected from the same locality at the same time on one sheet. If a herbarium sheet contains specimens either of different taxa or from a different locality or time, they are treated as separate records. Thus, the number of records (= rows in the database) is not equal to the number of herbarium sheets (= number of digital photos). When more than one settlement is given on the label, and the administrative affiliation of the locality is therefore ambiguous, the first settlement is provided as the location of gathering (e.g. in the case of " In picetis uliginosis *inter pag. Rábatótfalu et Apátistvánfalva*;", the administrative affiliation in the database is *Rábatótfalu*).

If collecting dates were given as intervals (very often in case of foreign collections) we record the earlier one. Data of *Sphagnum* database are summarized in an electronic appendix http://biologia.uni-eger.hu/en/biology/egr/database including: catalogue number, taxon name, collector, settlement, date of gathering and file name of the documentary photograph). Further data can be required from the curator of the herbarium.

RESULTS AND DISCUSSION

Geographical coverage

The database contains 2835 records altogether, and only 24% of the records (624 records) originates from Hungary, the rest comes from the neighboring and the overseas countries (USA: 17%, Poland: 8 %, Columbia: 8%, Romania: 6%, etc.) (*Figure 1*). We could not identify the country of origin in 368 further records (13%), though it is more than probable that these specimens were collected outside Hungary.

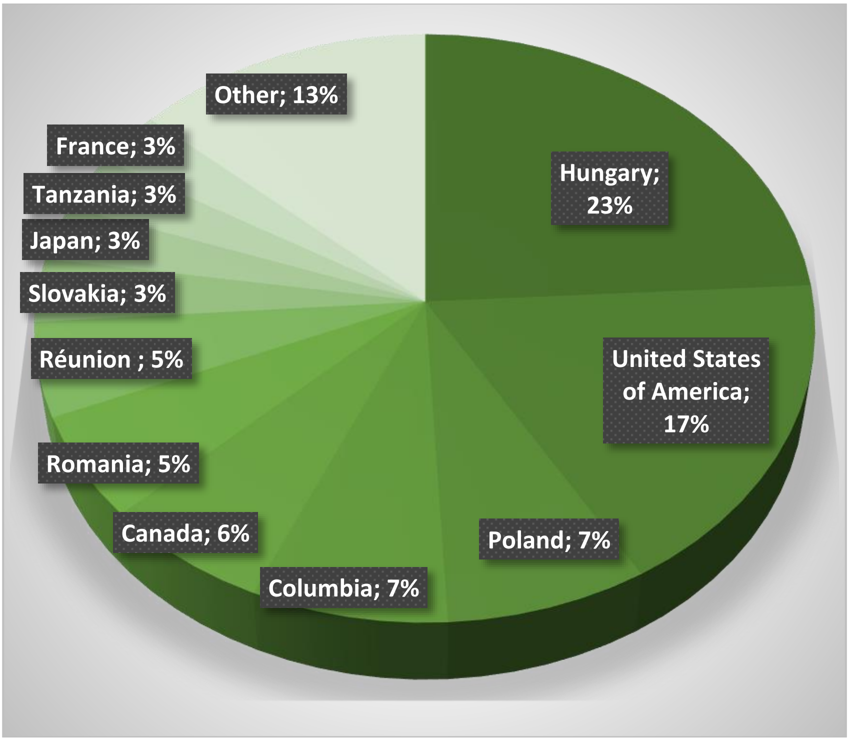


Figure 1. The number of collected specimens per countries.

Our analyses reveals that *Sphagnum* specimens collected in present-day Hungary (624 records) originated from only a smaller part of the country. Some counties are relatively overrepresented due to the special interests of the most prolific collectors of the herbarium and also probably due to the special researches conducted in peaty area and last but not least due to the climatic conditions.

The *Sphagnum* specimens of EGR under study were collected between 1850 and 2012. The intensity of collecting was very uneven, as the vast majority of specimens were obtained between 1950-1990 (*Figure 3*).

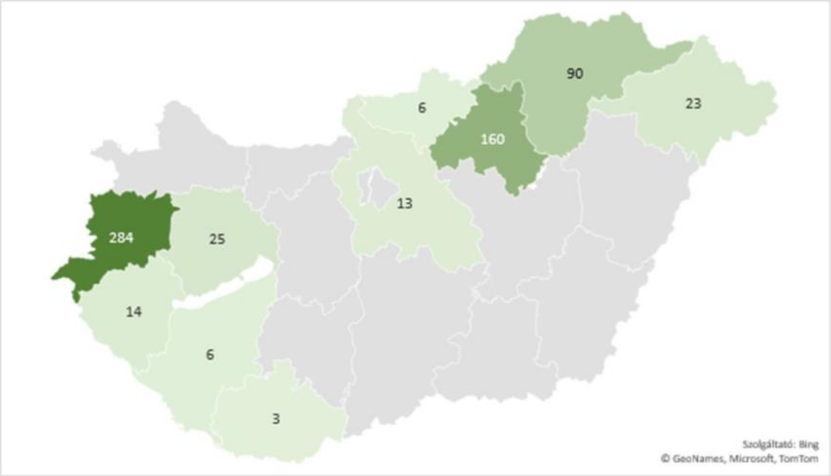


Figure 2. The number of collected specimens per counties in Hungary.

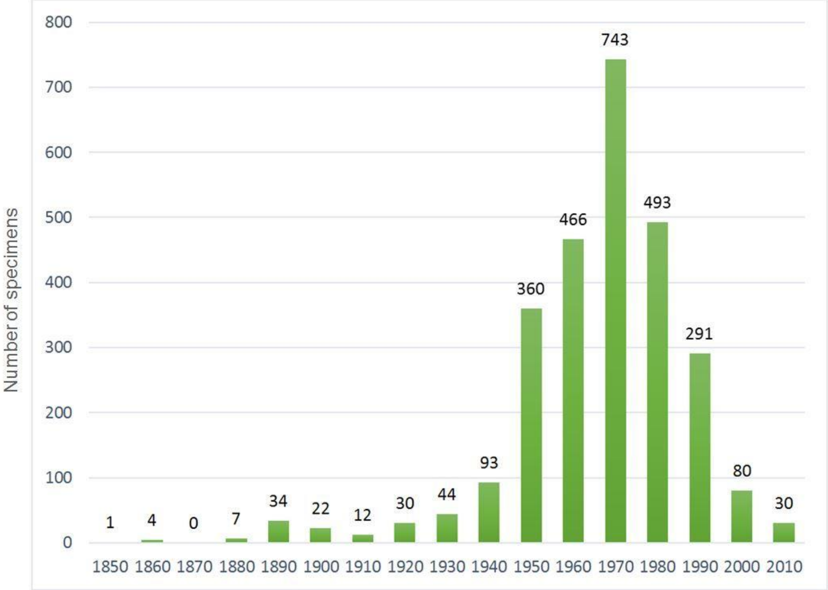
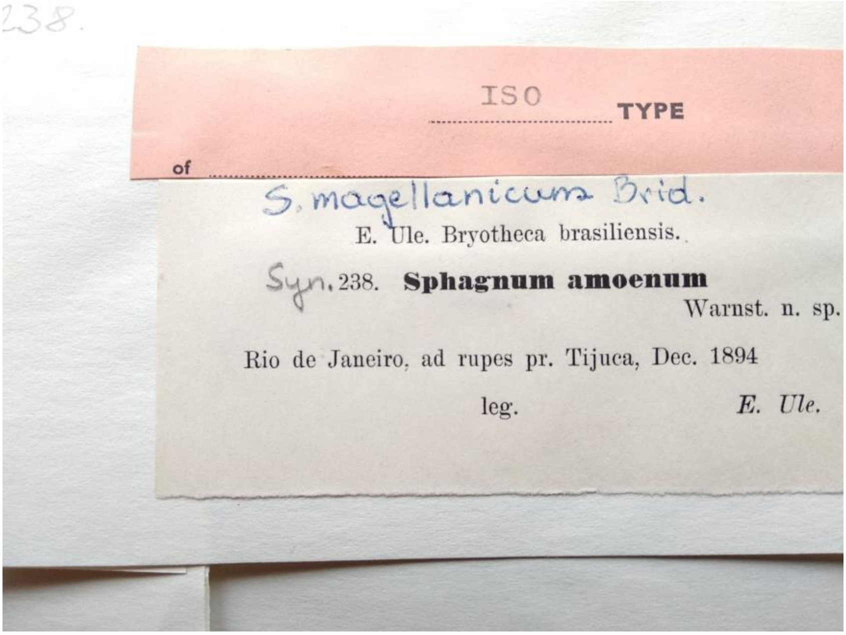


Figure 3. The number of collected specimens per decades.

Representations of the *Sphagnum* species and floristically significant records

More than 130 species, subspecies and varieties occur in the *Sphagnum* genus worldwide (Michaelis 2019). Some of the commonest species which occurs in the herbarium (EGR) are: *Sphagnum capillifolium* (Ehrh.) Hedw., *Sphagnum compactum* DC. ex. Lam. & DC., *Sphagnum cuspidatum* Ehrh. ex. Hoffm., *Sphagnum fimbriatum* Wilson and *Sphagnum girgensohnii* Russow. The two oldest *Sphagnum* specimens were collected by Weselsky in 1852, from Königgrätz and by J. E. Zetterstedt in June 23, 1860 from Sweden. Apart from the exsiccata specimens of *Sphagnotheca Boreali-americana* (Ed. R. E. Andrus and D.H. Vitt) we preserve the isotype of *Sphagnum amoenum* Warnst. (Bryotheca brasiliensis) and an isotype of *Sphagnum squarrosiforme* Dixon and Sherrin (Bryophytorum typorum exsiccata) in our collection (*Figure.4, 5*).



238.

ISO TYPE

of S. magellanicum Brid.

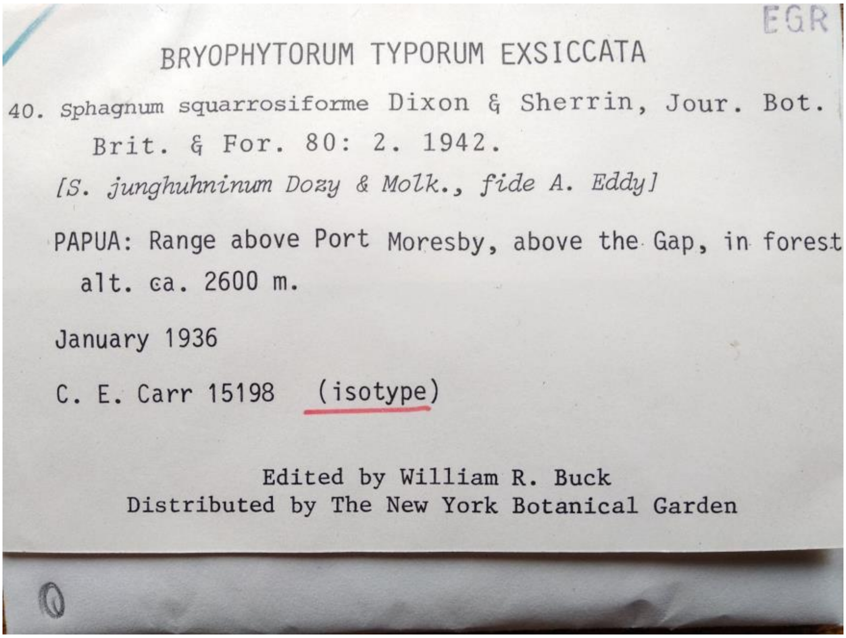
E. Ule. Bryotheca brasiliensis.

Syn. 238. **Sphagnum amoenum**
Warnst. n. sp.

Rio de Janeiro, ad rupes pr. Tijuca, Dec. 1894

leg. *E. Ule.*

Figure 4. Isotype of *Sphagnum amoenum* Warnst. (foto: A. Sass-Gy.)



EGR

BRYOPHYTORUM TYPORUM EXSICCATA

40. Sphagnum squarrosiforme Dixon & Sherrin, Jour. Bot. Brit. & For. 80: 2. 1942.

[S. junghuhninum Dozy & Molk., fide A. Eddy]

PAPUA: Range above Port Moresby, above the Gap, in forest alt. ca. 2600 m.

January 1936

C. E. Carr 15198 (isotype)

Edited by William R. Buck
Distributed by The New York Botanical Garden

Q

Figure 5. Isotype of *Sphagnum squarrosiforme* Dixon & Sherrin. (foto: A. Sass-Gy.)

CONCLUSIONS

In the *Sphagnum* collection, the specimens from Hungary are not separated from the foreign specimens, but can be found in a common storage cabinet. Only 24 % of the *Sphagnum* specimens comes from Hungary, and the other specimens comes as exchange or gift material to the herbarium through foreign researchers or collections made by Botany Department members (Madagascar 1994, 1996 and Réunion 1996). The greatest significance of the completed database is that the *Sphagnum* collection of the Herbarium is now electronically searchable, which facilitates the research work and more efficient management of the collection. Based on the database, which is also available online, the species, country, collector name and year of the *Sphagnum* specimens are now searchable. For more detailed data contact of the herbarium curator is required, who can assign additional data and borrow the material if necessary. Domestic and foreign herbariums can also access the data, which can help with loaning between herbaria and provide information on the size and diversity of the herbarium collection. Based on the results, we can get an idea of the spatial distribution of *Sphagnum* species both in Hungary and abroad. As *Sphagnum* species are the most important habitat for peat moss bogs, this provides feedback on their location or their actually existence. Subsequent research in the area may point to pollution factors if peat mosses are no longer found in the area.

Acknowledgement – The authors would like to express their gratitude to Professor Tamás Pócs and dr. Péter Szűcs for their useful comments. The second author's research was supported by the grant EFOP-3.6.1-16-2016-00001 ("Complex improvement of research capacities and services at Eszterházy Károly University").

REFERENCES

E. Vojtkó, A., Takács, A., Molnár, V.A. & Vojtkó, A. (2014). Herbarium database of the vascular collection of Eszterházy Károly College (EGR). *Kitaibelia* **19**: 339–348.

Index Herbariorum (2020). http://sweetgum.nybg.org/science/ih/ (accessed: June 10, 2020)

Michaelis, D. (2019). *The Sphagnum species of the World.* Schweizerbart Science Publishers. Bibliotheca Botanica, Vol. 162., 435 pp.

Molnár, V.A., Takács, A., Horváth, O., E. Vojtkó, A., Király, G., Sonkoly, J., Sulyok, J. & Sramkó, G. (2012). Herbarium Database of Hungarian Orchids I. Methodology, dataset, historical aspects and taxa. *Biologia* **67**: 79–86. https://doi.org/10.2478/s11756-011-0144-9

Papp, B. & Rajczy, M. (1998). Moharitkaságok Magyarországon – előkészületek az új Vörös Lista összeállításához (Rare bryophytes in Hungary – preliminary efforts on compiling a new Red List). *Kitaibelia* **3**(2): 355–356.

Pénzesné Kónya, E., Orbán, S., Pócs, T. & Sass-Gyarmati, A. (2013). Az Eszterházy Károly Főiskola megújult herbáriuma. *Acta Academiae Agriensis Sectio Biologiae* **40**: 5–9.

Sass-Gyarmati, A. & Vojtkó, A. (2010). The herbarium of the Botanical Department in Károly Eszterházy College, Eger. *Acta Biologica Plantarum Agriensis* **1**: 7–13.

Sass-Gyarmati, A., Pénzesné Kónya, E., Kapi, D., Stefancsik, G. & Kelecsényi, F. (2017). Bryophyte collection of Eszterházy Károly University (EGR): The digital database of peat moss (*Sphagnum*) species. *Acta Biologica Plantarum Agriensis* **5**: 61. https://doi.org/10.21406/abpa.2017.5.1.61

Szarvas, V.E., Takács, A. & Nagy, M. (2010). A Debreceni Egyetem Herbáriumának mohagyűjteménye (The bryophyte collection of the University of Debrecen). *Kitaibelia* **15**(1–2): 65–72.

Takács, A., Nagy, T., Fekete, R., Lovas-Kiss, Á., Ljubka, T., Löki, V., Lisztes-Szabó, Zs. & Molnár, V.A. (2014). A Debreceni Egyetem Herbáriuma (DE) I.: A "Soó Rezső Herbárium" (The herbarium of Debrecen University (DE) I. The "Rezső Soó Herbarium"). *Kitaibelia* **19**: 142–155.

(submitted: 25.05.2020, accepted: 10.06.2020)

Acta Biologica Plantarum Agriensis **8**(1): 29–39 (2020)
ISSN 2061-6716 (Print), 2063-6725 (Online)
http://abpa.ektf.hu/

DOI:10.21406/abpa.2020.8.1.29
Research article

BRYOPHYTE RECORDS FROM THE UDZUNGWA SCARP NATURE FOREST RESERVE, TANZANIA

Tamás Pócs

Institute of Biology, Eszterházy Károly University, Eger, Pf. 43, Hungary, H-3301;
E-mail: colura@upcmail.hu

Abstract: Christian Frimodt-Møller in 1997 collected an interesting bryophyte material in the Tanzanian Udzungwa Mountains, part of the Crystalline Eastern Arc. The material came from the Udzungwa Scarp Forest Reserve, an outsider satellite of the Udzungwa Mountains National Park, which is a bryologically completely unknown part of the south-western end of the mountains. The study site is located in a lower montane rainforest at an elevation of 1500–1700 m. These records fill a gap between the much better known north-eastern part of the Udzungwe Mountains and the Southern Highlands of Tanzania. Altogether 25 species of liverwort and five mosses were identified that in greater part are new to these mountains.

Keywords: Crystalline Eastern Arc, endemics, East Africa, Udzungwa Mountains

INTRODUCTION

The Udzungwa Mountains form the south-western end of the chain of the Crystalline Eastern Arc mountains extending from southern Kenya throughout eastern Tanzania and ending at the Makumbako Gap, which separates it from the Kipengere and Livingstone ranges in the Southern Highland (*Figure 1*). This mountain range has very old origin (Precambrian granitic and gneissic bedrocks) and continually favourable climatic conditions since its uplift 30 million years ago. It enjoys adequate rainfall throughout the year on the eastern, seaside slopes which are exposed to the rain carrying winds from the Indian Ocean. Therefore the Eastern Arc mountains on their windward side, ridges and high plateaux carry very luxurious rainforest vegetation. As a consequence, these rainforests are very rich both in plant and animal endemics and in elements common with Madagascar and other East African Indian Ocean islands (Pócs 1975, 1998, 1999, Burgess *et al.* 2007). This is in contrast to the much younger volcanic mountains located in drier areas located farther inland from the ocean. At the same time their lee side gets much less rainfall and, accordingly, it is covered by more xerotolerant vegetation, including dry forests and miombo woodland. This asymmetry can be observed in almost all mountains of the Eastern Arc (Pócs 1977). By the time of climatic fluctuations, for example during the Pleistocene, the ratio between the two types of vegetation could drastically change, but the higher slopes and summits were permanently covered by rainforests, at least in some core areas, like the Eastern Arc mountains (Hamilton 1982). The fluctuation of altitudinal belts resulted in repeated isolation of montane forest patches, promoting evolution of endemic species.

By the kindness of Christian Frimodt-Møller, that time botanist of University of Copenhagen (later becoming a tour operator and managing director of a travel agency in Colombia), I received a small collection of bryophytes for identification which he made in December 1997 in the Udzungwa Scarp Forest Reserve at the south-western end of the Crystalline Eastern Arc Mountains in Tanzania (*Figure 2*). The forest reserve, although not part of the Udzungwa Mountains National Park, enjoys special protection, as its outsider satellite, with a forest covering an area of 20,220 ha. This remote place hitherto was bryologically unknown, therefore a greatest part of the records are new to the mountains. Frimodt-Møller collected the bryophytes at one locality. All specimens were gathered at the site bearing according to the collector the following locality data: UDZUNGWA Mountains in Iringa District. Udzungwa Scarp Nature Forest Reserve. On steep E slopes at 1500-1700 m alt. 8°21′S, 35°58′E. Rainforest dominated by *Parinari excelsa, Syzygium* sp., *Myryanthus holstii* and *Ficalhoa laurifolia.*

The specimens were collected on various substrates, as indicated in the list below. The collecting numbers were given by Frimodt-Møller, the letters after the numbers indicate species segregated from a single gathering. One sample of each number went to the moss herbarium of Copenhagen University (C), while a fully identified set is deposited in the Herbarium of Eszterházy Károly University, Eger, (EGR). The nomenclature in most cases follows Wigginton (2018) and O'Shea (2006), apart from the names used in more recent revisions. Global distributional data are given for each species and for some species names some heterotypic and homotypic synonyms are provided.

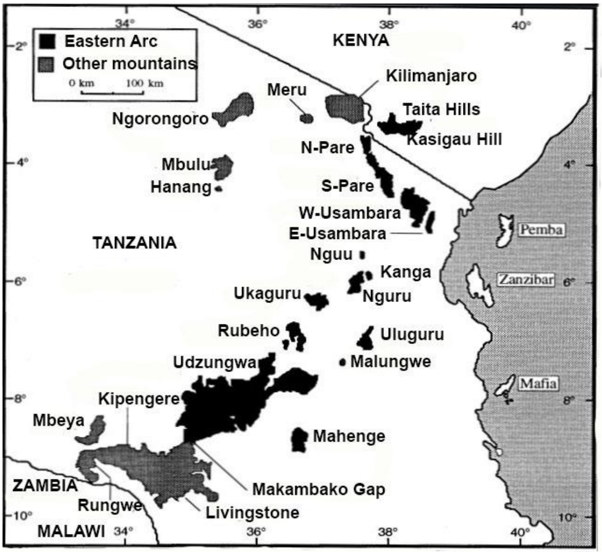


Figure 1. The Crystalline Eastern Arc mountain range from southern Kenya to southern Tanzania (Black. Other, mostly volcanic mountains are marked by grey color. After Burgess *et al.* 2007).

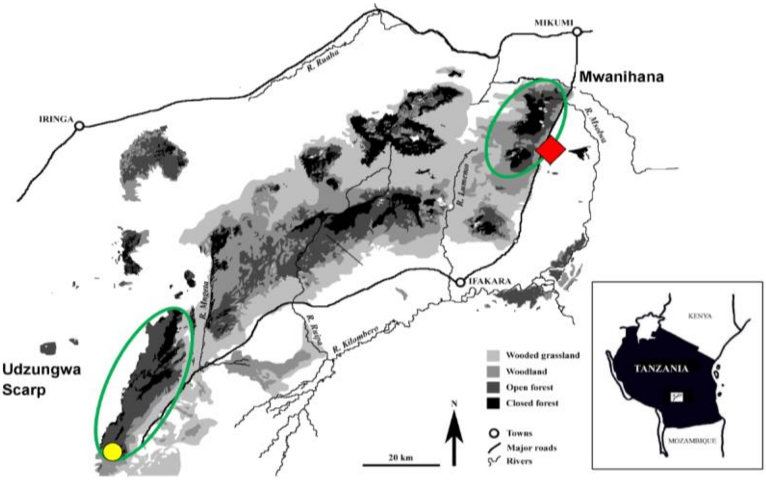


Figure 2. The Uzungwe Mountains. The bryophytes of northeastern part (Mwanihana Forest reserve within the National Park area, marked by diamond) are relatively well known, while the Udzungwe Scarp Forest Reserve at the southwestern end (area marked by light dot) was bryologically unknown (Map after Enghoff 2016).

RESULTS

List of species

Liverworts

Acrobolbus limbatus (Steph.) Briscoe & J.J.Engel (*Figure 5*) – On decaying wood. TZ634/A. Syn.: *Marsupidium limbatum* (Steph.) Grolle, *Tylimanthus limbatus* Steph. A southern temperate Afro-American disjunct (Gradstein *et al.* 1984), very rare in Africa, hitherto known from the Cape, Malawi, Tanzania (Kilimanjaro), and from the Mascarene Islands: Mauritius and Réunion (Wigginton 2018).

Bazzania decrescens (Lehm. & Lindenb.) Trev. – On decaying log. TZ710/A. A widespread Afromontane species, closely related to the Australasian *Bazzania adnexa* (Lehm. & Lindenb.) Trevis (Jones 1975).

Ceratolejeunea cornuta (Lindenb.) Steph. – On twigs. TZ649/D. A widespread tropical Afro-American or pantropical species, the African representatives used to be named as *Ceratolejeunea calabariensis* Steph. (Pócs 2011).

Cheilolejeunea trapezia (Nees) Kachroo & R.M.Schust. – On twigs. TZ636/B. A widespread palaeotropical species, common in Africa.

Cololejeunea malanjae Steph. (*Figure 6*) – Epiphyllous. TZ708/L. A southeast tropical African species distributed from the Democratic Republic of Congo and Kenya to Malawi.

Cololejeunea microscopica (Taylor) A.Evans – Epiphyllous. TZ708/P. A very variable pantropical species penetrating in the oceanic parts of the temperate regions.

Cololejeunea tanzaniae Pócs (*Figure 7*) – Epiphyllous. TZ708/D – An East African endemic distributed from Kenya and Tanzania to the Indian Ocean islands (Pócs 1980).

Cololejeunea zenkeri Steph. Syn.: *Cololejeunea duvigneaudii* E.W.Jones – Epiphyllous. TZ708/K. A very widespread epiphyllous species in tropical Africa.

Cryptolophocolea martiana (Sw.) L.Söderstr. & Váňa – On decaying log. TZ710/C. A widespread tropical Afro-American species (see map in Gradstein *et al.* 1984).

Diplasiolejeunea zakiae Tixier (*Figure 8*) – Epiphyllous. TZ708/H. A rare endemic species of East Africa known only from Kenya, Tanzania and Madagascar (Tixier 1977, Bizot and Pócs 1974, 1982, Pócs and Geissler 2002). In Tanzania it was previously known only from the Uluguru Mountains.

Drepanolejeunea cultrella (Mitt.) Steph. – Epiphyllous and among mosses. TZ634/B, TZ708/G & J, TZ636/A. A very widespread tropical African species.

Drepanolejeunea physifolia (Gottsche) Pearson – Epiphyllous. TZ708/C. A widespread tropical African species closely related or identical to the palaeotropical *Drepanolejeunea vesiculosa* (Mitt.) Steph.

Drepanolejeunea pocsii Grolle (*Figure 9*) – Epiphyllous. TZ708/M. An East African endemic known from Kenya, Tanzania, Malawi, Zimbabwe and from the Indian Ocean islands (Grolle 1976, Wigginton 2018).

Frullania serrata Gottsche – On bark. TZ713. A widespread palaeotropical species, common in tropical African mountains above 900 m elevation (Vanden Berghen 1976).

Lejeunea acuta Mitt. – Epiphyllous and ramicolous. TZ634/A, TZ708/A. Widespread in tropical African mountains (Vanden Berghen 1961, under *Hygrolejeunea acuta*).

Lejeunea flava (Sw.) Nees – Epiphyllous. TZ636/B. A common pantropical species.

Lejeunea grossecristata (Steph.) E.W.Jones – Epiphyllous. TZ708/B. A taxon of dubious status belonging to the *Lejeunea flava* complex, namely to *Lejeunea tabularis* (Spreng.) Gottsche, Lindenb. & Nees (Jones 1979, Pócs 1993). Widespread in tropical Africa.

Marchantia pappeana Lehm. subsp. ***pappeana*** (*Figure 10*) – On ground. TZ642, TZ716. The type subspecies is widespread in tropical Africa, while subsp. *robusta* (Steph.) Bischl. occurs in India and Sri Lanka (Perold 1999).

Metzgeria consanguinea Schiffn. – On twigs. TZ636/E. A widespread pantropical species occurring in all tropical African mountains.

Metzgeria leptoneura Spruce – On mosses. TZ649/C. A subcosmopolitan species common in the tropics and penetrating in the subtropics and the oceanic parts of the temperate zones. Widespread in African mountains.

Microlejeunea africana Steph. – Epiphyllous. TZ708/F, TZ634/C. A tropical African species close to *Microlejeunea bullata* (Taylor) Steph. and to the boreal temperate *Microlejeunea ulicina* (Taylor) A.Evans (Jones 1969, Wigginton and Van de Kerckhove 2004).

Plagiochila fusifera Taylor – Epiphyllous. TZ636/C. A widespread tropical African species.

Plagiochila kiaeri Gottsche – On twigs. TZ636/D. A widespread tropical African species.

Plagiochila pectinata (Willd.) Lindenb. – Epiphyllous. TZ708/O. A relatively rare tropical montane species occurring mostly on West and East African islands and in the Cristalline Eastern Arc mountains (Wigginton 2018).

Radula madagascariensis Gottsche – Epiphyllous. TZ708/E. A palaeotropic species, widespread from East Africa to Indonesia and the Philippines. In Africa it is known only from Kenya, Tanzania, Malawi, the Comoro Islands, Madagascar, Réunion and Mauritius (Yamada 1979, Pócs 1992, Wigginton 2018).

Mosses

Leucoloma subsecundifolium Broth. (*Figure 4*) Syn: *L. volkensii* Broth. – On bark. TZ648. A South-East African endemic, known from Kenya, Tanzania, Malawi and the Comoro Islands (La Farge 2002b). A typical member of mossy montane cloud forests.

Leucoloma zuluense Broth. & Bryhn (*Figure 3*)– On twigs. TZ635. A South-East African endemic, known from Kenya, Tanzania, Zimbabwe and South Africa. Its var. *ovatum* La Farge occurs also on the Comoro Islands (La Farge 2002a).

Plagiomnium rhynchophorum (Hook.) T.J.Kop. – On ground. TZ715. A palaeotropic species occurring scattered all over tropical Africa and Asia. (O'Shea 2006, Koponen *et al.* 2019)

Porotrichum elongatum (Welw. & Duby) Gepp – On twigs. TZ643/A. A very widespread epiphyte all over tropical Africa (O'Shea 2006).

Pyrrhobryum spiniforme (Hedw.) Mitt. – On decaying log. TZ710/B. A common pantropical moss.

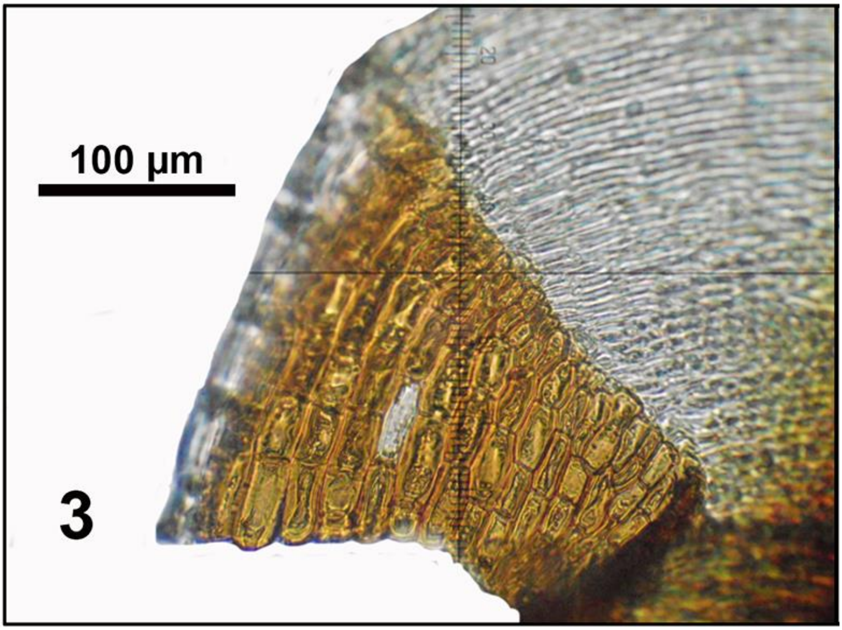


Figure 3. Alar region of *Leucoloma zuluense* Broth. & Bryhn (from *Frimodt-Møller TZ635*).

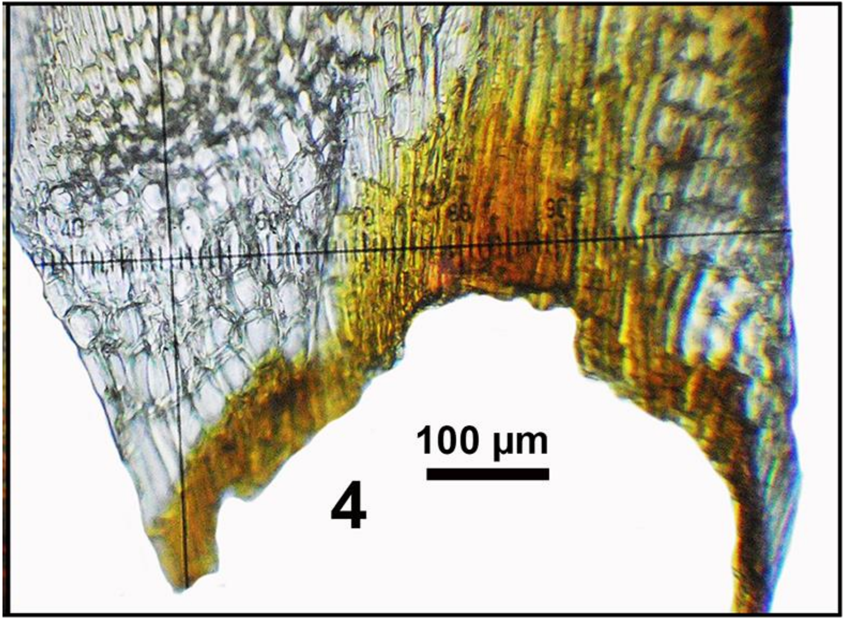


Figure 4. Alar region of *Leucoloma subsecundifolium* Broth. (from *Frimodt-Møller TZ648*).

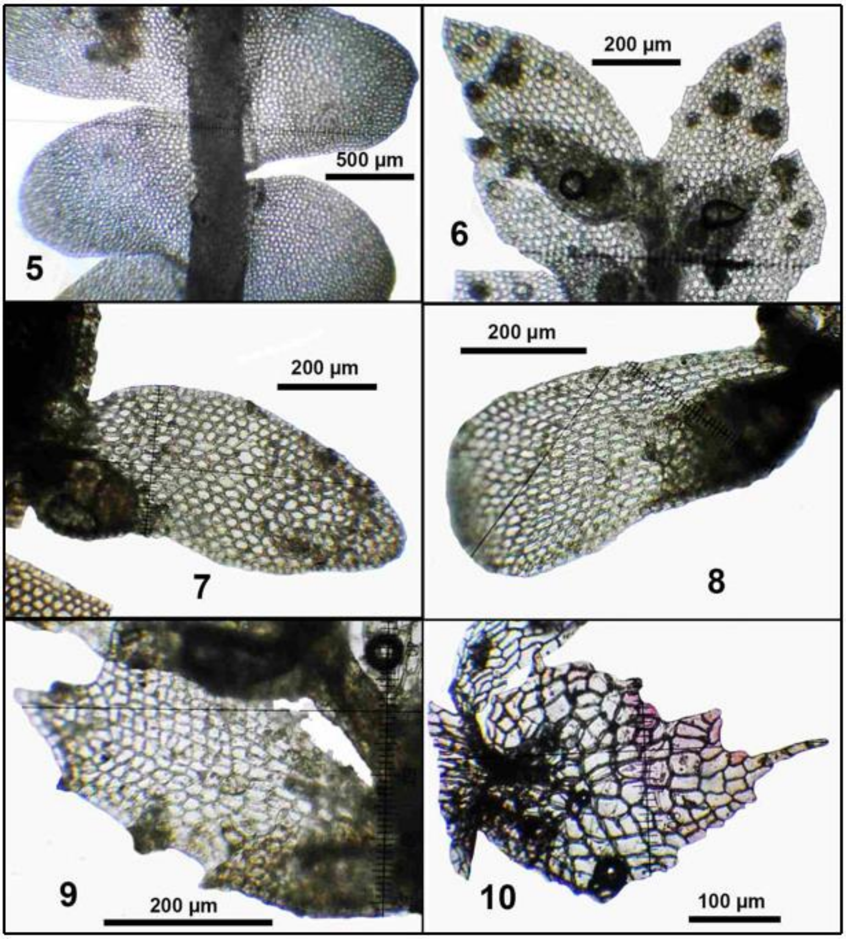


Figure 5. *Acrobolbus limbatus* (Steph.) Briscoe & J.J.Engel, habit, ventral view (from *Frimodt-Møller TZ634/A*). **Figure 6.** *Cololejeunea malanjae* Steph., habit, ventral view (from *Frimodt-Møller TZ708/L*). **Figure 7.** *Cololejeunea tanzaniae* Pócs, leaf, ventral view (from *Frimodt-Møller TZ708/D*). **Figure 8.** *Diplasiolejeunea zakiae* Tixier, leaf, ventral view (from *Frimodt-Møller TZ708/H).* **Figure 9.** *Drepanolejeunea pocsii* Grolle, leaf, ventral view (from *Frimodt-Møller TZ708/M*). **Figure 10.** *Marchantia pappeana* Lehm. Appendage of a ventral scale (from *Frimodt-Møller TZ642*).

DISCUSSION

The low number of species collected does not permit to draw extensive conclusions concerning the flora element composition of the study area, but even so, the relatively high proportion of endemic and subendemic species is conspicuous. Such elements are represented by *Cololejeunea malanjae, C. tanzaniae, Diplasiolejeunea zakiae, Drepanolejeunea pocsii, Leucoloma subsecundifolium* and *L. zuluense,* which constitute 20% of the total number of species. This high level of endemicity is even more striking among the animals. Among mammals, like the Udzungwa red colobus (*Cercocebus galeritus*), Kipunji (*Rungwecebus kipunji*) which is among the world's 25 most endangered mammals, the Abbot's duiker (*Cephalopuis spadix*); among birds the Udzungwa forest partridge (*Xenoperdix udzungwensis*) and last but not least the highly endangered Udzungwa Scarp tree frog (*Nectophrynoides wendyae*) which occurs only in one valley of the investigated area (Wikipedia 2019). Of course, the invertebrate fauna contains even higher ratio of endemics, probably thousands of still undescribed species (Enghoff 2016). Regrettably, the forest areas in the Eastern Arc mountains continuously shrivel, partly due to human activities, partly due to the global warming or the combination of the two. This is why the effective protection of these last primary forest covered blocks of the Crystalline Eastern Arc, and especially of the Udzungwe Scarp Forest Reserve would be essential (Seki *et al.* 2011, Rovero *et al.* 2005, 2010). Since 2008 the Tanzania Forest Conservation Group, cooperating closely with the Trento Museum of Natural History, Italy, have worked with communities around the reserve to promote awareness of the importance of conservation values.

Acknowledgement – I wish to express my gratitude towards Dr Christian Frimodt-Møller for putting his bryophyte collection at my disposal. Thanks are also due for the information and guidance provided by A.J. Isango and David C. Moyer during my last stay in the Udzungwa Mountains in May 1999. Finally I am grateful to my reviewers, Prof. Ryszard Ochyra and Dr. Andrea Sass-Gyarmati, for their useful corrections and advises.

REFERENCES

Bizot, M. & Pócs, T. (1974). East African bryophytes I. *Acta Academiae Paedagogicae Agriensis* **12**: 383–449.

Bizot, M. & Pócs, T. (1982). East African bryophytes V. *Acta Botanica Academiae Scientiarum Hungaricae* **28**: 15–64.

Burgess, N.D., Butynski, T.M., Cordeiro, N.J., Doggart, N.H., Fjeldsa, J., Howell, K.M., Kilahama, F.B., Loader, S.P., Lovett, J.C., Mbilinyi, B., Menegon, M. Moyer, D.C.,

NASHANDA, E., PERKIN, A., ROVERO, F., STANLEY, W.T. & STUART, S.N. (2007). The biological importance of the Eastern Arc Mountains of Tanzania and Kenya. *Biological Conservation* **134**: 209–231. https://doi.org/10.1016/j.biocon.2006.08.015

ENGHOFF, H. (2016). A mountain of millipedes IV: Species of *Prionopetalum* Attems, 1909, from the Udzungwa Mountains, Tanzania. With notes on "*P.*" *fasciatum* (Attems, 1896) and a revised species key (Diplopoda, Spirostreptida, Odontopygidae). *European Journal of Taxonomy* **215**: 1–23. https://doi.org/10.5852/ejt.2016.215

GRADSTEIN, S.R., PÓCS, T. & VÁŇA, J. (1984, „1983"). Disjunct hepaticae in tropical America and Africa. *Acta Botanica Hungarica* **29**: 127–171.

GROLLE, R. (1976). *Drepanolejeunea* subgen. *Kolpolejeunea* - eine neue Untergattung aus der Paläotropis. *Journal of the Hattori Botanical Laboratory* **40**: 191–216.

HAMILTON, A.C. (1982). *Environmental history of East Africa. A study of the Quaternary*. Academy Press, London & New York, 328 pp.

JONES, E.W. (1969). African Hepatics XXI. *Microlejeunea, Chaetolejeunea* and *Pleurolejeunea*. *Transactions of the British Bryological Society* **5**: 775–789.

JONES, E.W. (1975). African Hepatics XXVII. *Bazzania*. *Journal of Bryology* **8**: 299–316.

JONES, E.W. (1979). African Hepatics XXXI. Rare or little-known Lejeuneaceae and extensions of range. *Journal of Bryology* **10**: 387–400.

KOPONEN, T., NGUYEN, TH-L., LUONG, TH-T. & HUTTUNEN, S. (2019). Revision and checklist of the moss families Bartramiaceae and Mniaceae in Vietnam. *Hattoria* **10**: 69–107. https://doi.org/10.18968/hattoria.10.0_69

LA FARGE, C. (2002a). *Leucoloma* II: A revision of *Leucoloma* series *Holomitrioidea* (Dicranaceae). *Bryologist* **105**: 591–605. https://doi.org/10.1639/0007-2745(2002)105[0591:LIAROL]2.0.CO;2

LA FARGE, C. (2002b). *Leucoloma* III: A species synopsis: typification, synonymy and excluded names. *Bryologist* **105**: 606–624. https://doi.org/10.1639/0007-2745(2002)105[0606:LIASST]2.0.CO;2

O'SHEA, B.J. (2006). Checklist of the mosses of sub-Saharan Africa (version 5, 12/6). *Tropical Bryology Research Reports* **6**: 1–252.

PEROLD, S.M. (1999). *Hepatophyta. Part 1. Marchantiales. Fasc. 1. Marchantiidae*, In: LEISTNER, O.A. (ed.): *Flora of Southern Africa*. Pretoria: National Botanical Institute, 252 pp.

PÓCS, T. (1975). Affinities between the bryoflora of East Africa and Madagascar. In: MIÈGE, J. & STORK, A. (eds.): Origines des flores africaines et malgaches. Nature, spéciation. Comptes-rendues de la VIIIe réunion de l'AETFAT, 1. *Boissiera* **24a**: 125–128.

PÓCS, T. (1977, "1976"). Bioclimatic studies in the Uluguru Mountains (Tanzania, East Africa) II. Correlations between orography, climate and vegetation. *Acta Botanica Academiae Scientiarum Hungaricae* **22**: 163–183.

PÓCS, T. (1980). New or little known epiphyllous liverworts, II. Three new *Cololejeunea* from East Africa. *Journal of the Hattori Botanical Laboratory* **48**: 305–320.

PÓCS, T. (1992) Correlation between the tropical African and Asian bryofloras. II. In: KOPONEN T. & HYVÖNEN, J. (eds.): Proceedings of the Congress of East Asiatic Bryology, Helsinki, August 12-19, 1990. *Bryobrothera* **1**: 35–47.

Pócs, T. (1993). Taxonomic results of the BRYOTROP expedition to Zaire and Rwanda 12. Metzgeriaceae, Plagiochilaceae, Lejeuneaceae (the non epiphyllous collections). *Tropical Bryology* **8**: 105–125.

Pócs, T. (1998). Bryophyte diversity along the Eastern Arc. *Journal of East African Natural History* **87**: 75–84.

Pócs, T. (1999). Bryophyte speciation and diversity in the East African mountains. *Bryobrothera* **5**: 237–245.

Pócs, T. (2011). East african bryophytes XXIX. The *Ceratolejeunea* (Lejeuneaceae) species of the Indian Ocean islands. *Polish Botanical Journal* **56**(2): 131–153.

Pócs, T. & Geissler, P. (2002). Chapter 3. The bryophytes collected in the Réserve Spéciale de Manongarivo, Madagascar. In: Gautier, L. & Goodman, S.M. (eds.): Inventaire Floristique et Faunistique de la Réserve Spéciale de Manongarivo (NW Madagascar). *Boissiera* **59**: 41–76.

Rovero, F. & Menegon, M. (2005). *Uzungwa Scarp, an outstanding Eastern Arc forest: biological value and urgent need for protection.* In: Keyuu, J., Mduma, S. & Labora, A. (eds.): *Proceedings of the 5th Tanzania Wildlife Research Institute Scientific Conference*, TAWIRI, Arusha, pp. 183–187.

Rovero, F., Mtui, A., Kitegile, A., Nielsen, M. & Jones T. (2010). *Uzungwa Scarp Forest Reserve in crisis. An urgent call to protect one of Tanzania's most important forests.* USFR Report, Dar es Salaam, 15 pp.

Seki, H.A., Bowkett, A.E., Bungard, M.J., Doggart, N. & Menegon, M. (2011). *Current status of three endangered hyper-endemic amphibian species in the Uzungwa Scarp Forest Reserve, Tanzania.* Tanzania Forest Conservation Group and Whitley Wildlife Conservation Trust, Dar es Salaam, 12 pp.

Tixier, P. (1977). Espèces nouvelles malgaches du genre *Diplasiolejeunea* (Spruce) Schiffn. (Hepaticae). *Lindbergia* **4**: 117–125.

Vanden Berghen, C. (1961). Hépatiques récoltées par Dr J.-J. Symoens dans la région péri-tanganyikaise. *Bulletin de la Société Royale de Botanique de Belgique* **93**: 55–74.

Vanden Berghen, C. (1976). Frullaniaceae (Hépaticae) Africanae. *Bulletin du Jardin Botanique National de Belgique* **46**: 1–220.

Wigginton, M.J. (2018). Checklist and distribution of the liverworts and hornworts of sub-Saharan Africa, including the East African Islands. *Tropical Bryology Research Reports* **9**: 1–138.

Wigginton, M.J. & Van de Kerckhove, O. (2004). *EW Jones's liverwort and hornwort flora of West Africa.* National Botanic Garden, Meise, 443 pp.

Wikipedia (2019). Udzungwa Mountains. https://en.wikipedia.org/wiki/ Udzungwa Mountains. (Accessed: 28 September 2019)

Yamada, K. (1979). A revision of Asian taxa of *Radula*, Hepaticae. *Journal of the Hattori Botanical Laboratory* **45**: 201–322.

(submitted: 26.04.2020, accepted: 14.05.2020)

Acta Biologica Plantarum Agriensis **8**(1): 40–52 (2020)
ISSN 2061-6716 (Print), 2063-6725 (Online) *Research article*
http://abpa.ektf.hu/
https://doi.org/10.21406/abpa.2020.8.1.40

AURANTIPORUS CROCEUS, A FLAGSHIP SPECIES OF THE EUROPEAN FUNGAL CONSERVATION IS RE-DISCOVERED AFTER HALF CENTURY IN HUNGARY

Attila Koszka*[1] *& Viktor Papp*[2]

[1]Hungarian Mycological Society, Budapest, Hungary; [2]Department of Botany, Szent István University, 1518 Budapest, Hungary;
**E-mail: agaricum@gmail.com*

Abstract: The threatened polypore, *Aurantiporus croceus* has previously been known only from one locality in Hungary, and the basidiome of this species has not been seen in the country since 1972. In this study, a new Hungarian finding of *A. croceus* is reported from an old-growth forest reserve in the Vértes Mts (Central Transdanubia). We present the nrDNA ITS sequence, macro- and microscopical characteristics as well as photographs of the new specimen.

Keywords: Central Europe, *Hapalopilus*, Meruliaceae, phylogeny, Polyporales, *Quercus*

INTRODUCTION

The peculiar polypore species, *Aurantiporus croceus* (Pers.) Murrill (≡ *Hapalopilus croceus* (Pers.) Bondartsev & Singer) is easily recognised in the field by the often large sized, bright orange-red coloured basidiocarps growing predominantly on veteran oak (*Quercus*) trees. *Aurantiporus croceus* is widely distributed in Europe (Ryvarden and Melo 2014) and also reported from the eastern parts of Asia (Dai 2012) and eastern North America (Zhou *et al.* 2016). Despite its widespread distribution and easily recognisable basidiome, *A. croceus* is considered to be a rare species everywhere. As a result of its rarity and its spectacular appearance, it was placed in the spotlight of the European fungal conservation; it is listed amongst the 33 threatened fungi proposed to be included in the Bern Convention (Dahlberg and Croneborg 2003), and red-listed in 12 European countries (listed as CR in eight countries), and included in nine regional Red Data Books of Russia (Dahlberg 2019). Due to the significantly declined

Department of Botany and Plant Physiology

population, it became very rare and scattered, it has recently been assessed to the IUCN's Red List (Dahlberg 2019).

From Hungary, only historical specimens of *A. croceus* were known. These are originated from an old living *Quercus* tree located near Sitke, Western Transdanubia (Igmándy 1968, Szabó 2012). Despite the single locality of this rare polypore, it was not included in the proposed Hungarian Red List (Rimóczi *et al*. 1999). Considering the specific habitat preference and the rarity of *A. croceus* in Hungary, the second author urged to protect this species by law, which was achieved in 2013 (MK 2013). However, despite the greater attention, no new occurrence of this species was observed until 2018. During a mycological survey of Juhdöglő-völgy Forest Reserve (Vértes Mts, Central Transdanubia) in late May, a new location of *A. croceus* was found. In this study the ITS sequence, macro- and microscopical characteristics and photographs of the new Hungarian specimen of *A. croceus* are presented.

MATERIALS AND METHODS

Isolates and morphology

The new Hungarian specimen was deposited in the private herbarium of the authors. We report the macromorphological descriptions based on field notes. Micromorphological data were obtained from the dried specimens, which were observed under a Zeiss Axio Imager.A2 light microscope, equipped with AxioVision Release 4.8.2. software. Measurements were done with a 100× oil immersion objective (1000× magnification). Observations of microscopic features as well as measurements were made on slide preparations stained with Melzer's reagent. Spores were measured by cutting sections from the tubes. The following abbreviations were used in the description of the basidiospores: IKI = Melzer's reagent, IKI– = both inamyloid and indextrinoid, L = mean spore length, W = mean spore width, Q = variation in the L/W ratios, n = number of measured spores.

Molecular study

Primers ITS1F and ITS4 (White *et al*. 1990, Gardes and Bruns 1993) were used to amplify the ITS (internal transcribed spacer) region of the nuclear ribosomal DNA. For the amplification we used the

Phire® Plant Direct PCR Kit (Thermo Scientific, USA) following the manufacturer's recommendations. The PCR (polymerase chain reaction) protocols were set according to Papp and Dima (2017). The quality of PCR products were checked in 2% agarose gels. The amplicons were sequenced commercially at the Biological Research Centre (Szeged, Hungary) with the same primers used in the PCR reactions. The chromatograms were checked, assembled and edited with the CodonCodeAligner 7.0.1 (CodonCode Corporation, Centerville, MA, USA).

The newly generated *Aurantiporus croceus* sequence is deposited in GenBank (Benson *et al*. 2017); the accession numbers are included in *Table 1*. For the phylogenetic analysis, similar sequences were searched from GenBank using the BLASTn search tool (Altschul *et al*. 1990). The ITS region was aligned with PRANK (Löytynoja and Goldman 2005, 2008) as implemented in its graphical interface (PRANKSTER) using default settings. SeaView 4 (Gouy *et al*. 2010) was used to visually inspect and improve the alignment. The nucleotide dataset resulted an alignment length of 697 characters. The dataset was subjected to maximum likelihood (ML) and Bayesian inference (BI) phylogenetic analyses, which were performed in raxmlGUI (Silvestro and Michalak 2012) and MrBayes 3.1.2 (Ronquist and Huelsenbeck 2003), respectively. ML analysis was done using 1000 rapid bootstrap searches. For the nucleotide partition the GTRGAMMA substitution model, while for the indel partition the RAxML default set for binary characters were applied. In the BI analysis the GTR + G model of evolution for the nucleotide partition, and the two-parameter Markov model (Mk2 Lewis) for the indel partition were applied. The BI settings were: four Markov chain Monte Carlo (MCMC) over 5 million generations, sampling every 1000th generation, two independent runs, and burn-in of 20% (the first 1000 trees were discarded). Post burn-in trees were used to compute a 50% majority rule consensus phylogram. Phylogenetic trees from both ML and BI analyses resulted in congruent topologies. The best scoring tree from the ML analysis was edited with MEGA6 (Tamura *et al*. 2013) and presented in *Figure 1*.

Table 1. Details of the specimens comprised in this study. Species, herbarium voucher numbers, country, and GenBank accession numbers are presented.

Species	Specimen voucher	Collection site	GenBank number	References
Aurantipileus mayanensis	TJB10228, type	Belize	HM772140	Ginns *et al.* (2010)
A. mayanensis	JV 1504/128	Costa Rica	KT156706	unpublished
Aurantiporus croceus[1]	BRNM 737561	Czech Republic	JQ821320	Dvořák *et al.* (2014)
A. croceus[1]	H6-27	Lithuania	MH571407	unpublished
A. croceus	VPapp 300518-1	**Hungary**	MT876120	**this study**
Geesterania carneola	MCW 474/13	Brazil	KY175000	Westphalen *et al.* (2018)
G. carneola	MCW 388/12	Brazil	KY174999	Westphalen *et al.* (2018)
G. davidii	MCW 396/12, type	Brazil	KY174998	Westphalen *et al.* (2018)
G. davidii	MCW 370/12	Brazil	KY174997	Westphalen *et al.* (2018)
Luteoporia albomarginata	Dai 15229, type	China	KU598873	Wu *et al.* (2016)
L. albomarginata	GC 1702-1	Taiwan	MF496147	Chen *et al.* (2018a)
L. albomarginata	TNM GC 1702-1	Taiwan	LC379003	Chen *et al.* (2018b)
Odoria alborubescens	PC 0706595, type	France	MG097863	Papp & Dima (2018)
O. alborubescens	BP 106943	Hungary	MG097864	Papp & Dima (2018)
O. alborubescens[2]	BRNU 627479	Czech Republic	JQ821319	Dvořák *et al.* (2014)
Pappia fissilis[3]	SFC20140626-03	South Korea	KX792915	Kim *et al.* (2016)
P. fissilis[3]	BRNM 699803	Czech Republic	HQ728292	Tomšovský (2012)
P. fissilis[3]	HHB9530sp	USA	KY948774	Justo *et al.* (2017)
Phlebiporia bubalina	Dai 13168, type	China	KC782526	Chen & Cui (2014)
P. bubalina	Dai 15231	China	KU598876	Wu *et al.* (2016)
P. bubalina	Dai 9798	China	KY131842	Wu *et al.* (2017)
Hapalopilus rutilans	JV0407/34J	USA	KX752619	Miettinen *et al.* (2016)
H. rutilans	H 6012735	Finland	KX752614	Miettinen *et al.* (2016)

[1] as *Hapalopilus*; [2] as *Aurantiporus*; [3] as *Tyromyces*

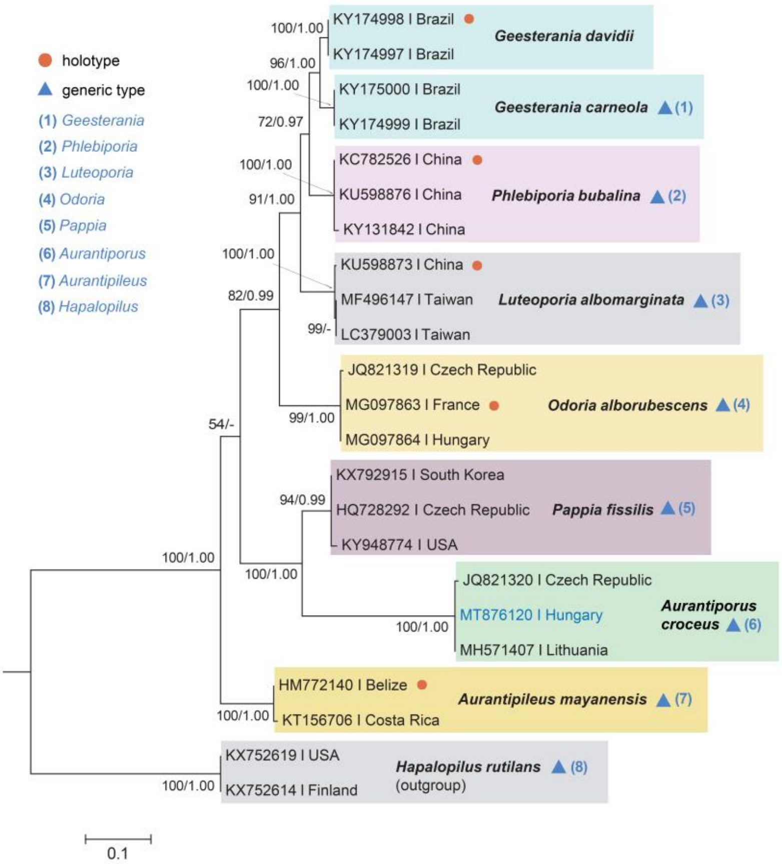


Figure 1. Phylogenetic tree of *Aurantiporus croceus* and related poroid species in Meruliaceae inferred from MrBayes and RAxML analyses of the nrDNA ITS sequences based on the best scoring maximum likelihood (ML) tree. *Hapalopilus rutilans* served for outgroup. Bayesian posterior probabilities (PP) > 0.9 and ML bootstrap values > 50% as evidences of statistical support are shown above or below branches. The bar indicates 0.1 expected change per site per branch.

RESULTS

ITS sequence analyses

The dataset represents 21 sequences of eight poroid Meruliaceae species, with *Hapalopilus rutilans* as outgroup. The Hungarian specimen (GenBank: MT876120) cluster together with other *A. croceus* specimens (labelled as *Hapalopilus croceus*) collected in

Czech Republic (GenBank: JQ821320) and Lithuania (GenBank: MH571407). The sequences of other morphologically similar European species formerly discussed in *Aurantiporus* (viz. *Odoria alborubescens* (Bourdot & Galzin) V. Papp & Dima and *Pappia fissilis* (Berk. & M.A. Curtis) Zmitr.) cluster in well-separated strongly supported clades, respectively.

Taxonomy

Aurantiporus croceus (Pers.) Murrill, Mycologia 12(1): 11, 1920 (*Figures 2–3*)

≡ *Boletus croceus* Pers., Observationes mycologicae 1: 87, 1796; ≡ *Polyporus croceus* (Pers.) Fr., Observ. mycol. (Havniae) 1: 124, 1815; ≡ *Phaeolus croceus* (Pers.) Pat., Essai taxonomique sur les familles et les genres des Hyménomycètes: 86, 1900; ≡ *Hapalopilus croceus* (Pers.) Bondartsev & Singer, Annales Mycologici 39 (1): 52, 1941; ≡ *Tyromyces croceus* (Pers.) J. Lowe, Mycotaxon 2 (1): 21, 1975

= *Polyporus pilotae* Schwein., Transactions of the American Philosophical Society 4 (2): 156, 1832

Basidiocarps annual, occasionally semi-perennial (*Figure 3*), broadly attached, pileate; upper surface bright orange-yellow at first, and finely pubescent, becomes vivid orange later, finely brownish orange and almost smooth when old; flesh vivid orange, dark wine-red to almost black when touched with KOH. Context soft and watery when fresh, shrinking considerably and becomes hard and rigid when dry, taste sourish or slightly bitter. Pore surface bright reddish-orange when fresh, brownish when dry, pores angular, 2–3 per mm. Tubes 2–3 cm thick, bright orange, spongiose and watery when fresh, drying darker orange to brownish, becomes hard and resinous. Hyphal system monomitic, hyphae hyaline and thin-walled, moderately branched, 3–6 µm in diam., septa with clamps. Hyphae richly encrusted with golden yellow crystals, forming a dense and loose covering around the wall. Yellow incrustation layer lose its elements in small pieces easily. Basidia 4-spored, 18–30 × 7–10 µm, clamped. Cystidia or other sterile elements absent. Basidiospores broadly ellipsoid, (4.02–)4.12–4.36(–4.48) × (2.82–)3.00–3.22(–3.26) µm, L = 4.25 µm, W = 3.1 µm, Q = 1.37 (n = 30), hyaline, thin-walled, smooth, negative in Melzer's reagent.

Specimens examined: HUNGARY. Vas County, near Sitke, Bajti, on old living *Quercus robur*, leg. Z. Igmándy, Pagony et Varga, 14 Sept 1964 (Igmándy 1488); leg. Haracsi et Igmándy, 17 Sept 1965

(Igmándy 1599), leg. Z. Igmándy et Varga, 3 Dec 1966 (Igmándy 1675); leg. Z. Igmándy, 8 Nov 1968 (Igmándy 1807); leg. Z. Igmándy, 28 Oct 1969 (Igmándy 1848); leg. Z. Igmándy, 11 Oct 1972 (Igmándy 2010); Fejér County, Vértes Mts, near Csákberény, Juhdöglő-völgy Forest Reserve, on the underside of large *Quercus* sp. log, leg. A. Koszka et V. Papp, 30 May 2018 (VPapp 300518-1), GenBank: MT876120. CZECH REPUBLIC. Moravia, on *Quercus robur*, leg. A. Černý, 2 May 1958 (Igmándy 10129).

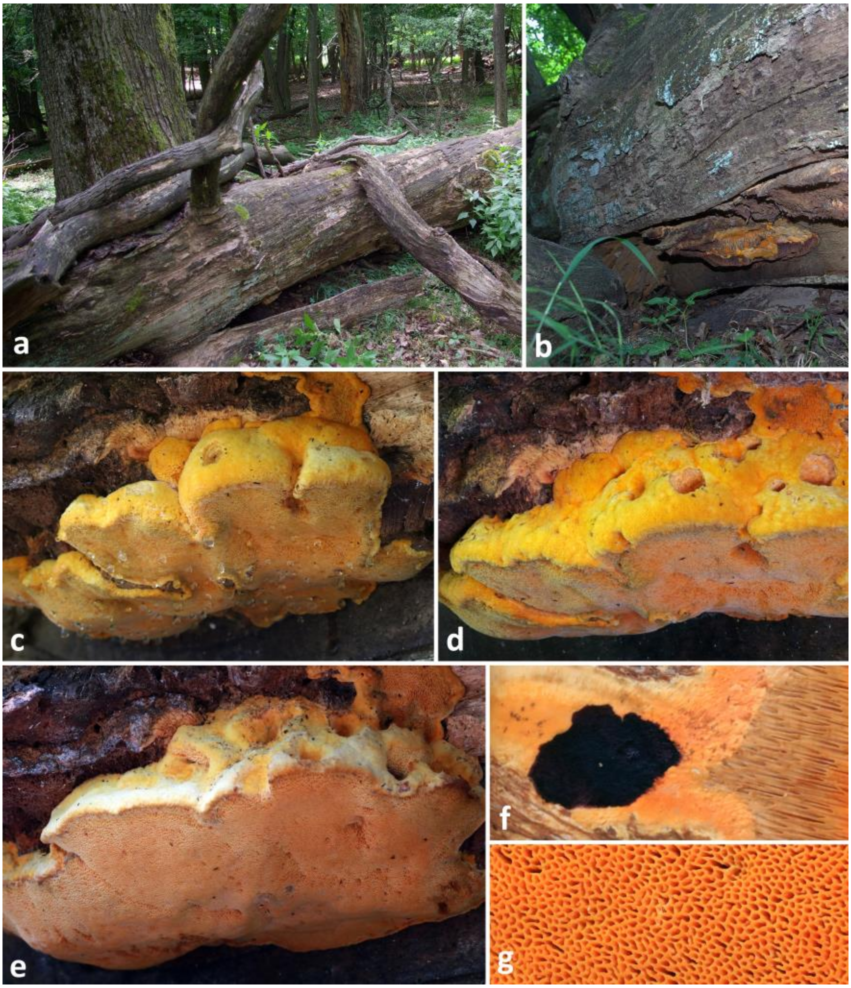


Figure 2. Macromorphology and habitat of *Aurantiporus croceus*. a–b: habitat in Juhdöglő-völgy Forest Reserve. c–e: basidiomata. f: KOH reaction of the basidiomata. g: pore surface (fresh material). Photos (a, b: V. Papp, c, e, f, g: A. Koszka, d: P. Finy).

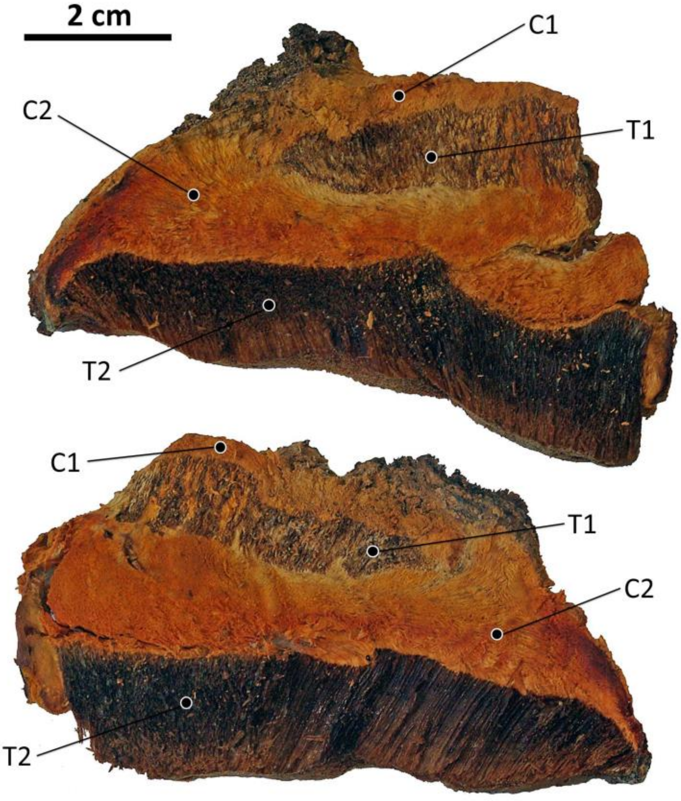


Figure 3. Cross-section of *Aurantiporus croceus* basidiome (dried material). C1: old context, C2: new context, T1: old trama, T2: new trama.

DISCUSSION

Most observations of *A. croceus* are originated from living, old and coarse veteran trees, mainly oak (*Quercus*) and more rarely on chestnut (*Castanea*) in parks and old growth forests (Dahlberg 2019). The new Hungarian location of *A. croceus* is reported from an old oak forest site at the Juhdöglő-völgy Forest Reserve, which is considered as one of the few European primary forests extant in the Pannonian Biogeographic region (Sabatini *et al*. 2018). The lignicolous funga of the Juhdöglő-völgy Forest Reserve have intensely been studied in the last decade, and several rare and threatened wood-inhabiting basidiomycetes were documented (Papp 2011, 2012, Papp and Szabó 2013, Papp and Dima 2014, 2018, Papp *et al*. 2012, 2016; Crous *et al*. 2018, Liu *et al*. 2018). Despite of the consecutive, targeted search of *A*. *croceus* at the old oak forest sections of the core area, only one location was observed. The basidiocarp grew 10 meters from the root zone of an old oak log (*Figure 2 a,b*). Although Juhdöglő-völgy Forest Reserve are protected and no forest management practices are being applied on the collection site, the crowded population of boar, red deer and mouflon creates a discontinuity in the forest. Thus, the genotype (or population) of *A*. *croceus* investigated in the Juhdöglő-völgy Forest Reserve is threatened by the decreasing habitat quality.

Acknowledgement: The authors wish to thank Jenő Jakab (Institute of Sylviculture and Forest Protection, University of Sopron) for providing additional information on the specimens collected by Zoltán Igmándy.

REFERENCES

ALTSCHUL, S.F., GISH, W., MILLER, W., MYERS, E.W. & LIPMAN, D.J. (1990). Basic local alignment search tool. *Journal of Molecular Biology* **215**: 403–410. https://doi.org/10.1016/S0022-2836(05)80360-2

BENSON, D.A., CAVANAUGH, M., CLARK, K., KARSCH-MIZRACHI, I., LIPMAN, D.J., OSTELL, J. & SAYERS, E.W. (2017). GenBank. *Nucleic Acids Research* **45**(D1): D37–D42. https://doi.org/10.1093/nar/gkw1070

CHEN, J.J. & CUI, B.K. (2014). *Phlebiporia bubalina* gen. et. sp. nov. (Meruliaceae, Polyporales) from Southwest China with a preliminary phylogeny based on rDNA sequences. *Mycological Progress* **13**: 563–573. https://doi.org/10.1007/s11557-013-0940-4

CHEN, C.C., WU, S.H. & CHEN, C.Y. (2018a). Twelve species of polypores (basidiomycetes) new to Taiwan. *Fungal Science* **33**(1): 7–19.

CHEN, C.C., WU, S.H. & CHEN, C.Y. (2018b). *Hydnophanerochaete* and *Odontoefibula*, two new genera of phanerochaetoid fungi (Polyporales, Basidiomycota) from East Asia. *MycoKeys* **39**: 75–96. https://doi.org/10.3897/mycokeys.39.28010

CROUS, P.W., WINGFIELD, M.J., BURGESS, T.I., HARDY, G.E.ST.J., GENÉ, J., GUARRO, J., BASEIA, I.G., GARCÍA, D., GUSMÃO, L.F.P., SOUZA-MOTTA, C.M., THANGAVEL, R., ADAMČÍK, S., BARILI, A., BARNES, C.W., BEZERRA, J.D.P., BORDALLO, J.J., CANO-LIRA, J.F., DE OLIVEIRA, R.J.V., ERCOLE, E., HUBKA, V., ITURRIETA-GONZÁLEZ, I., KUBÁTOVÁ, A., MARTÍN, M.P., MOREAU, P.-A., MORTE, A., ORDOÑEZ, M.E., RODRÍGUEZ, A., STCHIGEL, A.M., VIZZINI, A., ABDOLLAHZADEH, J., ABREU, V.P., ADAMČÍKOVÁ, K., ALBUQUERQUE, G.M.R., ALEXANDROVA, A.V., Álvarez Duarte, E., Armstrong-Cho, C., Banniza, S., Barbosa, R.N., Bellanger, J.-M., Bezerra, J.L., Cabral, T.S., Caboň, M., Caicedo, E., Cantillo, T., Carnegie, A.J., Carmo, L.T., Castañeda-Ruiz, R.F., Clement, C.R., Čmoková, A., Conceição, L.B., Cruz, R.H.S.F., Damm, U., da Silva, B.D.B., da Silva, G.A., da Silva, R.M.F., Santiago, A.L.C.M. de A., de Oliveira, L.F., de Souza, C.A.F., Déniel, F., Dima, B., Dong, G., Edwards, J., Félix, C.R., Fournier, J., Gibertoni, T.B., Hosaka, K., Iturriaga, T., Jadan, M., Jany, J.-L., Jurjević, Ž., Kolařík, M., Kušan, I., Landell, M.F., Leite Cordeiro, T.R., Lima, D.X., Loizides, M., Luo, S., Machado, A.R., Madrid, H., Magalhães, O.M.C., Marinho, P., Matočec, N., Mešić, A., Miller, A.N., Morozova, O.V., Neves, R.P., Nonaka, K., Nováková, A., Oberlies, N.H., Oliveira-Filho, J.R.C., Oliveira, T.G.L., Papp, V., Pereira, O.L., Perrone, G., Peterson, S.W., Pham, T.H.G., Raja, H.A., Raudabaugh, D.B., Iřehulka, J., Rodrāguez-Andrade, E., Saba, M., Schauflerovā¡, A., Shivas, R.G., Simonini, G., Siqueira, J.P.Z., Sousa, J.O., Stajsic, V., Svetasheva, T., Tan, Y.P., Tkalčec, Z., Ullah, S., Valente, P., Valenzuela-Lopez, N., Abrinbana, M., Viana Marques, D.A., Wong, P.T.W., Xavier de Lima, V. & Groenewald, J.Z. (2018). Fungal Planet description sheets 716–784. *Persoonia* **40**: 240–387. https://doi.org/10.3767/persoonia.2018.40.10

Dahlberg, A. (2019). *Hapalopilus croceus*. The IUCN Red List of Threatened Species 2019: e.T58521209A58521216.

Dahlberg, A. & Croneborg, H. (eds.) (2003). 33 threatened fungi in Europe. Complementary and revised information on candidates for listing in Appendix 1 of the Bern Convention. Swedish Environmental Protection Agency (EPA) and European Council for the Conservation of Fungi (ECCF), 82 pp.

Dai, Y.C. (2012). Polypore diversity in China with an annotated checklist of Chinese polypores. *Mycoscience* **53**: 49–80. https://doi.org/10.1007/s10267-011-0134-3

Dvořák, D., Běťák, J. & Tomšovský, M. (2014). *Aurantiporus alborubescens* (Basidiomycota, Polyporales) – first record in the Carpathians and notes on its systematic position. *Czech Mycology* **66**(1): 71–84. https://doi.org/10.33585/cmy.66105

Gardes, M. & Bruns, T.D. (1993). ITS primers with enhanced specificity for basidiomycetes – application to the identification of mycorrhizae and rusts. *Molecular Ecology* **2**(2): 113–118. https://doi.org/10.1111/j.1365-294x.1993.tb00005.x

Ginns, J., Lindner, D.L., Baroni, T.J. & Ryvarden, L. (2010). *Aurantiopileus mayanensis* a new genus and species of polypore (Polyporales, Basidiomycota) from Belize with connections to existing Asian species. *North American Fungi* **5**(4): 1–10. https://doi.org/10.2509/naf2010.005.004

Gouy, M., Guindon, S. & Gascuel, O. (2010). SeaView version 4: a multiplatform graphical user interface for sequence alignment and phylogenetic tree building. *Molecular Biology and Evolution* **27**(2): 221–224. https://doi.org/10.1093/molbev/msp259

Igmándy, Z. (1968). Die Porlinge Ungarns und ihre phytopathologische Bedeutung III. (*Polipori Hungariae* III.). *Acta Phytopathologica Academiae Scientiarum Hungaricae* **3**(3): 349–359.

Justo, A., Miettinen, O., Floudas, D., Ortiz-Santana, B., Sjökvist, E., Lindner, D., Nakasone, K., Niemelä, T., Larsson, K.H., Ryvarden, L. & Hibbett, D.S. (2017). A revised family-level classification of the Polyporales (Basidiomycota). *Fungal Biology* **121**: 798–824. https://doi.org/10.1016/j.funbio.2017.05.010

Kim, N.K., Park, J.Y., Park, M.S., Lee, H., Cho, H.J., Eimes, J.A., Kim, C. & Lim, Y.W. (2016). Five new wood decay fungi (Polyporales and Hymenochaetales) in Korea. *Mycobiology* **44**(3): 146–154. https://doi.org/10.5941/myco.2016.44.3.146

Liu, L.-N., Razaq, A., Atri, N.S., Bau, T., Belbahri, L., Chenari Bouket, A., Chen, L.-P., Deng, C., Ilyas, S., Khalid, A.N., Kitaura, M.J., Kobayashi, T., Li Y., Lorenz, A.P., Ma, Y.-H., Malysheva, E., Malysheva, V., Nuytinck, J., Qiao, M., Saini, M.K., Scur, M.C., Sharma, S., Shu, L.-L., Spirin, V., Tanaka, Y., Tojo, M., Uzuhashi, S., Valério-Júnior, C., Verbeken, A., Verma, B., Wu, R.-H., Xu, J.-P., Yu, Z.-F., Zeng, H., Zhang, B., Banerjee, A., Beddiar, A., Bordallo, J.J., Dafri, A., Dima, B., Krisai-Greilhuber, I., Lorenzini, M., Mandal, R., Morte, A., Nath, P.S., Papp, V., Pavlík, J., Rodríguez, A., Ševčíková, H., Urban, A., Voglmayr, H. & Zapparoli, G. (2018). Fungal Systematics and Evolution: FUSE 4. *Sydowia* **70**: 211–286. https://doi.org/10.12905/0380.sydowia70-2018-0211

Löytynoja, A. & Goldman, N. (2005). An algorithm for progressive multiple alignment of sequences with insertions. *Proceedings of the National Academy of Sciences of the United States of America* **102**(30): 10557–10562. https://doi.org/10.1073/pnas.0409137102

Löytynoja, A. & Goldman, N. (2008). Phylogeny-aware gap placement prevents errors in sequence alignment and evolutionary analysis. *Science* **320**: 1632–1635. https://doi.org/10.1126/science.1158395

Miettinen, O., Spirin, V., Vlasák, J., Rivoire, B., Stenroos, S. & Hibbett, D.S. (2016). Polypores and genus concepts in Phanerochaetaceae (Polyporales, Basidiomycota). *MycoKeys* **17**: 1–46. https://doi.org/10.3897/mycokeys.17.10153

MK (2013). 83/2013. (IX. 25.) VM rendelet. A védett és a fokozottan védett növény- és állatfajokról, a fokozottan védett barlangok köréről, valamint az Európai Közösségben természetvédelmi szempontból jelentős növény- és állatfajok közzétételéről szóló 13/2001. (V. 9.) KöM rendelet módosításáról. *Magyar Közlöny* **2013**(156): 67479–67503. (in Hungarian)

Papp, V. (2011). Adatok a *Xylobolus* nemzetség magyarországi előfordulásáról. *Mikológiai Közlemények, Clusiana* **50**(2): 173–182. (in Hungarian)

Papp, V. (2012). A *Frantisekia mentschulensis* első magyarországi előfordulása. *Mikológiai Közlemények, Clusiana* **51**(2): 181–186. (in Hungarian)

Papp, V. & Dima, B. (2014). A *Pholiota squarrosoides* első magyarországi előfordulása és előzetes filogenetikai vizsgálata. *Mikológiai Közlemények, Clusiana* **53**(1–2): 33–42. (in Hungarian)

Papp, V. & Dima, B. (2017). *Favolus gracilisporus* (Polyporaceae, Basidiomycota), an East Asian polypore species new to the European mycobiota. *Mycosphere* **8**(6): 1177–1184. https://doi.org/10.5943/mycosphere/8/6/7

Papp, V. & Dima, B. (2018). New systematic position of *Aurantiporus alborubescens* (Meruliaceae, Basidiomycota), a threatened old-growth forest polypore. *Mycological Progress* **17**(3): 319–332. https://doi.org/10.1007/s11557-017-1356-3

Papp, V., Dima, B., Koszka, A. & Siller, I. (2012). A *Donkia pulcherrima* (Polyporales, Basidiomycota) első magyarországi előfordulása és taxonómiai értékelése. *Mikológiai Közlemények, Clusiana* **53**(1–2): 43–53. (in Hungarian)

PAPP, V., KIRÁLY, G., KOSCSÓ, J., MALATINSZKY, Á., NAGY, T., TAKÁCS, A. & DIMA, B. (2016). Taxonomical and chorological notes 2 (20–27). *Studia botanica hungarica* **47**(1): 179–191. https://doi.org/10.17110/studbot.2016.47.1.179

PAPP, V. & SZABÓ, I. (2013). Distribution and host preference of poroid basidiomycetes in Hungary I. – Ganoderma. *Acta Silvatica et Lignaria Hungarica* **9**: 71–83. https://doi.org/10.2478/aslh-2013-0006

RIMÓCZI, I., SILLER, I., VASAS, G., ALBERT, L., VETTER, J. & BRATEK, Z. (1999). Magyarország nagygombáinak javasolt vörös listája. *Mikológiai Közlemények, Clusiana* **38**(1–3): 107–132. (in Hungarian)

RONQUIST, F. & HUELSENBECK, J.P. (2003). MrBayes 3: Bayesian phylogenetic inference under mixed models. *Bioinformatics* **19**: 1572–1574. https://doi.org/10.1093/bioinformatics/btg180

RYVARDEN, L. & MELO, I. (2014). Poroid fungi of Europe. *Synopsis Fungorum* **31**: 1–450.

SABATINI, F.M., BURRASCANO, S., KEETON, W.S., LEVERS, C., LINDNER, M., PÖTZSCHNER, F., VERKERK, P.J., BAUHUS, J., BUCHWALD, E., CHASKOVSKY, O., DEBAIVE, N., HORVÁTH, F., GARBARINO, M., GRIGORIADIS, N., LOMBARDI, F., DUARTE, I.M., MEYER, P., MIDTENG, R., MIKAC, S., MIKOLAS, M., MOTTA, R., MOZGERIS, G., NUNES, L., PANAYOTOV, M., ÓDOR, P., RUETE, A., SIMOVSKI, B., STILLHARD, J., SVOBODA, M., SZWAGRZYK, J., TIKKANEN, O.-P., VOLOSYANCHUK, R., VRSKA, T., ZLATANOV, T.M. & KUEMMERLE, T. (2018). Where are Europe's last primary forests? *Diversity and Distributions* **24**(12): 1890–1892. https://doi.org/10.1111/ddi.12778

SILVESTRO, D. & MICHALAK, I. (2012). raxmlGUI: a graphical front-end for RAxML. *Organisms Diversity & Evolution* **12**: 335–337. https://doi.org/10.1007/s13127-011-0056-0

SZABÓ, I. (2012). Poroid fungi of Hungary in the collection of Zoltán Igmándy. *Acta Silvatica et Lignaria Hungarica* **8**: 113–122. https://doi.org/10.2478/v10303-012-0009-0

TAMURA, K., STECHER, G., PETERSON, D., FILIPSKI, A. & KUMAR, S. (2013). MEGA6: molecular evolutionary genetics analysis version 6.0. *Molecular Biology and Evolution* **30**(12): 2725–2729. https://doi.org/10.1093/molbev/mst197

TOMŠOVSKÝ, M. (2012). Delimitation of an almost forgotten species *Spongipellis litschaueri* (Polyporales, Basidiomycota) and its taxonomic position within the genus. *Mycological Progress* **11**(2): 415–424. https://doi.org/10.1007/s11557-011-0756-z

WESTPHALEN, M.C, RAJCHENBERG, M., TOMŠOVSKÝ, M. & GUGLIOTTA, A.M. (2018). A re-evaluation of Neotropical *Junghuhnia* s.lat. (Polyporales, Basidiomycota) based on morphological and multigene analyses. *Persoonia* **41**: 130–141. https://doi.org/10.3767/persoonia.2018.41.07

WHITE, T.J., BRUNS, T., LEE, S. & TAYLOR, J. (1990). Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: INNIS, M.A., GELFAND, D.H., SNINSKY, J.J. & WHITE, T.J. (eds.): PCR protocols: a guide to methods and applications. Academic Press, New York, pp. 315–332.

WU, F., YUAN, Y., CHEN, J.J. & HE, S.H. (2016). *Luteoporia albomarginata* gen. et sp. nov. (Meruliaceae, Basidiomycota) from tropical China. *Phytotaxa* **263**(1): 31–41. https://doi.org/10.11646/phytotaxa.263.1.3

WU, F., CHEN, J.J., JI, X.H., VLASÁK, J. & DAI, Y.C. (2017). Phylogeny and diversity of the morphologically similar polypore genera *Rigidoporus*, *Physisporinus*, *Oxyporus*, and *Leucophellinus*. *Mycologia* **109**: 749–765. https://doi.org/10.1080/00275514.2017.1405215

ZHOU, L.W., NAKASONE, K.K., BURDSALL, H.H. JR, GINNS, J., VLASÁK, J., MIETTINEN, O., SPIRIN, V., NIEMELÄ, T., YUAN, H.S., HE, S.H., CUI, B.K., XING, J.H. & DAI, Y.C. (2016). Polypore diversity in North America with an annotated checklist. *Mycological Progress* **15**: 771–790. https://doi.org/10.1007/s11557-016-1207-7

(submitted: 20.05.2020, accepted: 12.08.2020)

Acta Biologica Plantarum Agriensis **8**(1): 53–68 (2020)
ISSN 2061-6716 (Print), 2063-6725 (Online) *Research article*
http://abpa.ektf.hu/
https://doi.org/10.21406/abpa.2020.8.1.53

EXTRACTION AND IDENTIFICATION OF MAJOR POLYPHENOL CONSTITUENTS OF NORTHERN HUNGARIAN HORSEMINT (*MENTHA LONGIFOLIA* L. (L.))

Katalin Patonay[1]*, Orsolya Szabó-Hudák[1], Helga Szalontai[1], Mihály Jánószky[1], Erika Pénzesné Kónya[2] & Éva Zámboriné Németh[3]

*[1]Food and Wine Research Institute, Eszterházy Károly University, 6G Leányka u., H-3300, Eger, Hungary; [2]Department of Botany and Plant Physiology, Eszterházy Károly University, 6D Leányka u., H-3300, Eger, Hungary; [3]Department of Medicinal and Aromatic Plants, Szent István University, 29-43 Villányi u., H-1118, Budapest, Hungary; *E-mail: patonay.katalin@uni-eszterhazy.hu*

Abstract: (*Mentha longifolia* (L.) L.) is a less studied species from the viewpoint of its nonvolatile constituents and this work is the first phytochemical screening of it in Hungary. The aim of the experiments is to elaborate an efficient extraction method of its polyphenolic antioxidants. Thirty-six accessions were sampled. Soxhlet and an ultrasonic extraction were applied, both with methanol (MeOH) and ethanol:water 7:3 (WA). The analysis of the extracts was carried out by HPLC-DAD method. Rosmarinic acid, hesperidin, diosmin and cynaroside were observed to be common constituents of all samples. Other major constituents were observed in all of the chromatograms at ca. t_R=13.5 min. and t_R=19.7 min (unknown A and B). Their identification needs further examination. Regarding antioxidant activity, WA extraction was more efficient than MeOH Soxhlet extraction.

Keywords: *Mentha longifolia*, horsemint, polyphenolic antioxidants, HPLC, antioxidant activity

INTRODUCTION

Horsemint, *Mentha longifolia* (L.) L., *Lamiaceae* (further: ML) is a wild-growing perennial species. It has a large distribution area in Eurasia and non-tropical Africa (Tucker and Naczi 2007), grows in multiple types of habitats such as wet meadows, roadsides and urban areas (Başer *et al.* 1999). Active ingredients of ML originate from the mevalonate and shikimate pathways. The former produces the volatile terpenes of ML, most frequently ketones as carvone, piperitone, piperitenone and corresponding epoxides

(Başer *et al*. 1999; Sharopov *et al*. 2012). Triterpenes in ML (ursolic, oleanolic acids etc.) also originate from this pathway. The shikimate pathway is the origin of phenolics of ML. Caffeic acid derivatives (predominantly the known antiradical molecule rosmarinic acid) are present. But flavonoids also deserve attention: the presence of 55 different flavonoids was reported (Bourwieg and Pohl 1973; Ghoulami *et al*. 2001; Pereira and Cardoso 2013 Hawrył *et al*. 2016, Patonay *et al.* 2017; Park *et al*. 2019), six of them *de novo* (Sharaf *et al*. 1999; Jahan *et al*. 2001; Ghoulami *et al*. 2001; Ali *et al*. 2002). Regarding flavonoid subclasses, flavones seem to be present in the largest variability.

There are several studies dealing with the phytochemistry of horsemint, although the majority of them deals with volatile terpenoids. Studies focusing to polyphenolic constituents most frequently evaluate a single sample of the species (e. g. Bourwieg and Pohl 1973; Sharaf *et al*. 1999; Jahan *et al*. 2001; Ghoulami *et al*. 2001; Ali *et al*. 2002; Fialová *et al*. 2008; Krzyzanowska *et al*. 2011; Shen *et al*. 2011; Stanislavljević *et al*. 2012; Benedec *et al*. 2013; Bahadori *et al*. 2018; Park *et al*. 2019). Relevant studies on a larger representative sample pool dealing with flavonoid composition of horsemint are not available, in contrary to e.g. peppermint (Guédon and Pasquier 1994). For the major nonflavonoid phenolics of horsemint, one single large-sample study has been found (Dudai *et al*. 2006) providing the rosmarinic and caffeic acid contents of 40 different clones. Therefore, it can be established, that conclusions on the distribution pattern of polyphenols in horsemint populations and on the factors influencing their accumulation would be difficult. Additionally, a part of the former studies does not provide a detailed description of the extraction method applied, making the comparison of data difficult (e. g. Ghoulami *et al*. 2001; Jahan *et al*. 2001; Benedec *et al*. 2013). Thus, contrary to the abundance of this species, knowledge on its phytochemistry, especially in case of the polyphenolic compounds, still seems to be incomplete. The present study on the extractability and polyphenol composition of the Northern Hungarian ML populations has been carried out in the framework of a wider project which intends to utilize this species as a cultivated source of antioxidant and/or preservative preparations in the food industry. Thus, one step is to define an effective extraction method of horsemint resulting in the highest antioxidant capacity and polyphenol content of the samples. In parallel, screening the polyphenol composition supplements our knowledge on this species and may help in elucidating the background of the antioxidant properties.

MATERIALS AND METHODS

Sampling: Thirty-six shoot samples of ML were collected in the flowering stage, from the end of June to the beginning of October, 2016. Populations in the Bükk Mountains, Mátra Mountains, Heves-Borsodi Hills and Eger were surveyed. Identification: one item (ID: NOSZ) by the collector Jana Táborská (Eszterházy Károly University, Department of Botany and Plant Physiology) ten items (JOF, HV1, HV2, HV3, HOV1, HOV2, HOV3, KÜH, JÁV and EGR4) by the collector Helga Szalontai and other samples: by Erika Pénzesné Kónya. The identification was performed on the basis of macromorphology, using the manual of the vascular flora of Hungary (Simon 1994). Voucher specimen deposition: at the Eszterházy Károly University in the chemotaxonomical collection of the herbarium (EGR-CH). Samples were hung in a shady place for 21 days. After drying, leaves and inflorescences were separated from stems, collected and deposited in the freezer.

Extraction. Extraction in a cooled ultrasonic bath (45 kHz, 3 stages, 45 min/stage, at maximum 30°C) was applied. Sonication may be expected to be milder toward many plant polyphenols of heat sensitivity as it can be performed at room temperature and normal pressure. Another advantageous traits of this method are the much lesser time demand and moderate solvent demand in comparison with Soxhlet extraction or the conventional room temperature maceration of plant material (Medina-Torres *et al.* 2017). Furthermore, for rosmarinic and carnosic acids it was observed that the sonication gives better yield of them in comparison with conventional room-temperature maceration of rosemary leaves, both in case of methanolic and ethanolic extractions (Paniwnyk *et al.* 2009) and the antioxidant activity of extracts gained by ultrasonic treatments is higher than the values of extracts prepared by maceration. A comparative study on extraction of polyphenols of *Tilia cordata* inflorescences established that the sonication was the most efficient method to extract phenolic acids of *Tiliae flos* and the second efficient method to gain flavonol constituents of it when sonication, microwave extraction, accelerated solvent extraction, heat reflux extraction, and Soxhlet were compared (Oniszczuk *et al*. 2015)

In parallel, the Soxhlet method was applied as a reference method, as nowadays it is still frequently used to extract different plant materials (e. g. Ahmed *et al*. 2015) including horsemint (e. g. Güllüce *et al*. 2007; Iqbal *et al*. 2013) and in comparative investigations on extractability of some plants (Karabegović *et al*. 2011). The solvents were methanol (MeOH) and 7:3 ethanol: water mixture (WA). Methanol (MeOH) was taken as a reference, as for this species a significant part of investigators use it when polar extracts are prepared (e.g.: Hajlaoui *et al.* 2009; Güllüce *et al*. 2007; Iqbal *et al*. 2013; Murad *et al*. 2016). The aqueous ethanol with 7:3 ethanol: water ratio (WA) is not frequently mentioned in the references on this species (Mimica-Dukić *et al*. 1999; Stanislavljević *et al.* 2012; Benedec *et al*. 2013) however, the solvent is applied in pharmaceutical practice to prepare polyphenol rich tinctures i. e. from common tormentil (*Potentilla erecta*) rhizome (Kőszeginé Szalai *et al*. 2003). In this manner, four different extracts of each of the thirty-six horsemint samples were obtained.

In vitro antioxidant parameters Antiradical activity was investigated by DPPH assay (0.1 mM DPPH in ethanol/30 min/room temperature/normal light). Results are given in EC_{50} mg/l extract. This means the concentration of the extract consumes half of the initial concentration of DPPH, thus a lower value means stronger activity. Reducing ability was investigated on FRAP assay. Briefly, a modified method of Benzie and Strain (1996) was used at the assay. Fifty ml of buffer (46.3 ml 0.2M acetic acid plus 3.7 ml of 0.2M Na-acetate filled to 100 ml) was measured in with 5 ml 10mM 2,4,6-s-tripyridyl-triazine (TPTZ) acidified with 84 µl cc HCl/25 ml solution. In the last step 5 ml 20mM ferric chloride was added to the mixture. 50 µl of sample with 1450 µl of this mixed reagent was reacted for 5 min (under normal light and room temperature). Spectra were collected and at 596 ± 1 nm evaluated. The results were determined as mg ascorbic acid equivalents (AAE) per kg dry weight (DW). Total polyphenol content was investigated by Folin-Ciocalteu assay, with modifications of the protocol of Waterhouse (Waterhouse 2002) and given in gallic acid eq. (GAE) mg/kg DW. Suppliers of chemicals used for the antioxidant assays are as follows: DPPH, a. r., Sigma-Aldrich. Folin and Ciocalteu's reagent, 2N, VWR. TPTZ, 98%, Alfa Aesar.

HPLC investigations Apparatus: Agilent 1200 Series with diode array detector G1315D Column: ACE C18250 mm*4.6 mm*5 µm. Eluent A: aqueous acetic acid (5 V/V%) and B: acetonitrile. Injection: 10 µl. The applied gradient is shown in *Table 1*.

Table 1. Timetable of the gradient program applied to the horsemint extracts.

t min	ACN %
0	5
5	5
8	20
11	20
13	25
16	30
22.5	25
24.5	25
30	50
32	95
34	5
37	5

Identification of components was based on retention times of analytical standards. Standards were Caffeic acid, Sigma-Aldrich, min. 99%, predominantly trans; rosmarinic acid, Aldrich, min. 99%; Hesperidin, Sigma-Aldrich, min. 97%, analytical standard; Cynaroside, Sigma-Aldrich, min. 98%, analytical standard; Diosmin, Sigma-Aldrich, min. 90%, analytical standard. Solvents: Acetonitrile, VWR Chromanorm, super gradient grade Reag. Ph Eur, USP, ACS. Acetic acid, glacial: VWR AnalAR Normapur.

Quantitation of the identified compounds were based on calibration with their standards. Results here are expressed as the maximal extractable concentration, in mg component/kg dry plant material weight (DW). For each sample, four concentration values were gained for the main phenolics according the different extraction methods described above. These were compared and the maxima were chosen. This approach may better approximate the real concentrations of the ingredients in the plant thus the variability of the concentrations among the 36 populations may be more recognizable.

RESULTS AND DISCUSSION

Extractability and antioxidant activity

The polyphenol contents calculated from the four different extract types has shown a strong solvent dependence. Extractions made with WA were more efficient. *Table 2* depicts the basic statistical data for the TPC of four extract types, which are calculated from the detailed results provided in our previous study (Patonay *et al.* 2019). The similarity of TPC values of the two WA extractions are observable, together with the weak efficiency of MeOH Soxhlet extraction. In Welch tests, significant difference of the obtained TPC values were observable between the two ultrasound extractions ($p<0.025$), the two Soxhlets ($p<0.01$), and between the two methanolic extract types but as it was expected from their similar values, no significant difference was found between the two hydroalcoholic treatments.

Table 2. Comparison of descriptive statistics of TPC of ML samples, calculated from the different extracts. All data is given in mg GAE/kg DW. US stands for ‘ultrasonic’.

	MeOH Soxhlet	**MeOH US**	**WA Soxhlet**	**WA US**
Q1	25836	39143	41875	46148
Median	35373	47520	53721	51684
Q3	41295	55901	61351	58333
Min	6295	7105	18381	23390
Max	49033	67952	87024	67910

The solvent dependence appears in the results of the antioxidant assays, too. WA extractions provide extracts with higher antioxidant activity, than methanolic ones. But additionally, in these assays, dependence on the extraction method is also noticeable. WA sonication was observed to be more efficient than WA Soxhlet, contrary to their similarity in total polyphenol contents. Methanolic Soxhlet extraction resulted in the least efficiency, contrary to its usage appearing in the literature. Basic descriptive statistical data of DPPH EC_{50} measurements on each extract series are shown in *Table 3*. The antiradical activity of the four extract types has shown significant differences on the Welch test (from $p<0.025$ to $p<0.001$) in all arrangements of comparisons (two hydroalcoholic extractions, two methanolic extractions, two Soxhlets and two ultrasonic extractions).

Table 3. Comparison of descriptive statistics of EC_{50}–mg/L values of the different ML extracts. US stands for 'ultrasonic'.

	MeOH Soxhlet	MeOH US	WA Soxhlet	WA US
Q1	466.7	403.9	303.2	323.1
Q3	727.8	608.3	462.6	383.3
Median	583.1	501.6	362.1	355.8
Min	353.3	291.7	168.5	226.9
Max	3183	1736	983.4	806.4

The ferrous reducing activities follow a similar trend as the DPPH data. Methanolic extraction is less efficient, than the WA extractions – partially, methanolic Soxhlet which is found to be the least effective. Sonication with WA is noticed to be more efficient, than WA Soxhlet extraction. The statistical characterization of FRAP values are presented in *Table 4*.

Table 4. Comparison of descriptive statistics of FRAP values calculated from the different extracts. All data is given in mg AAE/kg DW. US stands for 'ultrasonic'.

	MeOH Soxhlet	MeOH US	WA Soxhlet	WA US
Q1	5236	5498	6587	7173
Q3	6704	7301	9975	10447
Median	6118	6275	8017	9102
Min	3264	2875	3441	5671
Max	7463	9101	11969	12543

The constituents detected in all the thirty-six samples of the horsemint

Six major peaks were observed to appear in all of the extracts and until now, four of them have been identified and quantified. These are, in order of their elutions, the following: i) cynaroside, the 7-O-glucoside of luteolin (5,7,3',4'-tetrahydroxyflavone) ii) diosmin, the 7-O-rutinoside of diosmetin (5,7,3'-trihydroxy-4'-methoxyflavone) iii) hesperidin, 7-O-rutinoside of hesperetin (5,7,3'-trihydroxy-4'-

methoxyflavanone) and iv) rosmarinic acid, the caffeoyl ester of 3,4-dihydroxyphenyllactic acid. *Figure 2* is the chromatogram of a WA Soxhlet extract, also showing the structure of the identified four ingredients and designation of their peaks. Their concentrations in the studied extracts are different, depending on the plant material and, as it is expected, on solvent type. For example, hesperidin is concentrated in methanolic extracts up to 29815 mg/kg calculated to DW, while it is present in significantly lower concentrations in hydroalcoholic extracts. Identification of the two other major peaks Unknown A and B requires further investigations.

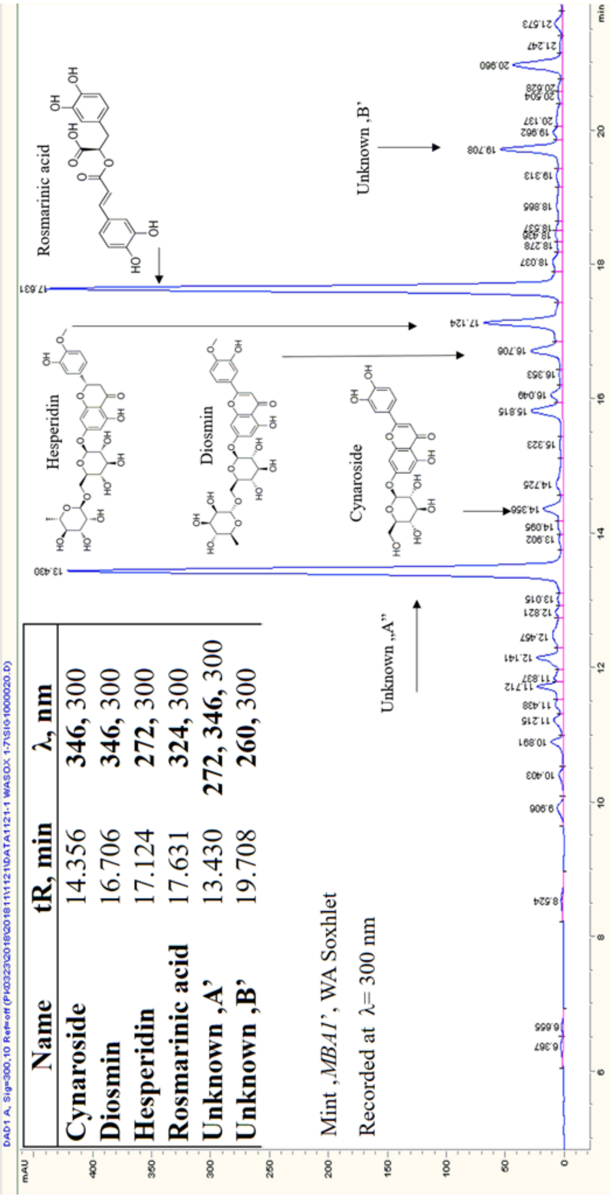
As it was expected, rosmarinic acid is a dominant constituent in the horsemint samples while other hydroxycinnamic acids as major components were not found. Another peculiar trait of the samples is the high concentration of hesperidin. This compound is frequently detected in *Mentha* plants (Guédon and Pasquier 1994) (Hanafy *et al*. 2017) but in varying concentrations, not often as a dominant constituent. Diosmin is also frequent in mints, e. g. in peppermint (Guédon and Pasquier 1994). In the case of the horsemint, presence of this compound was only reported once, from a single sample (Bourwieg and Pohl 1973).

Figure 2 shows the distribution of the identified four polyphenols in the thirty-six plant samples and *Table 2* summarizes the concentration intervals of them.

As it was expected, a high variability is observable in the concentration of each polyphenol amongst the plant samples.

Table 5. Intervals of the highest extracted concentrations of the identified polyphenols in the thirty-six horsemint samples.

Component	Highest extracted concentration, mg/kg DW
Rosmarinic acid	7043-38667
Cynaroside	<200-2944
Hesperidin	1985-29815
Diosmin	398-7987



Name	tR, min	λ, nm
Cynaroside	14.356	346, 300
Diosmin	16.706	346, 300
Hesperidin	17.124	272, 300
Rosmarinic acid	17.631	324, 300
Unknown ‚A'	13.430	272, 346, 300
Unknown ‚B'	19.708	260, 300

Mint ‚*MBA1*', WA Soxhlet

Recorded at λ= 300 nm

Figure 1. Chromatogram of mint MBA1, WA Soxhlet extract. Table included to the left upper corner contains the retention times of the six common constituents of the mint samples, together with their most intensive detection wavelengths. Structural formulae of the identified four polyphenols are inserted above their peaks.

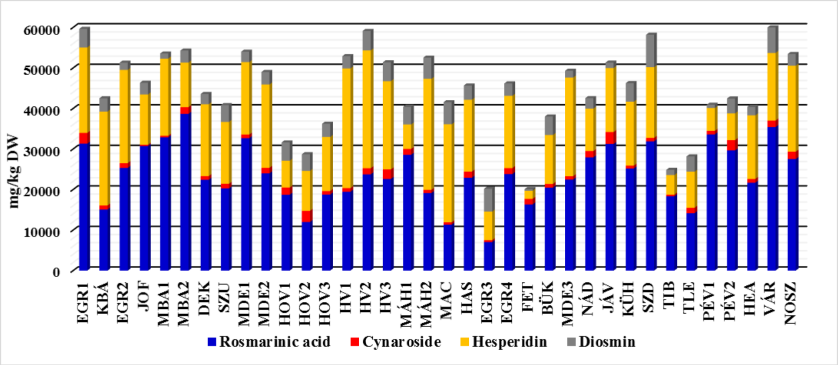


Figure 2. Highest extracted concentrations of the identified polypenols in the horsemint samples, given in mg/kg DW. On the x-axis of the chart the sample ID-s are provided.

Antioxidant properties of the identified polyphenols and their connection with the antioxidant capacities of the extracts

Table 6 contains the DPPH and FRAP data measured on the standards of the four identified polyphenols, compared with the values of a widely used control antioxidant, di(*tert*-)butyl-hydroxytoluene (*BHT* or *E321*).

Table 6. DPPH and FRAP values of the four identified polyphenol compounds in comparison with BHT as positive control.

Standard	**DPPH EC$_{50}$ mg/l**		**FRAP mg AAE/kg**	
	Mean	**S. D.**	**Mean**	**S. D.**
Rosmarinic acid	16	0.75	172499	1044
Cynaroside	33	0.03	68637	2910
Hesperidin	6352	84.00	13333	873
Diosmin	>25000	NA	3098	42
BHT	*87.35*	*4.51*	*12169*	*34*

Rosmarinic acid and cynaroside were observed to be strong antiradical and reducing agents, being more active than BHT. Rosmarinic acid shows outstandingly strong activity in both antioxidant activity assays, especially in FRAP. This is in accordance with the available literature data each in ferrous reducing activity (Hadjimehdipoor *et al.* 2014) and in DPPH testing (Damien-Dorman *et al.* 2003; Koşar *et al.* 2004). Hesperidin was observed to be a weak antiradical agent and diosmin is practically inactive in both assays.

The difference in the antiradical activity of cynaroside and the other two flavonoids may be justified with structural background: cynaroside has free C3'-OH and C4'-OH (free catechol moiety) which is highly important in effective radical scavenging (Bors *et al.* 1990). Contrary to this, in the case of hesperidin and diosmin the C4' site is blocked by a methoxy group. The correlation between the antioxidant properties of the extracts and the concentrations of the four phenolics in them was investigated. Rosmarinic acid concentration was found to be in significant correlation both with the antiradical and the ferrous reducing activity. Strength of correlations of DPPH EC_{50} values and rosmarinic acid content of the extracts was medium (R between 0.4990 and 0.5555 depending on extract type) with $p<0.01$. To ferrous reducing activity, R was calculated between 0.3551 and 0.5530 (also in dependence with the extract type) with $p<0.01$. To cynaroside concentrations, contrary to the high antioxidant activity of this substance, no significant correlation was found with the antioxidant properties of the extracts. No correlation between the two other identified flavonoids and the antioxidant capacities was detected, which is not unexpected regarding the weaker activity of them.

Concluding remarks

Based on our results, WA may be the appropriate extract type for assuring a high *in vitro* antiradical activity. Besides, it can be used in food industry without restrictions, contrary to methanol to which strict limitations have been declared (The European Parliament and Council 2009, 2010). The better efficacy of WA than methanol in our study is especially interesting as in numerous former references methanol was frequently used as a polar medium to gain polyphenolic antioxidants of the plant.

The investigated horsemint samples contained high levels of rosmarinic acid. Concentration of this constituent may reach high values also in other mints: 0.97-3.86 m/m% DW, ca. 9700-38600 mg/kg were observed in a survey dealing with 40 different samples of different peppermint cultivars (Guédon and Pasquier 1994). Our findings show, that horsemint may be a useful source of polyphenolic antioxidants, too. The high amount of hesperidin and the lack of caffeic acid in higher ratios was unexpected in comparison with literature of ML (Dudai *et al.* 2006). The presence of diosmin and its concentration intervals are also similar to the values of peppermint detected in the aforementioned study (0.03-0.47 m/m %, ca. 300-4700 mg/kg DW). A medium, significant correlation between rosmarinic acid content and antioxidant properties of samples was determined while the connection of antioxidant power and content of cynaroside was not significant. These results indicate that identification and quantification of Unknown A and B in the extracts would be necessary to decide how flavonoids and other polyphenols beyond rosmarinic acid contribute to the antioxidant properties of horsemint. The investigation discussed above is part of the first phytochemical survey of horsemint populations in Hungary. The high heterogeneity of concentration of the phenolics in the horsemint samples supports the assumption that the wild-collected material is not suitable for gaining extracts of standard quality. Selection, propagation and cultivation of ML chemotypes bearing the required polyphenol composition may provide a sustainable alternative.

Acknowledgements: Authors gratefully acknowledge support of the grant EFOP-3.6.1-16-2016-00001 'Complex improvement of research capacities and services at Eszterhazy Karoly University' and the valuable support of Bükk National Park in this project.

REFERENCES

AHMED, D., KHAN, M.M. & SAEED, R. (2015). Comparative analysis of phenolics, flavonoids, and antioxidant and antibacterial potential of methanolic, hexanic and aqueous extracts from *Adiantum caudatum* leaves. *Antioxidants* **4**: 394–409. https://doi.org/10.3390/antiox4020394

ALI, M. S., SALEEM, M., AHMAD, W., PARVEZ, M. & YARUDANI, R. (2002). A chlorinated monoterpene ketone, acylated glycosides of beta-sitosterol and a flavonoid glycoside of *Mentha longifolia*. *Phytochemistry* **59**: 889–895. https://doi.org/10.1016/S0031-9422(01)00490-3

BAHADORI, B. M., ZENGIN, G., BAHADORI, S. & DINPARAST, L. (2018). Phenolic composition and functional properties of wild mint (*Mentha longifolia* var. *calliantha* (Stapf) Briq.). *International Journal of Food Properties* **21**: 183–193. https://doi.org/10.1080/10942912.2018.1440238

BAŞER, K.H.C., KÜRKÇÜOĞLU, M., TARIMCILAR, G. & KAYNAK, G. (1999). Essential oils of *Mentha* species from Northern Turkey. *Journal of Essential Oil Research* **11**: 579–588. https://doi.org/10.1080/10412905.1999.9701218

BENEDEC, D., VLASE, L., ONIGA, I., MOT, A.C., SILAGHI-DUMITRESCU, R., HANGANU, D., TIPERCIUC, B.T. & CRISAN, G. (2013). LC-MS analysis and AO activity of phenolic compounds from two indigenous species of *Mentha*. Note I. *Farmacia* **61**: 262–267.

BENZIE, I.F.F. & STRAIN, J.J. (1996). The ferric reduction ability of plasma (FRAP) as a measure of "Antioxidant Power": the FRAP assay. *Analytical Biochemistry* **239**: 70–76. https://doi.org/10.1006/abio.1996.0292

BORS, W., HELLER, W., MICHEL, C. & SARAN, M. (1990). Flavonoids as antioxidants: Determination of radical-scavenging efficiencies. *Methods in Enzymology* **186**: 343–355. https://doi.org/10.1016/0076-6879(90)86128-I

BOURWIEG, D., & POHL, R. (1973). The flavonoids of *Mentha longifolia. Planta Medica* **24**: 304–314.

DAMIEN-DORMAN, H. J., KOŞAR, M., KAHLOS K, HOLM, Y., HILTUNEN, R. (2003). Antioxidant properties and composition of aqueous extracts from *Mentha* species, hybrids, varieties, and cultivars. *Journal of Agricultural and Food Chemistry* **51**: 4563–4569. https://doi.org/10.1021/jf034108k

DUDAI, N., SEGEV, D., HAVKIN-FRENKEL, D. & ESHEL, A. (2006). Genetic variation of phenolic compounds content, essential oil composition and antioxidative activity in Israel-grown *Mentha longifolia* L. *Acta Horticulturae* **709**: 69–78. https://doi.org/10.17660/ActaHortic.2006.709.8

FIALOVÁ, S., TEKEL'OVÁ, D., MRLIANOVÁ, M. & GRANČAI, D. (2008). The determination of phenolic compounds and antioxidant activity of mints and balms cultivated in Slovakia. *Acta Facultatis Pharmaceuticae Universitatis Comenianae* **54**: 96–102.

GHOULAMI, S., IL-IDRISSI, A. & FKIH-TETOUANI, S. (2001). Phytochemical study of *Mentha longifolia* of Morocco. *Fitoterapia* **72**: 596–598. https://doi.org/10.1016/S0367-326X(01)00279-9

GUÉDON, D.J. & PASQUIER, B.P. (1994). Analysis and distribution of flavonoid glycosides and rosmarinic acid in 40 *Mentha x piperita* clones. *Journal of Agricultural and Food Chemistry* **42**: 679–684. https://doi.org/10.1021/jf00039a015

GÜLLÜCE, M., SAHIN, F., SOKMEN, M., OZER, H., DAFERERA, D., SOKMEN, A., POLISSIOU, M., ADIGUZEL, A., & OZKAN, H. (2007). Antimicrobial and antioxidant properties of essential oils and methanolic extract from *Mentha longifolia* ssp. *longifolia. Food Chemistry* **103**: 1449–1456. https://doi.org/10.1016/j.foodchem.2006.10.061

HAJIMEHDIPOOR, H., SHAHRESTANI, R. & SHEKARCHI, M. (2014). Investigating the synergistic antioxidant effects of some flavonoid and phenolic compounds. *Research Journal of Pharmacognosy* **1**: 35–40.

HAJLAOUI, H., TRABELSI, N., NOUMI, E., SNOUSSI, M., FALLAH, H., KSOURI, R. & BAKHROUF, A. (2009). Biological activities of the essential oils and methanol extract of two cultivated mint species (*Mentha longifolia* and *Mentha pulegium*) used in the Tunisian folkloric medicine. *World Journal of Microbiology and Biotechnology* **25**: 2227–2238. https://doi.org/10.1007/s11274-009-0130-3

HANAFY, D.M., PRENZLER, P.D., BURROWS, G.E., RYAN, D., NIELSEN, S., EL SAWI, S.A., EL ALFY, T.S., ABDELRAHMAN, E.H., HASSAN, K., OBIED, H.K. (2017). Biophenols of mints: Antioxidant, acetylcholinesterase, butyrylcholinesterase and histone deacetylase inhibition activities targeting Alzheimer's disease treatment. *Journal of Functional Foods* **33**: 345–362. https://doi.org/10.1016/j.jff.2017.03.027

HAWRYŁ, M. A., NIEMIEC, M., SŁOMKA, K., WAKSMUNDZKA-HAJNOS, M. & SZIMCZAK, G. (2016). Micro-2D-TLC Separation of phenolics in some species of mint and their fingerprints on diol bonded polar stationary phase. *Acta Chromatographica* **28**: 119–127. https://doi.org/10.1556/achrom.28.2016.1.9

IQBAL, T., HUSSAIN, A. I., SHAHID CHATHA, S. A., NAQVI, S. A. R. & HUSSAIN BOKHARI, T. (2013). Antioxidant activity and volatile and phenolic profiles of essential oil and different extracts of wild mint (*Mentha longifolia*) from the Pakistani flora. *Journal of Analytical Methods in Chemistry* **2013**: ID 536490. https://doi.org/10.1155/2013/536490

JAHAN, N., MALIK, A. & MUHAMMAD, P. (2001). New flavonoid from *Mentha longifolia. Heterocycles* **55**: 1951–1956. https://doi.org/10.3987/COM-01-9281

KARABEGOVIĆ, I. T., NIKOLOVA, M., VELICKOVIĆ, D., STOJIČEVIĆ, S., VELJKOVIĆ, V. & LAZIĆ, M. (2011). Comparison of antioxidant and antimicrobial activities of methanolic extracts of the *Artemisia* sp. recovered by different extraction techniques *Chinese Journal of Chemical Engineering* **19**: 504–511. https://doi.org/10.1016/S1004-9541(11)60013-X

KOŞAR, M., DAMIEN DORMAN, H.J., BAŞER, K.H.C. & HILTUNEN, R. (2004). Screening of free radical scavenging compounds in water extracts of *Mentha* samples using a postcolumn derivatization method *Journal of Agricultural and Food Chemistry* **52**: 5004–5010. https://doi.org/10.1021/jf0496189

KŐSZEGINÉ SZALAI, H., TÖRÖK, I., RÁFLINÉ ROMVÁRI, ZS., NAGY, A., FEJÉR, I., KERTÉSZ, P., POSGAYNÉ KOVÁCS, E., & VANKÓ, É. (eds.) (2003). *Magyar Gyógyszerkönyv* I–II. (Pharmacopoea Hungarica). Medicina Könyvkiadó Rt., Budapest, 2504+76 p.

KRZYZANOWSKA, J., JANDA, B., PECIO, L., STOCHMAL, A. & OLESZEK, W. (2011). Determination of polyphenols in *Mentha longifolia* and *M. piperita* field-grown and in vitro plant samples using UPLC-TQ-MS. *Journal of AOAC International* **94**: 43–50. https://doi.org/10.1093/jaoac/94.1.43

MEDINA-TORRES, N., AYORA-TALAVERA, T., ESPINOSA-ANDREWS, H., SÁNCHEZ-CONTRERAS, A. & PACHECO, N. (2017). Ultrasound assisted extraction for the recovery of phenolic compounds from vegetable sources. *Agronomy* **7**(3): 47. https://doi.org/10.3390/agronomy7030047

MIMICA-DUKIĆ, N., JAKOVLJEVIC, V., POPOVIC, M., SZABO, A. & GAŠIC, O. (1999). Pharmacological studies of *Mentha longifolia* phenolic extracts II. Hepatoprotective activity. *Pharmaceutical Biology* **37**:221–224. https://doi.org/10.1076/phbi.37.3.221.6306

MURAD, H.A.S., ABDALLAH, H.M., & ALI, S.S. (2016). *Mentha longifolia* protects against acetic-acid induced colitis in rats. *Journal of Ethnopharmacology* **190**: 354–361. https://doi.org/10.1016/j.jep.2016.06.016

ONISZCZUK, A. & PODGÓRSKI, R. (2015). Influence of different extraction methods on the quantification of selected flavonoids and phenolic acids from *Tilia cordata* inflorescence. *Industrial Crops and Products* **76**: 509–514. https://doi.org/10.1016/j.indcrop.2015.07.003

PANIWNYK, L., CAI, H., ALBU, S., MASON, T.J., & COLE, R. (2009). The enhancement and scale up of the extraction of anti-oxidants from *Rosmarinus officinalis* using ultrasound. *Ultrasonics Sonochemistry* **16**: 287–292. https://doi.org/10.1016/j.ultsonch.2008.06.007

PARK, Y.J., BAEK, S.A., CHOI, Y., KIM, J.K. & PARK, S.U. (2019). Metabolic profiling of nine *Mentha* species and prediction of their antioxidant properties using chemometrics. *Molecules* **24**: 258–273. https://doi.org/10.3390/molecules24020258

PATONAY, K., KORÓZS, M., MURÁNYI, Z. & PÉNZESNÉ KÓNYA, E. (2017). Polyphenols in northern Hungarian *Mentha longifolia* (L.) L. treated with ultrasonic extraction for potential oenological uses *Turkish Journal of Agriculture and Forestry* **41**: 208–217. https://doi.org/10.3906/tar-1701-61

PATONAY, K., SZALONTAI, H., CSUGÁNY, J., SZABÓ-HUDÁK, O., HELGA PÉNZESNÉ, KÓNYA E. & ZÁMBORINÉ NÉMETH, É. (2019). Comparison of extraction methods for the assessment of total polyphenol content and in vitro antioxidant capacity of horsemint (*Mentha longifolia* (L.) L.) *Journal of Applied Research on Medicinal and Aromatic Plants* **15**: 100220. https://doi.org/10.1016/j.jarmap.2019.100220

PEREIRA, O.R., & CARDOSO, S.M. (2013). Overview of *Mentha* and *Thymus* polyphenols. *Current Analytical Chemistry* **9**: 382–396. https://doi.org/10.2174/1573411011309030008

SHARAF, M., EL-ANSARI, M., & SALEH, N.A.M. (1999). Flavonoid glycosides from *Mentha longifolia. Fitoterapia* **70**: 478–483. https://doi.org/10.1016/S0367-326X(99)00062-3

SHAROPOV, F.S., SULAIMANOVA, V.A., & SETZER, W.N. (2012). Essential oil composition of *Mentha longifolia* from wild populations growing in Tajikistan. *Journal of Medicinally Active Plants* **1**: 76–84. https://doi.org/10.7275/R5736NTN

SHEN, D., PAN, H.M., WU, Q.L., PARK, C.H., RODOLFO JULIANI, H., HO, C.T. & SIMON, J.E. (2011). A rapid LC/MS/MS method for the analysis of nonvolatile antiinflammatory agents from *Mentha* spp. *Journal of Food Science* **76**(6): 900–908. https://doi.org/10.1111/j.1750-3841.2011.02281.x

SIMON, T. (1994). *A magyarországi edényes flóra határozója: Harasztok – virágos növények.* (Handbook of the Hungarian vascular flora. Ferns and seed plants). Nemzeti Tankönyvkiadó Rt., Budapest, 845 p.

STANISLAVLJEVIĆ, D.M., STOJIČEVIĆ, S.S., ĐORĐEVIĆ, S.M., ZLATKOVIĆ, B.P., VELIČKOVIĆ, D.T. & KARABEGOVIĆ, I.T. (2012). Antioxidant activity, the content of total phenols and flavonoids in the ethanol extracts of *Mentha longifolia* L. dried by the use of different techniques. *Chemical Industry and Chemical Engineering Quarterly* **18**: 411–420. https://doi.org/10.2298/CICEQ110919017S

THE EUROPEAN PARLIAMENT AND COUNCIL (2009). Directive 2009/32/EC of the European Parliament and Council of 23 April 2009 on the approximation of the laws of the Member States on extraction solvents used in the production of foodstuffs and food ingredients. *Official Journal of the European Union* **141**: 3–7.

THE EUROPEAN PARLIAMENT AND COUNCIL (2010). Directive 2010/59/EC of the European Parliament and Council of 26 August 2010 amending Directive 2009/32/EC of the European Parliament and of the Council on the approximation of the laws of the Member States on extraction solvents used in the production of foodstuffs and food ingredients. *Official Journal of the European Union* **225**: 10–12.

TUCKER, A.O. & NACZI, R.F.C. (2007). *Mentha: An overview of its classification and relationships.* In: LAWRENCE, B.M.(ed.): Mint. *The genus* Mentha – *Medicinal and* *aromatic plants – Industrial profiles*. CRC Press, Taylor and Francis Group, Boca Raton, pp. 3–39.

WATERHOUSE, A.L. (2002). Determination of total phenolics. *Current Protocols in Food Analytical Chemistry* **6**(1): I1.1.1–I1.1.8. https://doi.org/10.1002/0471142913.fai0101s06

(submitted: 07.01.2020, accepted: 09.09.2020)

Acta Biologica Plantarum Agriensis 8(1): 69–84 (2020)
ISSN 2061-6716 (Print), 2063-6725 (Online) *Research article*
http://abpa.ektf.hu/
https://doi.org/10.21406/abpa.2020.8.1.69

NOTES ON THE BRYOPHYTES OF MADAGASCAR 2. NEW LIVERWORT AND HORNWORT RECORDS

Tamás Pócs

Institute of Biology, Eszterházy Károly University, Eger, Pf. 43, Hungary, H-3301;
E-mail: pocs.tamas33@gmail.com

Abstract: During the identification of the epiphyllous liverworts collected in Madagascar for the purposes of description of foliicolous communities, 11 species turned out as new to the flora of the island, one of them (*Cololejeunea ocelloides*) to the whole of Africa. In addition, 8 other species hitherto known only from one place in Madagascar became known from other localities. Finally two new synonyms were established.

Keywords: biodiversity, endemics, Indian Ocean islands, Madagascar, new species

INTRODUCTION

The author with colleagues from the Eger College and other institutions and with his late wife visited Madagascar during 1990, 1994, 1998 and in 2004. The results of these collecting trips are only partly published (Allen and Magill 2007, Bechteler *et al.* 2015, Dong *et al.* 2012, 2013, Dorr 1997, He and Grolle 2001, Heinrichs *et al.* 2005, 2014, Hodgetts 2008, Jones 1992, Kis and Pócs 2006, Lee *et al.* 2015, Lee and Pócs 2018, Lindner *et al.* 2004, Orbán 1995, 2000, Pócs 1995, 1997, 2001, 2004, 2006a, 2006b, 2006c, 2010a, 2010b, 2011a, 2011b, 2011c, 2020a, 2020b, 2021, Pócs *et al.* 2015, 2016, 2018, 2019, Pócs and Geissler 2002, Pócs and Sass-Gyarmati 2006, Pócs and Schäfer-Verwimp 2006, Pócs and Váňa 2002, 2015, Reeb *et al.* 2018, 2019, Sass-Gyarmati 2001, 2017, Sass-Gyarmati and Pócs 2002, 2019, Szabó and Pócs 2016, Wilding 2016), which were summarized until 2011 in Marline *et al.* (2012). Another part of the specimens is still under identification by himself and by specialists of the concerned groups [S.R. Gradstein (GOET): *Plagiochila*, G.E. Lee: *Lejeunea*, A.E. Newton (BR): Rutenbergiaceae,

A. Sass-Gyarmati (EGR): *Bazzania,* J. Wilbraham (BM): Orthotrichaceae].

MATERIALS AND METHODS

In the following the author wishes to display records from his most recent investigations done mostly on epiphyllous liverwort communities. Some of them are widely distributed in the continental part of Africa but somehow were not yet reported from Madagascar. For the nomenclature the 'World checklist of hornworts and liverworts' (Söderström *et al.* 2016) was considered. In the Madagascar and total African distribution apart from smaller reports we took in account the publications of Marline *et al.* (2012) and of Wigginton (2018). Specimens of the collection are deposited in the herbarium of Eszterházy Károly University in Eger, Hungary (EGR), in the Tsimbazaza Botanic Garden Herbarium (TAN), partly also in the Missouri Botanic Garden Herbarium (MO) and in the Herbarium of Field Museum in Chicago (F). Our present records originate from only a few localities and almost all are epiphylls (if not otherwise stated), therefore it seemed to be reasonable to enumerate the localities first in details than in the list just to mention them by their collecting numbers:

The collecting localities

90111. Antsiranana prov., Sava reg., Réserve Intégrale Nationale de Marojezy. Lowland rainforest near the first campsite in Andampibe valley, 200-500 m, with giant trees. *T. Pócs, with R.E. Magill & C. LaFarge-England, 23–24. March 1990.*

90113. Antsiranana prov., Sava reg.,réserve Intégrale Nationale de MAROJEZY. Montane rainforest on the ridge N of Andampibe falls, 780-1050 m., trees 10-20 m high with many epiphytes. *T. Pócs, with R.E. Magill & C. LaFarge-England, 24–29. March 1990.*

90114. Antsiranana prov., Sava reg., réserve Intégrale Nationale de Marojezy. Elfin forest on the SE ridge near camp III, at 1830 m alt. *T. Pócs, with R.E. Magill & C. LaFarge-England, 26–28. March 1990.*

9448. Antsiranana prov., Sava reg. Masoala Peninsula. Submontane rainforest on the W slopes E of AMBANIZANA village, 450-600 m, 15°37'S, 49°59'30"E. *T. Pócs, G. Kis, 9. Sept. 1994.*

9472. Fianarantsroa Prov., Haute Matsiatra reg., Parc national de l'Andringitra . Montane rainforests on the W side of KOROKORO river, around camp II, 750-1000 m, 22°13'S, 47°01-02'E. *T. Pócs, G. Kis & A. Szabó, 20–23. Sept. 1994.*

9473. Fianarantsroa prov., Haute Matsiatra reg., Parc national de l'Andringitra. Mossy montane rainforest along a W tributary of KOROKORO river, near camp III, 1000-1270 m, 22°12'40"S, 47°0 E . *T. Pócs, G. Kis & A. Szabó, 20–23. Sept. 1994.*

9474. Fianarantsroa prov., Haute Matsiatra, Parc national de l'Andringitra. Mesic montane evergreen forest on the ridge E of camp III, 1270-1450 m, with many climbing bamboos (*Nastus* sp.), 22°12'30"S, 47°0'15"E. *T. Pócs, 22. Sept. 1994.*
9875. Toamasina prov., Analanjirofo reg., Mananara Nord Biosphere Reserve. Perhumide coastal dune forest with rich moss layer on the sandy ground, 1 km W of Antanambe village, 5 m, 16°25.8'S, 49°50.2'E. *K., S. & T. Pócs and A. Szabó, 13. Aug. 1998.*
9877. Toamasina prov., Analanjirofo reg., Mananara Nord Biosphere Reserve and National Park. Lowland rainforest on the NW slope of BEHAFOTRA hill, 250-300 m, with 3500 mm annual rainfall, 16°27.1-3'S, 49°47.6-9'E. *K. & T. Pócs and A.& E. Szabó, 14–16. Aug. 1998.*
9883. Toamasina prov., Atsinanana reg.. Coastal dune forest with stilt rooted *Uapaca* trees and *Memecylon* bushes along the road between Andovoranto and Ambila-Lemaitso, 4-5 m , 18°54.2-55.4'S, 49°7.3-6'E. *S. & T. Pócs and A. Szabó, 22. Aug. 1998.*
9887. Toamasina prov, Atsinanana reg. Peat forest & woodland often with thick *Sphagnum* layer on coastal white sand, with shallow lakes, between Pampangalana Canal and the ocean, 5 km N of Andovoranto, between Andombo and Andovoa, 10 M , 18°54.1'S, 49°7.24'E. *Pócs And A. Szabó, 24. Aug. 1998.*
9888. Toamasina prov., Atsinanana reg. Open dune forest dominated by *Pandanus* sp. and *Garcinia* sp. on coastal white sands between Pampangalana canal and the ocean, 5 km N of Andovoranto, between Andombo and Andovoa, at 10 m alt. 18°54.1'S, 49°7.24'E. *T. Pócs and A. Szabó, 24. Aug. 1998.*
9890. Toamasina prov., Alaotra-Mangoro, reg. Maromizaha forest, mossy montane rainforest with bamboo (*Nastus* sp.) undergrowth on the summit ridge of Mt. Maromizaha, south of the Andasibe Nat. Park and the Antananarivo – Toamasina road, 2 km SW of Anevoka village, 1080-1214 m , 18°57.8'S, 48°27.5'E. *S. & T. Pócs, 26. Aug. 1998*.
04116. Fianarantsoa prov. Forest Reserve on the E side of Antananarivo – Fianarantsoa road, 5 km N of Ambohima-Hasoa town, S of Ialatsara village. *Eucalyptus* plantation replacing montane rainforest, 1360 m alt. 21°04.69'S, 47°12.70'E. *S.& T. Pócs, N. Rakotonirina, R. Ranaivojaona and Z. Tuba, 24 July 2004.*
04120. Fianarantsoa prov., Atsinanana reg;, parc National de Ranomafana. Sahamalaotra Forest, 1.5 km E of Vohiparara village. Montane rainforest on undulating terrain, 1120-1160 m,21°14.36'S, 47°23.67'E. *T. Pócs, N. Rakotonirina & R. Ranaivojaona, 26 July 2004*.
04126. Fianarantsoa prov., Atsinanana reg; parc National de Ranomafana. Along the N side of main road at the head the big waterfalls on the NAMORONA river. Dripping granitic cliffs surrounded by montane rainforest, 1080-1150 m, 21°14.70-86'S, 47°23.82'E. *S. & T. Pócs, 29 July 2004*.
04128. Fianarantsoa prov., Atsinanana reg;, parc National de Ranomafana. On the summit ridge of Mt. Vatolampy (with a telecom aerial), 2.3 km W of Ambatolahy village. Disturbed, relatively open montane rainforest with 5-10 (-20)m high trees, rich in micro-epipytes, 1200 m, 21°15.10*'S,* 47°24.44'E *S.& T. Pócs, 30 July 2004.*
04130. Fianarantsoa prov., Atsinanana reg;, beyond the W boundary of Ranomafana National park. 1.6 km E of Ambatovaku Avaratra village, on the S side ridge of Mt. Namatoana. Dense, but not too shady microphyllous elfin forest of

thin, 6-8 (10) m tall trees, 1250 m,. 21°17.10'S, 47°19.14'E. S. & *T. Pócs, K. Tuba, 24 July 2004.*

RESULTS

Records new for Madagascar

Caudalejeunea africana (Steph.) Steph. **90111/BB, 9875/CC**. Widespread epiphyll in continental Africa, new to the Indian Ocean islands, including Madagascar.

Cheilolejeunea adnata (Lehm.) Grolle var. ***autoica*** Gradst. & Ilk.-Borg. (Syn.: *Cheilolejeunea larsenii* Mizut., C*heilolejeunea exinnovata* E.W. Jones (fide Bastos & Gradstein 2020). **90111/CE,** a pantropical species hitherto known from several places in continental West-Africa and Zimbabwe under the name of *C. larsenii*, but new for all East African islands (Wigginton 2018). It is characterized by its bottle shaped lobule and the the variability of its teeth often crossing each other.

Cheilolejeunea ventricosa (Schiffn. ex P.Syd.) Xiao L. He. **9472/BV**, on dry, dead leaf. Malesian-Pacific species known in Africa only from Mauritius (Pócs 1997 as *Pycnolejeunea ventricosa*, Pócs *et al.* 2019).

Cololejeunea haskarliana (Lehm. et Lindenb.) Schiffn**. 90113/FL, 9472/AS, 9473/GV, 9474/AA.** A widespread species in Asia and in the Pacific, but in Africa only known from the Seychelles and from Réunion Island.

Cololejeunea ocelloides (Hook.) Mizut. (Syn.: *Cololejeunea leonidens* Benedix). **90114/KE.** Hitherto known as a Malesian species known from Sumatra to New Guinea (Pócs and Piippo 2011), new for Africa.

Cololejeunea pteroporum Tixier **90111/BR.** A species described and hitherto known only from Rodrigues Island (Tixier 1985, Grolle 1995), new for Madagascar.

Dendroceros borbonicus Steph. **9888/F.** Quite widespread in Mauritius, Réunion and the Seychelles, not yet known from Madagascar. A species equally occurring on leaves, bark and twigs in wet habitats.

Lejeunea exilis (Reinw. et al.) Grolle **90114/HA.** A widespread Indomalesian-Pacific species, known only from Réunion in Africa, new for Madagascar.

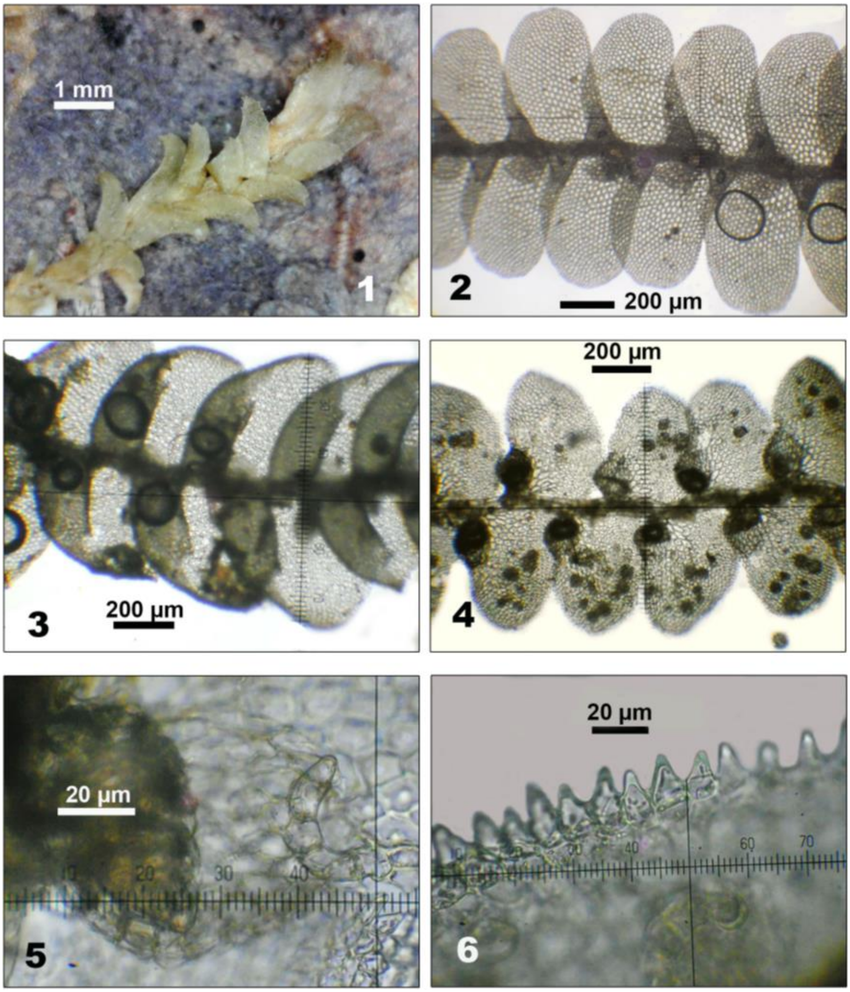


Plate I. Figure 1. *Caudalejeunea africana* (Steph.) Steph. (from *Pócs & Szabó 9875/CC*). **Figure 2.** *Cheilolejeunea andnata var. autoica* Gradst. & Ilkiu-Borges, ventral view (from *Pócs 90111/CE*). **Figure 3.** *Cheilolejeunea ventricosa* (Schiffn. ex P.Syd.) Xiao L. He, ventral view (from *T. Pócs & Kis 9472/BV*). **Figures 4–6.** *Cololejeunea haskarliana* (Lehm. & Lindenb.) Steph., 4 = ventral view, 5 = lobule with crossing teeth, 6 = lobe margin (from *Pócs & Kis 9472/AS*).

Leptolejeunea maculata (Mitt.) Schiffn. **94111/BC,BW, 9883/BA.** A palaeotropic species, widespread in mainland Africa and tropical Asia and the Pacific, known also from the Seychelles, Réunion and Rodrigues Island but not yet in Madagascar (Müller and Pócs 2002).

Microlejeunea dispar Jov.-Ast **90113/FO.** Endemic of the East African islands, known only from Réunion, Mauritius (Grolle 1995, Marline *et al.* 2012, Wigginton 2018).

Radula flaccida (Lindenb.) Gottsche **90111/BH, 9883/BR**. A common Afro-American epiphyll. From the Indian Ocean region it was known only from Anjouan (Dzouani) Island in the Comores (Castle 1939).

Records of species known before only from one area in Madagascar

Cololejeunea mariana Tixier **90111/CG.** Known only from its type locality in St. Marie Island.

Colura obvoluta Jov.-Ast **9875/CN.** Apart from its type locality in St. Marie Island of Madagascar it was known only from the Seychelles Islands (Pócs 1995).

Colura humbertii Jov.-Ast **9890/DB.** In Pócs (2020b) this record by mistake under the name of *C. calyptrifolia* was communicated. **04126/QC.** In Madagascar it is known only from its type locality (Jovet-Ast 1954). Since became known from Mauritius and Réunion (Grolle 1995) and from Tanzania (Pócs 2020b).

Diplasiolejeunea magnistipula Tixier **04128/AT.** The species was described from Cameroon (Tixier 1975) and later only one other record was published from the Lokoho Basin in northern Madagascar (under the synonym of *Diplasiolejeunea drepanolejeuneoides*, Tixier 1977).

Drepanolejeunea cultrella (Mitt.) Steph. **04128/AT.** On dry, dead leaf. A very widespread tropical African species known only from one locality in north-western Madagascar (Pócs and Geissler 2002).

Leptolejeunea epiphylla (Mitt.) Steph. **94111/BC.** Palaeotropical species, widespread from tropical Africa through tropical Asia to the Pacific. It is known in the Indian Ocean Islands from the Seychelles, Comores and Mauritius and in Madascar only from the Manongarivo Reserve (Pócs and Geissler 2002).

Otolejeunea moniliata Grolle. **9448/K, 9472/AD, AZ** epiphyllous, **BW** On dry, dead leaves. It was previously known only from the type area of Marojezy Reserve in northern Madagascar. Now it becomes known also from the Masoala Peninsula and further south in the Andringitra mountains. It is worth to mention that the original description does not mention the scattered ocelli, which can be observed only on fresh material (and also seen on the micrograph of Bechteler *et al.* 2015: fig. 1c and f). Furthermore there are specimens with denticulate lobes, where the teeth are tipped by hyaline papillae.

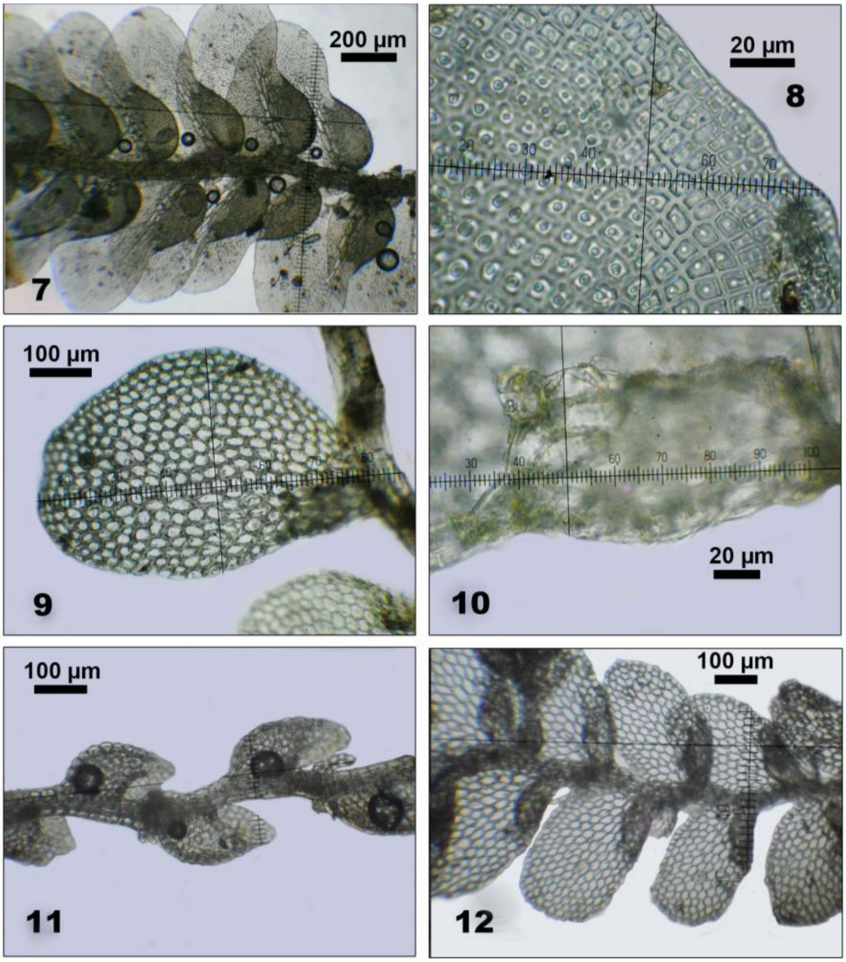


Plate II. Figures 7–8. *Cololejeunea ocelloides* (Hook.) S.Hatt. 7 = ventral view, 8 = lobe margin (from *Pócs et al. 90114/KE*). **Figures 9–10.** *Cololejeunea pteroporum* Tixier, 9 = leaf, dorsal view, 10 = lobule, ventral view (from *Pócs et al. 90111/BR).* **Figure 11.** *Lejeunea exilis* (Reinw. et al.) Grolle, ventral view (from *Pócs et al. 90114/HA*). **Figure 12.** *Leptolejeunea maculata* (Mitt.) Schiffn., ventral view (from *Pócs & Szabó 9883/BA*).

Radula pinnata Pócs **9875/CK.** New record of the recently described species (Pócs 2020a) at 5 km distance from the type locality. It was supposed that it is mainly corticolous and only facultative or incidentally epiphyll, but at the new locality *R. pinnata* occurred also only on leaf surface.

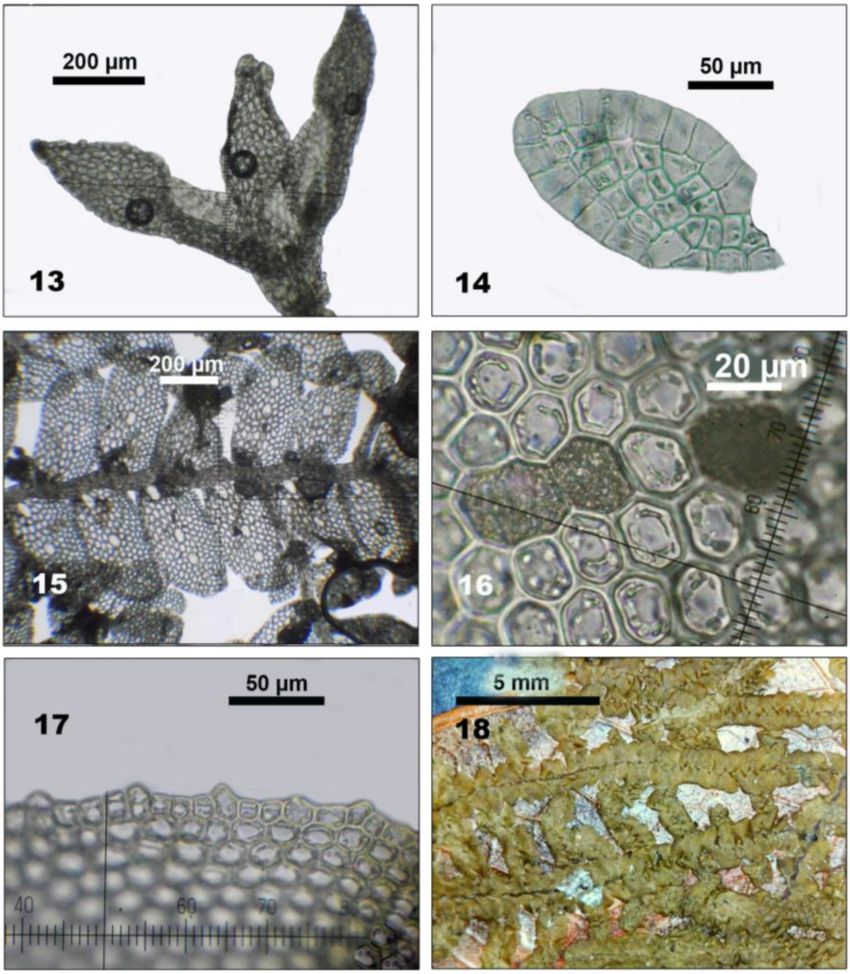


Plate III. Figures 13–14. *Colura obvoluta* Jov.-Ast , 12 = ventral view, 14 = unseparable valve (from *Pócs & Szabó 9875/CN*). **Figure 15.** *Leptolejeunea maculata* (Mitt.) Schiffn., ventral view from *Pócs et al. 90111/BC*). **Figures 16–17.** *Otolejeunea moniliata* Grolle. 16 = scattered mid lobe ocelli, 17 = sparsely denticulate lobe margin (from *Pócs et al. 9472/AZ*). **Figure 18.** *Radula pinnata* Pócs (from *Pócs et al. 9875/CK*).

New synonyms of Madagascar liverworts

During the study of many epiphyllous material from different Madagascar localities was found that some species described on the base of only few specimens are interconnected by transitional forms. Hence is impossible to demarcate species limits between them. On this base the following new synonyms were established:

Cololejeunea cuneata (Lehm. et Lindenb.) Herzog, Bot. Not. 100 (4): 320, 1947 (Herzog 1947).
Basionym: *Jungermannia cuneata* Lehm. et Lindenb., Nov. Stirp. Pug. 4: 56, 1832 (Lehmann 1832). Type: MAURITIUS (G).
Syn.: *Leptocolea subciliata* (Mitt.) E.W.Jones, Trans Brit. Bryol. Soc. 2 (E.W. Jones 1953). Type: RODRIGUEZ I. *I.B. Balfour, 1874* (BM).
Syn. nov.: *Cololejeunea cristata* (Steph.) R.M. Schust. Beih. Nova Hedwigia 9: 173, 1963 (Schuster 1963).
Basionym: *Physocolea cristata* Steph., Sp. Hepat. (Stephani) 5: 869, 1916 (Stephani 1916).
Type: MADAGASCAR *Leloutre, 13 July 1898* (G).
Other synonyms see in Tixier (1985).
Tixier (1985: 52) had already mentioned the difficulties to distinguish *Cololejeunea cristata* from *Cololejeunea cuneata* and from some other species. The difference between the two species is established on the lobule dentition, whether 1 or 2 lobule teeth are present. In our collections (90111/BQ, 90113/FU, 9473/HJ, 9875CH, 9883/BC, 9887/Y), even within one specimen, both cases occur and the development of second tooth is very variable, from very well developed to obsolete or nothing. This indicates that the two species are practically not separable.

Diplasiolejeunea kraussiana (Lindenb.) Spruce, Sp. Hepat. (Stephani) 5: 919, 1916 (Stephani 1916).
Basionym.: *Lejeunea kraussiana* Lindenb., Syn. Hepat. 3: 393, 1845, (Gottsche *et al.* 1845).
Type: SOUTH AFRICA, near Knysna, in terries “Outaniquanis”, *Krauss* (W, Lindenb. Hep. 6757, isotypes BM (H 10003), G).
Syn. nov.: *Diplasiolejeunea auriculata* Tixier, Rev. Bryol. Lichenol. 45 (2): 210, 1979 (Tixier 1979).
Type: MADAGASCAR, Fianarantsdoa, forêt du Tsarafidy, *Tixier 8908* (holotype PC).

According to Tixier (1979:212) *D. auriculata* is close to *D. kraussiana.* To distinguish the two species, he used the formation of lobule apex and the shape of perianths. In the original diagnosis of *Diplasiolejeunea kraussiana* (Lindenberg in Gottsche *et al.* 1845), the lobule apex is described as "*lobulo magno bidentato (dente margini propinquo maiore) inflexo*". Tixier writes in the diagnosis of *Diplasiolejeunea auriculata*: "*dente media cum duobus cellulis ad apicem*". The T shaped teeth, like for *Diplasiolejeunea cornuta* Steph., is a very unreliable character occurring on many lobules indeed and missing in others, within the same or in other plants. The perianth shape is also very variable, even on the Tixier's drawings of (1979: 211-212, figs. 1:11 and fig 2: 10–12) from obcordate with auriculate wings to simple obconic with narrow, straight wings. According to the author's observation several specimens are transitional between the two. The combinations of these two characters at different degree can occur. Hence there is no reason to distinguish them at the species level. For example in our specimen 04116/H the shape of the perianth is typically "auriculate", while the majority of the apices of the lobule teeth are simple.

DISCUSSION

The diversity of Madagascar bryoflora is very high, compared even to continental Africa. From the near 1200 bryophyte species almost 400 are liverworts and hornworts, although the greater part of the island is under high anthropogenic influence. A great proportion of the former primary vegetation types are lost. Many rainforests, which house the highest proportion of the bryophyte species, are already converted to pastures, to secondary savoka bushes, as the result of shifting, slash and burn cultivation and of illegal timber cutting. Even so, careful collection and identification work every year brings to light many new records and even taxa new to science. Therefore the existing protected areas, as national parks and biosphere reserves have great significance and merit international aid to be saved.

As it is seen from the above enumeration, the bryoflora of Madagascar is still inadequately known. From a moderate amount of collections 11 species turned out to be new for the flora of the island and the same amount were known only from one locality. It should be mention, that from these few localities already 7 species were described as new to science and further 4 are waiting for description. The herbarium of Eszterházy Károly University in Eger (EGR) has an immense amount of collected specimens (mostly shared with the Herbarium of Tsimbazaza Botanical garden, Antananarivo: TAN). We are looking for specialists of the different groups, especially of mosses and are ready to send the specimens to them for identification and revision, as we already did before.

Acknowledgement – The author wishes to express his gratitudes to all instutions and sponsoring agencies mentioned in his last Madagascar contribution (Pócs 2020b), who supported his travel costs and collecting activities. He is also grateful to his late wife and to his Hungarian and Malagasy colleagues, who participated in the collecting expeditions and finally to the careful reviewers, Catherine Reeb (Sorbonne Université and PC) and to Alfons Schäfer-Verwimp (Herdwangen-Schönach, Germany) for their useful advises and corrections.

REFERENCES

Allen, B. & Magill, R. (2007). A revision of *Orthotrichella* (Neckeraceae). *The Bryologist* **110**(1):1–45.

Bastos, C.J.P. & Gradstein, S.R. (2020). The genus *Cheilolejeunea* (Marchantiophyta: Lejeuneaceae) in tropical America. *Nova Hedwigia* **111**(3-4): 287–335. https://doi.org/10.1127/nova_hedwigia/2020/0596

Bechteler, J., Lee, G.E., Schäfer-Verwimp, A., Pócs, T., Peralta, D.F., Renner, M.A.M., Schneider, H. & Heinrichs, J. (2015). Towards a monophyletic classification of Lejeuneaceae IV: reinstatement of *Allorgella*, transfer of Microlejeunea aphanella to *Vitalianthus* and refinements of the subtribal classification. *Plant Systematics and Evolution* **302**: 187–201. https://doi.org/10.1007/s00606-015-1252-8

Castle, H. (1939). A revision of the genus *Radula*. Part II. Subgenus *Acroradula*. Section 1. *Epiphyllae*. *Annales Bryologici* **12**: 21–47.

Dong, S., Schäfer-Verwimp, A., Meinecke, Ph., Feldberg, K., Bombosch, A., Pócs, T., Schmidt, A.R., Reitner, J., Schneider, H. & Heinrichs, J. (2012). Tramps, narrow endemics and morphologically cryptic species in the epiphyllous liverwort *Diplasiolejeunea. Molecular Phylogenetics & Evolution* **65**: 582–594. https://doi.org/10.1016/j.ympev.2012.07.009

Dong, S., Schäfer-Verwimp, A., Pócs, T., Feldberg, K., Czumaj, A., Schmidt. A.R., Schneider, H. & Heinrichs, J. (2013). Size doesn't matter – recircumscription of Microlejeunea (Lejeuneaceae, Porellales) based on molecular and morphological evidence. *Phytotaxa* **85**(2): 41–55. https://doi.org/10.11646/phytotaxa.85.2.2

Dorr, L.J. (1997). *Plant collectors in Madagascar and the Comoro Islands: A biographical and bibliographical guide to individuals and groups who have collected herbarium material of algae, bryophytes, fungi, lichens, and vascular* *plants in Madagascar and the Comoro Islands.* Royal Botanic Gardens, Kew, 524 pp.

GOTTSCHE, C.M., LINDENBERG, J.B.W. & NEES, C.G. (1845). *Synopsis Hepaticarum*, fasc. 3. Meissner, Hamburg, pp. 305–464. https://doi.org/10.5962/bhl.title.15221

GROLLE, R. (1995). The Hepaticae and Anthocerotae of the East African Islands. An annotated catalogue. *Bryophytorum bibliotheca* **48**: 1–178.

HE, X.-L. & GROLLE, R. (2001). *Xylolejeunea,* a new genus of the Lejeuneaceae (Hepaticae) from the Neotropics, Madagascar and the Seychelles. *Annales Botanici Fennici* **38**: 25–44.

HERZOG, T. (1947). Hepaticae von der Comoreninsel Johanna. *Botaniska Notiser* **100**(4): 317–334.

HEINRICHS, J., LINDNER, M., GRADSTEIN, S.R., GROTH, H., BUCHBENDER, V., SOLGA, A. & FISCHER, E. (2005). Origin and subdivision of Plagiochila (Jungermanniidae: Plagiochilaceae) in tropical Africa based on evidence from nuclear and chloroplast DNA sequences and morphology. *Taxon* **54**(2): 317–333. https://doi.org/10.2307/25065360

HEINRICHS, J., DONG, S., SCHÄFER-VERWIMP, A, PERALTA, D.F. , FELDBERG, K., , ALEXANDER R. SCHMIDT, A.R., & SCHNEIDER, H. (2014). Towards a monophyletic classification of Lejeuneaceae II: subtribes Pycnolejeuneinae and Xylolejeuneinae subtr. nov., transfer of *Otolejeunea* to Lepidolejeuninae, and generic refinements. *Phytotaxa* **163**(2): 61–76. https://doi.org/10.11646/phytotaxa.163.2.1

HODGETTS, N.G. (2008). A morphological revision of the genus *Herbertus* S.Gray (Herbertaceae, Marchantiophyta) in Africa, including the East African Islands. *Journal of Bryology* **30**: 239–263. https://doi.org/10.1179/174328208X300714

JONES, E.W. (1953). African Hepatics II. *Leptocolea* with hyaline-margined leaves. *Transactions of the British Bryological Society* **2**: 144–157.

JONES, E.W. (1992). African Hepatics XLII. *Radula marojezica* E. W. Jones, a new species from Madagascar. *Journal of Bryology* **17**(2): 307–311. https://doi.org/10.1179/jbr.1992.17.2.307

JOVET-AST, S. (1954, "1953"). Le genre *Colura*, Hepatiques, Lejeuneacees, Diplasiae. *Revue Bryologique et Lichénologique* **22**: 206–312.

KIS, G. & PÓCS, T. (2006). Supplementum and corrections of the non European types and list of exsiccata in the Eger Cryptogamic Herbarium (EGR). *Folia Historico Naturalia Musei Matraensis* **30**: 35–41.

LEE, G.E. & PÓCS, T. (2018). Additions to the distribution of some Palaeotropic *Lejeunea* (Marchantiophyta) species, with the description of *Lejeunea konratii* sp. nov. from Fiji. *Phytotaxa* **349**(1): 31–38. https://doi.org/10.11646/phytotaxa.349.1.3

LEE, G.E., BECHTELER, J., PÓCS, T., SCHÄFER-VERWIMP, A. & HEINRICHS, J. (2015 online, 2016). Molecular and morphological evidence for an intercontinental range of the liverwort *Lejeunea pulchriflora* (Marchantiophyta: Lejeuneaceae). *Organisms Diversity & Evolution* **16**(1):13–21. https://doi.org/10.1007/s13127-015-0243-5

LEHMANN, J.G.C. (1832). *Novarum et minus cognitarum stirpium pugillus quartus.* Meissner, Hamburg, 64 pp. https://doi.org/10.5962/bhl.title.45011

LINDNER, M., PÓCS, T. & HEINRICHS, J. (2004). On the occurrence of *Plagiochila stricta* on Madagascar, new to Africa. *Journal of the Hattori Botanical Laboratory* **96**: 261–271.

MARLINE, L., ANDRIAMIARISOA, R. L., BARDAT, J., CHUAH-PETIOT, M., HEDDER-SON, T. A. J., REEB, C., STRASBERG, D., WILDING, N. & AH-PENG, C. (2012). Checklist of the bryophytes of Madagascar. *Cryptogamie, Bryologie* **33**(3): 199–255. https://doi.org/10.7872/cryb.v33.iss3.2012.199

MÜLLER, F. & PÓCS, T. (2002). Contribution to the hepatic flora of Rodrigues (East African islands). *Tropical Bryology* **22**: 107–113.

ORBÁN, S. (1995). Studies on African Calymperaceae, VI. New data to continental Africa and Madagascar. *Acta Botanica Hungarica* **39**(3–4): 227–234.

ORBÁN, S. (2000). *Calymperes venezuelanum*, a newly discovered American-African disjunct element in the flora of Madagascar. *The Bryologist* **103**(1): 145–146. https://doi.org/10.1639/0007-2745(2000)103[0145:CVANDA]2.0.CO;2

PÓCS, T. (1995). East African Bryophytes, XIV. Hepaticae from the Indian Ocean Islands. *Fragmenta Floristica et Geobotanica* **40**: 251–277.

PÓCS, T. (1997). The distribution and origin of the foliicolous bryophyta in the Indian Ocean Islands. – Cryptogams in the Phyllosphaere: Systematics, Distribution, Ecology and Use. Proceedings of the IAB & IAL Symposium on Foliicolous Cryptogams, 29 August – 2 September 1995, Eger, Hungary. *Abstracta Botanica* **21**: 123–134.

PÓCS, T. (2001). East African Bryophytes, XVI. New taxa of Lejeuneoideae (Lejeuneaceae) collected by Patricia Geissler in Manongarivo Special Reserve, NW Madagascar. *Candollea* **56**(1): 69–78.

PÓCS, T. (2004). New or little known epiphyllous liverworts, XI. *Otolejeunea subana* sp. nov. from Madagascar. *Acta Academiae Paedagogicae Agriensis, Nov. Ser., Sect. Biol.* **25**: 49–57.

PÓCS, T. (2006a). East African Bryophytes, XX. Observations on some Calypogeiaceae. *Acta Academiae Paedagogicae Agriensis*, *Nov. Ser., Sect. Biol.* **25**: 29–35.

PÓCS, T. (2006b). East African Bryophytes. XXI. Two new species of *Telaranea*, sect. *Tenuifoliae* and records on *Amazoopsis* from the Indian Ocean Islands. *Acta Botanica Hungarica* **48** (1–2): 119–137. https://doi.org/10.1556/abot.48.2006.1-2.14

PÓCS, T. (2006c). East African Bryophytes, XX. Observations on some Calypogeiaceae. *Acta Academiae. Paedagogicae Agriensis, Nov. Ser., Sect. Biol.* **25**: 29–35.

PÓCS, T. (2010a). On some less known *Lejeunea* (Lejeuneaceae, Jungemanniopsida) species in tropical Africa. East African Bryophytes, XXVII (With 9 plates). *Nova Hedwigia, Beiheft* **138**: 99–116.

PÓCS, T. (2010b). East African Bryophytes, XXVI. New records from Mayotte (Maore) Island (French Comoro). In: KOPONEN, T., PIIPPO, S. & REINIKKA, E. (eds.): Dr. Ming-Jou Lai Memorial Volume. *Acta Bryolichenologica Asiatica* **3**: 105–114.

PÓCS, T. (2011a). Type studies of some African Lejeuneaceae. *Acta Botanica Hungarica* **53**(1-2): 181–192. https://doi.org/10.1556/abot.53.2011.1-2.18

PÓCS, T. (2011b). *Bazzania orbanii* (Lepidoziaceae), a new species from Madagascar. East African Bryophytes, XXVIII. *Acta Biologica Plantarum Agriensis* **1**: 15–22.

PÓCS, T. (2011c). East African Bryophytes XXIX. The *Ceratolejeunea* (Lejeuneaceae) species in the Indian Ocean islands. *Polish Botanical Journal* **56**(2):131–153.

PÓCS, T. (2020a). New or little known epiphyllous liverworts, XXIII. *Radula pinnata* (Radulaceae), a new species from Madagascar. *Hattoria* **11**: 1–8.

PÓCS, T. (2020b). Notes on some species of *Colura* (Lejeuneaceae, Jungermanniopsida), with description of *Colura cataractarum* from Madagascar. *Acta Botanica Hungarica* **62**: 393–406. https://doi.org/10.1556/034.62.2020.3-4.6

PÓCS, T. (2020c). *Cheilolejeunea hodgettsii sp. nov.* with comments on other African members of sect. *Cyrtolejeunea.* — Notes on the bryophytes of Madagascar 1. *Bryophyte Diversity & Evolution* **42**(1): 49–55. https://doi.org/10.11646/bde.42.1.4

PÓCS, T., CALLAGHAN, A.D., REEB,C. & ANDRIAMIARISOA, R.L. (2018). *Microlejeunea strasbergii* Bardat & Ah Peng. In: ELLIS, T.L. (ed.): New national and regional bryophyte records, 55. *Journal of Bryology* **40**: 179. https://doi.org/10.1080/03736687.2018.1454161

PÓCS, T. & GEISSLER, P. (2002). Chapter 3. The bryophytes collected in the Réserve Spéciale de Manongarivo, Madagascar. In: GAUTIER, L. & GOODMAN, S.M. (eds.): Inventaire Floristique et Faunistique de la Réserve Spéciale de Manongarivo (NW Madagascar). *Boissiera* **59**: 41–76.

PÓCS, T., MIZUTANI, M. & KOPONEN T. (2019). Bryophyte flora of the Huon Peninsula, Papua New Guinea. LXXX. *Cheilolejeunea* and *Drepanolejeunea*, with contributions to *Ceratolejeunea, Cololejeunea*, *Diplasiolejeunea*, *Lejeunea*, *Leptolejeunea*, *Metalejeunea* and *Microlejeunea* (Lejeuneaceae, Marchantiophyta*). Acta Bryolichenologica Asiatica* **8**: 41–84.

PÓCS, T., OCHYRA, R. & BEDNAREK-OCHYRA, H. (2016). *Lepidozia cupressina* (Marchantiopsida, Lepidoziaceae) in sub-Saharan Africa, with a note on the taxonomic status of *L. chordulifera. Cryptogamie, Bryologie* **37**(2):125–147. https://doi.org/10.7872/cryb/v37.iss2.2016.125

PÓCS, T. & PIIPPO, S. (2011). Bryophyte flora of the Huon Peninsula, Papua New Guinea. LXXIV. *Cololejeunea* (Lejeuneaceae, Hepaticae). *Acta Bryolichenologica Asiatica* **4**: 59–137.

PÓCS, T. & SASS-GYARMATI, A. (2006). New or little known epiphyllous liverworts, XII. *Archilejeunea helenae* Pócs & Gyarmati, sp. nova. *Cryptogamie, Bryologie* **27**(1): 103–109.

PÓCS, T. & SCHÄFER-VERWIMP, A. (2006). East African Bryophytes, XXIII. Three new species of *Diplasiolejeunea* (Lejeuneaceae, Jungermanniopsida) from Madagascar*. Cryptogamie, Bryologie* **27**(4): 439–452.

PÓCS, T. & VÁŇA, J. (2002, "2001"). A new species of *Amphicephalozia* (Hepaticae) from Madagascar. *Polish Botanical Journal* **46**(2): 145–150.

PÓCS, T. & VÁŇA, J. (2015). East African Bryophytes XXX. New liverwort and hornwort records. *Acta Biologica Plantarum Agriensis* **3:** 3–21.

PÓCS, T., ZHU, R-L., REINER-DREHWALD, E., SÖDERSTRÖM, L., HAGBORG, A. & VON KONRAT, M. (2015). Notes on Early Land Plants Today. 71. New synonyms, new names and new combinations in Lejeuneaceae (Marchantiophyta). *Phytotaxa* **208**(1): 97–102. https://doi.org/10.11646/phytotaxa.208.1.10

Reeb, C., Marline, L., Rabeau, L., Andriamanantena, A., Andriamiarisoa, R.L., Ranarijaona, H-L. & Pócs, T. (2018). A survey of Marchantiales from Madagascar. *Acta Biologica Plantarum Agriensis* **6**: 3–72.

Reeb, C., Letsara, R. Andriamiarisoa, R.L., Brinda, J.C., Abalo-Loko, G.-A., Boucheron-Dubuisson, E., Fischer, E., Hodgetts, N.G., Lavocat-Bernard, E., Naniarimino, F., Phillipson, P.B., Sass-Gyarmati, A. & A. Vanderpoorten, A. (2019). Un atelier pilote sur les Bryophytes a l'herbier d'Antananarivo (TAN) met en évidence la présence de 7 espèces supplémentaires a Madagascar. *Candollea* **74**: 153–158. https://doi.org/10.15553/c2019v742a5

Sass-Gyarmati, A. (2001). *Lopholejeunea leioptera* Gyarmati (Lejeuneaceae, subfam. Ptychanthoideae), une nouvelle espèce récoltée dans la Réserve Spéciale de Manongarivo (Nord-Ouest de Madagascar). *Candollea* **56**(1): 79–83.

Sass-Gyarmati, A. & Pócs, T. (2002). A second species of *Symbiezidium* from Africa. *Cryptogamie, Bryologie* **23**(1): 31–39.

Sass-Gyarmati, A. (2017). *Bazzania konratiana* sp. nov. (Marchantiophyta: Lepidoziaceae) from Madagascar. *Cryptogamie Bryologie* **38**(2): 119–124. https://doi.org/10.7872/cryb/v38.iss2.2017.119

Sass-Gyarmati, A. & Pócs, T. (2019): A new combination in Lepidoziaceae (Marchantiophyta). *Phytotaxa* **403**: 142. https://doi.org/10.11646/phytotaxa.403.2.8

Schuster, R.M. (1963). An annotated synopsis of the genera and subgenera of Lejeuneaceae. I. Introduction; annotated keys to subfamilies and genera. *Beihefte zur Nova Hedwigia* **9**: 1–203.

Söderström, L., Hagborg, A., von Konrat, M., Bartholomew-Began, Sh., Bell, D., Briscoe, L., Brown, E.† , Cargill, DC., Costa, D.P., Crandall-Stotler, B.J., Cooper, E.D., Dauphin, G., Engel, J.J., Feldberg, K., Glenny, D., Gradstein, S..R., He, X., Heinrichs, J., Hentschel, J., Ilkiu-Borges, A., Katagiri, T., Konstantinova, N.A., Larraín, J., Long, DG., Nebel, M., Pócs, T., Puche, F., Reiner-Drehwald, E., Renner, M.A.M., Sass-Gyarmati, A., Schäfer-Verwimp, A., Segarra, J.G.M., Stotler, R.E.†, Sukkharak, Ph., Thiers, B.M., Uribe, J., Vaňa, J., Villareal, J.C., Wigginton, M., Zhang, L. & Zhu, R.L. (2016): World checklist of hornworts and liverworts. *PhytoKeys* **59**: 1–828. https://doi.org/10.3897/phytokeys.59.6261

Stephani, F. (1916) *Species Hepaticarum 5* Genève & Bâle, George & Cie, pp. 833–1008. https://doi.org/10.5962/bhl.title.95494

Szabó, A. & Pócs, T. (2016). New or little known epiphyllous liverworts, XX. *Cololejeunea nosykombae* A. Szabó & Pócs *sp. nov.* from Madagascar. *Journal of Bryology* **38**(4): 302–307. https://doi.org/10.1080/03736687.2016.1156357

Tixier, P. (1977). Espèces nouvelles malgaches du genre *Diplasiolejeunea* (Spruce) Schiffn. (Hepaticae). *Lindbergia* **4**: 117–125.

Tixier, P. (1979). La famille des *Cololejeunoideae* Grolle dans l'Océan Indien Occidental. – Essai monographique. *Bulletin de l'Academie Malgache* **55**: 173–247.

Tixier, P. (1985). Contribution à la connaissance des Cololejeuneoideae. *Bryophytorum Bibliotheca* **27**: 1–439.

WIGGINTON, M. J. (2018). Checklist and distribution of the liverworts and hornworts of sub- Saharan Africa, including the East African Islands. *Tropical Bryology Research Reports* **9**: 1–138.

WILDING, N. (2016). *Systematics, biogeography and morphological evolution in Entosthodon Schwägr. (Bryopsida, Funariaceae) with a revision of the genus in Africa.* University of Cape Town, 330 pp.

(submitted: 04.12.2020, accepted: 23.12.2020)

Contents

PÉTER SZŰCS, GABRIELLA FINTHA & GERGŐ FAZEKAS:
The bryophyte diversity of central park (Archbishop's Garden) of Eger town (Hungary) 3–16

SÁNDOR DULAI, RÉKA TARNAI, DÓRA SZOPKÓ, ANNA E-VOJTKÓ, DÓRA SALAMON, AMMAR ALEM & ANDRÁS VOJTKÓ:
Iris sibirica has unusual, practically non-different heat tolerance of PS II at different geographical exposures on the Bükk-Plateau (North-Hungary) 17–20

DÁVID KAPI & ANDREA SASS-GYARMATI:
Bryophyte collection in the Herbarium of Eszterházy Károly University (EGR): The digital database of peat moss (*Sphagnum*) species 21–28

TAMÁS PÓCS:
Bryophyte records from the Udzungwa Scarp Nature Forest Reserve, Tanzania 29–39

ATTILA KOSZKA & VIKTOR PAPP:
Aurantiporus croceus, a flagship species of the european fungal conservation is re-discovered after half century in Hungary 40–52

KATALIN PATONAY, ORSOLYA SZABÓ-HUDÁK, HELGA SZALONTAI, MIHÁLY JÁNÓSZKY, ERIKA PÉNZESNÉ KÓNYA & ÉVA ZÁMBORINÉ NÉMETH:
Extraction and identification of major polyphenol constituents of northern hungarian horsemint (*Mentha longifolia* L. (L.)) 53–68

TAMÁS PÓCS:
Notes on the bryophytes of Madagascar 2. New liverwort and hornwort records 69–84