

DOI: 10.17110/StudBot.2015.46.2.95

Studia bot. hung. 46(2), pp. 95–110, 2015

NEW MEMBERS OF THE PERTUSARIALES (ASCOMYCOTA) PROVED BY COMBINED PHYLOGENETIC ANALYSIS

Sergii Y. KONDRATYUK^{1*}, László LÖKÖS², Jung A. KIM³,
Anna S. KONDRATIUK⁴, Min-Hye JEONG³,
Seol Hwa JANG³, Soon-Ok OH³ and Jae-Seoun HUR³

¹*M. H. Kholodny Institute of Botany, Tereshchenkivska str. 2, 01601 Kyiv-601, Ukraine; *ksya_net@ukr.net*

²*Department of Botany, Hungarian Natural History Museum, H-1476 Budapest, Pf. 222, Hungary*

³*Korean Lichen Research Institute, Suncheon National University, Suncheon 540-742, Republic of Korea*

⁴*Institute of Biology' Scientific Educational Centre, Taras Shevchenko National University of Kyiv,
Volodymyrska str. 64/13, 01601 Kyiv, Ukraine*

Kondratyuk, S. Y., Lökös, L., Kim, J. A., Kondratiuk, A. S., Jeong, M.-H., Jang, S. H., Oh, S.-O. & Hur, J.-S. (2015): New members of the Pertusariales (Ascomycota) proved by combined phylogenetic analysis. – *Studia bot. hung.* 46(2): 95–110.

Abstract: New genus *Marfloraea* for the “*Variolaria*” *amara*-group as well as new members of the genera *Dibaeis* and *Ochrolechia* proved by results of the combined phylogenetic analysis based on nuclear ITS1/ITS2 portion of ribosomal nrDNA and 12S SSU mtDNA sequences are described and compared with closely related taxa. Fifteen new combinations are proposed, i.e. *Dibaeis yurii*, *Marfloraea albescens*, *M. amara*, *M. aspergilla*, *M. corallina*, *M. corallophora*, *M. erythrella*, *M. excludens*, *M. mammosa*, *M. ophthalmiza*, *M. panyrga*, *M. pulvinata*, *M. scaberula*, *M. subventosa* and *Ochrolechia dactylina*. *Dibaeis yurii* is recorded for the first time from South Korea.

Key words: *Marfloraea*, new genus, *Ochrolechia*, *Variolaria*

INTRODUCTION

During a field trip to the Russian Far East (Khasan district of Primorsky region) new sterile crustose soredious lichen was found and described as *Ochrolechia yurii* S. Y. Kondr., L. Lökös, S.-O. Oh et J.-S. Hur (KONDRATYUK *et al.* 2014).

The molecular study of recent collections of *O. yurii* allowed to clarify the position of the lichen mentioned on the basis of the results of combined phylogenetic analysis based on ribosomal nuclear and mitochondrial DNA data.

Results of the phylogenetic analysis using both nuclear ribosomal and mitochondrial DNA data (not shown) and combined molecular data show that this lichen is undoubtedly a member of the genus *Dibaeis* Clem. of the Icmadophilaceae Triebel (Fig. 1). During phylogenetic analysis of the members of the order Pertusariales the localisation of “*Pertusaria*” *dactylina* in the *Ochrolechia* clade

was determined. Furthermore, a new genus *Marfloraea* was proposed for the “*Variolaria*” *amara* group positioned in a separate robust branch within the Per-tusariales based on combined molecular data.

MATERIAL AND METHODS

Specimens were examined using standard microscopic techniques, i.e. Nikon SMZ-645 dissecting microscope (Nikon Corp., Tokyo, Japan) for hand-sections, sections were observed using Nikon E-200 and Olympus BX-51 microscope (same as above). Spot test reactions were performed on thalli under a dissecting microscope. Chemicals were extracted in analytical grade acetone in a 1 mL Eppendorf tube. Thin layer chromatography (TLC) was performed using a glass plate coated with TLC Silica gel 60, in solvent system A (toluene : dioxin : acetic acid = 180 : 45 : 5) (ORANGE *et al.* 2010).

Total DNA was extracted directly from the thalli according to EKMAN (1999) and was purified with DNeasy Plant Mini Kit (QIAGEN, Germany). The nuclear ribosomal RNA gene region including the internal transcribed spacers 1 and 2 and the 5.8S subunit (ITS) was amplified using the primers ITS1F (GARDES and BRUNS 1991) and ITS4 (WHITE *et al.* 1990), the 28S LSU using the primer LR5 (VILGALYS and HESTER 1990), and the 12S mtSSU using the primers mtSSU1-mtSSU3R and mtSSU2R (FEDORENKO *et al.* 2009, 2012).

The amplification was done using a Takara JP/TP600 PCR machine (Takara Bio Inc., Japan). One initial cycle of 5 min at 94 °C was followed by 30 cycles of the following steps: 30 seconds at 94 °C, 39 seconds at 57 °C and 1 min at 72 °C. Amplifications were ended with a final cycle at 72 °C for 10 min. PCR products were then sent to the sequencing facilities of the Genotech Cooperation, Seoul, South Korea for cleaning and sequencing. Sequencing was carried out using the fluorescent marker BigDye and an ABI 3730xl sequencing machine (Applied Biosystems, Carlsbad, CA, USA).

The consensus sequence was aligned with all related species sequences retrieved from the GenBank database. The consensus sequences were then deposited into GenBank under the accession numbers KT877997–KT878018. Phylogenetic analysis was performed using the ITS region and LSU nrDNA as well as 12S SSU mtDNA sequences of the treated fungi retrieved from the GenBank database and the lichen-forming fungi investigated in this study. Sequence alignment was conducted in BioEdit and a phylogenetic tree was generated by the maximum parsimony (MP), minimum evolution (ME), and maximum likelihood (ML) analysis methods performed in mega 5.0 (TAMURA *et al.* 2011) with the number of bootstrap trials set to 1,000.

RESULTS AND DISCUSSION

We have tried to include in our comparison the type species of the genera of the order Pertusariales. Unfortunately, data on both nuclear ribosomal and mitochondrial DNA sequences are available only for few taxa of this order.

The order Pertusariales M. Choisy ex D. Hawksw. et O. E. Erikss. includes five families. The family Coccotremataceae Henssen is represented in the phylogenetic analysis by the genus *Coccotrema* Müll. Arg., while molecular data on the genus *Parasiphula* Kantvilas et Grube are incomplete.

The family Icmadophilaceae Triebel, including 6 genera, is represented only by five genera, i.e. *Dibaeis*, *Icmadophila* Trev., *Thamnomia* Ach. ex Schaer., *Siphula* Fr., and *Chirleja* Lendemer et B. P. Hodk. Molecular data on the genera *Pseudobaeomyces* M. Sato, and *Siphuella* Kantvilas, Elix et P. James are incomplete or absent. Since position of the genera *Knightiella* Müll. Arg. and *Endocena* Cromb. is unclear due to the absence of molecular data there is an urgent need of the further studies of these taxa.

Among the genera of the Icmadophilaceae abundant molecular data are present only for the genus *Thamnomia*, while there are data only for single species of some genera, i.e. *Dibaeis baeomyces* (L. f.) Rambold et Hertel, the type species of the genus *Dibaeis*, *Siphula ceratites* (Wahlenb.) Fr. (the type species of the genus *Siphula*) and *Chirleja buckii* Lendemer et B. P. Hodk., the type species of the genus *Chirleja*. At the same time, there are data only on the ITS1/ITS2 portion of nuclear ribosomal DNA for *Icmadophila japonica* (Zahlbr.) Rambold et Hertel (OHMURA 2011), while only mitochondrial data are present for *Icmadophila ericetorum* (L.) Zahlbr. (the type species of the genus *Icmadophila*) in GenBank (MIADŁIKOWSKA *et al.* 2006, PLATT and SPATAFORA 1999, WEDIN *et al.* 2005).

There are only two voucher specimens of the type species of the genus *Ochrolechia* A. Massal. (i.e. *O. tartarea* (L.) A. Massal.) in Ochrolechiaceae R. C. Harris ex Lumbsch et I. Schmitt, as well as data on two taxa of the genus *Varicellaria* Nyl. (see also Table 1) with both nuclear ribosomal and mitochondrial DNA data.

The family Megasporaceae Lumbsch, including three or more genera, is represented in our combined phylogenetic tree by the genera *Megaspora* (Lumbsch, Feige et K. Schmitz) Haf. et V. Wirth, *Lobothallia* (Clauzade et Cl. Roux) Hafellner, and *Aspicilia* A. Massal.

Molecular data on the Pertusariaceae Körb. ex Körb. are provided in several publications. However, there are only data for the genus *Pertusaria* DC. s. l., whereas molecular data on nuclear ribosomal and mitochondrial DNA for the genera *Loxosporopsis* Henssen (type species *L. corallifera* Brodo, Henssen et Imshaug) and *Thamnoochrolechia* Aptroot et Sipman (type species *T. verticillata* Aptroot et Sipman) are totally absent.

Table 1. Specimen vouchers used in the phylogenetic analysis with GenBank numbers.

Species name	Voucher/source	ITS1/ITS2	12S mt SSU
<i>Aspicilia caesiocinerea</i>	NORDIN <i>et al.</i> (2007)	EU057897	
<i>Aspicilia caesiocinerea</i>	Orange, unpubl.	FJ532372	
<i>Chirleja buckii</i>	LENDEMER and HODKINSON (2012)	JX673914	JX673913
<i>Circinaria calcarea</i>	NORDIN <i>et al.</i> (2007)	EU057898	
<i>Circinaria calcarea</i>	OWE-LARSSON <i>et al.</i> (2011)	HQ406804	
<i>Circinaria desertorum</i>	NORDIN <i>et al.</i> (2007)	EU057905	
<i>Circinaria desertorum</i>	OWE-LARSSON <i>et al.</i> (2011)	HQ406802	
<i>Coccotrema coccophorum</i>	SCHMITT <i>et al.</i> (2001 <i>as Lepolichen coccophorus</i>)	AF329169	AF329168
<i>Coccotrema cucurbitula</i>	SCHMITT <i>et al.</i> (2001)	AF329162	AF329161
<i>Coccotrema maritimum</i>	SCHMITT <i>et al.</i> (2001)	AF329165	AF329163
<i>Coccotrema pocillarium</i>	SCHMITT <i>et al.</i> (2001)	AF329167	AF329166
<i>Dibaeis baeomyces</i>	JAMES <i>et al.</i> (2006)	DQ782844	
<i>Dibaeis baeomyces</i>	LUTZONI <i>et al.</i> (2004)		AY584704
<i>Dibaeis baeomyces</i>	LUMBSCH <i>et al.</i> (2004)		AY300883
<i>Dibaeis yurii</i>	SK A31, Russia: Primorsky region; leg. Kondratyuk, S. Y. (RU 06), Oh, S.-O., Hur, J.-S., 04.08.2013; KoLRI-019869 (RU 130342) – holotype	KT877997	KT878008
<i>Dibaeis yurii</i>	SK A32, Russia: the same locality; leg. Kondratyuk, S. Y. (RU 06), Oh, S.-O., Hur, J.-S., 04.08.2013; KoLRI-019866 (RU 130339) – topotype	KT877998	KT878009
<i>Dibaeis yurii</i>	SK B17, Russia: the same local- ity; leg. Oh, S.-O., Hur, J.-S., 04.08.2013; KoLRI-019806 (RU 130279) – topotype	KT877999	KT878010
<i>Dibaeis yurii</i>	SK C68, South Korea: Chuja-do Island; leg. Lökös, L., Kondratyuk, S. 20.06.2014; KoLRI-023218 (140750/1)	KT878000	KT878011
<i>Dibaeis yurii</i>	SK C69, South Korea: Chuja-do Island; leg. Lökös, L., Kondratyuk, S. 20.06.2014; KoLRI-023219 (140750/2)	KT878001	KT878012

Table 1. (cont.).

Species name	Voucher/source	ITS1/ITS2	12S mt SSU
<i>Icmadophila ericetorum</i>	MIADEIKOWSKA <i>et al.</i> (2006)		DQ986897
<i>Icmadophila ericetorum</i>	WEDIN <i>et al.</i> (2005)		AY853327
<i>Icmadophila japonica</i>	OHMURA (2011)	AB623070	
<i>Lecanora allophana</i>	ARUP and GRUBE (2000)	AF159939	
<i>Lecanora allophana</i>	ANDERSEN and EKMAN (2005)		AY567710
<i>Marfloraea albescens</i>	SCHMITT <i>et al.</i> (2001)	AF329177	AF329175
<i>Marfloraea amara</i>	KELLY <i>et al.</i> (2011)	FR799258	
<i>Marfloraea amara</i>	Schmitt, unpubl.	JN943621	
<i>Marfloraea amara</i>	LUTZONI <i>et al.</i> (2004)		AY584713
<i>Marfloraea amara</i>	LUMBSCH <i>et al.</i> (2004)		AY300900
<i>Marfloraea corallina</i>	KELLY <i>et al.</i> (2011)	FR799261	
<i>Marfloraea corallina</i>	LUMBSCH <i>et al.</i> (2004)		AY300901
<i>Marfloraea corallina</i>	SCHMITT <i>et al.</i> (2006)		DQ780286
<i>Marfloraea excludens</i>	Hur, unpubl. (2006)	DQ534475	
<i>Marfloraea excludens</i>	SCHMITT and LUMBSCH (2004)		AY567987
<i>Marfloraea pulvinata</i>	SK C72/B37, Ukraine, "Novy Svit" Botanical Reserve; leg. Kondra- tyuk, S. 21327, 2013; (KW-L)	KT878002	KT878013
<i>Marfloraea pulvinata</i>	SK C72/B38, Ukraine, "Novy Svit" Botanical Reserve; leg. Kondra- tyuk, S. 21327, 2013; (KW-L)	KT878003	KT878014
<i>Marfloraea pulvinata</i>	SK C72/B39, Ukraine, "Novy Svit" Botanical Reserve; leg. Kondra- tyuk, S. 21324, 2013; (KW-L)	KT878004	KT878015
<i>Megaspora verrucosa</i>	IVANOVA and HAFELLNER (2002)	AF332121	
<i>Megaspora verrucosa</i>	IVANOVA and HAFELLNER (2002)	AF332122	
<i>Megaspora verrucosa</i>	SOHRABI <i>et al.</i> (2013)	KC667053	
<i>Ochrolechia androgyna</i>	KELLY <i>et al.</i> (2011)	FR799239	
<i>Ochrolechia androgyna</i>	LUMBSCH <i>et al.</i> (2004)		AY300897
<i>Ochrolechia balcanica</i>	SCHMITT <i>et al.</i> (2001)	AF329172	AF329170
<i>Ochrolechia dactylina</i>	JAMES <i>et al.</i> (2006)	DQ782843	
<i>Ochrolechia dactylina</i>	MIADEIKOWSKA <i>et al.</i> (2006)		DQ972973
<i>Ochrolechia dactylina</i>	MIADEIKOWSKA <i>et al.</i> (2006)		DQ912307
<i>Ochrolechia frigida</i>	LUMBSCH <i>et al.</i> (2004)		AY300898
<i>Ochrolechia frigida</i>	Hur, unpubl.	DQ534474	

Table 1. (cont.)

Species name	Voucher/source	ITS1/ITS2	12S mt SSU
<i>Ochrolechia parella</i>	SCHMITT <i>et al.</i> (2010)		GU980977
<i>Ochrolechia parella</i>	SCHMITT <i>et al.</i> (2001)	AF329174	AF329173
<i>Ochrolechia tartarea</i>	Schmitt, unpubl.	JN943620	
<i>Ochrolechia tartarea</i>	LUMBSCH <i>et al.</i> (2004)		AY300899
<i>Ochrolechia tartarea</i>	Lim and Hur, unpubl.	DQ219304	
<i>Pertusaria australis</i>	SK C70/B71, Ukraine, "Novy Svit" Botanical Reserve; leg. Kondratyuk, S. 21325, 2013; (KW-L)	KT878005	KT878016
<i>Pertusaria flavida</i>	SK C73/B44 Ukraine, "Novy Svit" Botanical Reserve; leg. Kondratyuk, S. 21324, 2013; (KW-L)	KT878006	KT878017
<i>Pertusaria pertusa</i>	SK C71/B40 Ukraine, "Novy Svit" Botanical Reserve; leg. Kondratyuk, S. 21327, 2013; (KW-L)	KT878007	KT878018
<i>Pertusaria pertusa</i>	Schmitt, unpubl.	JN943618	
<i>Pertusaria pertusa</i>	SCHMITT <i>et al.</i> (2003)		AF381565
<i>Pertusaria pertusa</i>	IVANOVA and HAFELLNER (2002)	AF332127	
<i>Pertusaria pertusa</i>	IVANOVA and HAFELLNER (2002)	AF332126	
<i>Protoparmeliopsis muralis</i>	Guzow-Krzeminska, unpubl.	HM209239	
<i>Protoparmeliopsis muralis</i>	CRESPO <i>et al.</i> (2004)		AY464076
<i>Rhizoplaca melanophthalma</i>	LEAVITT <i>et al.</i> (2013)	JX948232	
<i>Rhizoplaca melanophthalma</i>	ARUP <i>et al.</i> (2007)		DQ787352
<i>Siphula ceratites</i>	SCHMULL <i>et al.</i> (2011)	HQ650642	
<i>Siphula ceratites</i>	WEDIN <i>et al.</i> (2005)		AY853344
<i>Thamnomia vermicularis</i>	NELSEN and GARGAS (2009)	EU714429	
<i>Thamnomia vermicularis</i>	WEDIN <i>et al.</i> (2005)		AY853345
<i>Thamnomia subuliformis</i>	Knight <i>et al.</i> , unpubl.	AY961605	
<i>Thamnomia subuliformis</i>	SCHMULL <i>et al.</i> (2011)	HQ650718	
<i>Thamnomia subuliformis</i>	LUTZONI <i>et al.</i> (2004)		AY584728
<i>Varicellaria hemisphaerica</i>	SCHMULL <i>et al.</i> (2011)	HQ650676	
<i>Varicellaria hemisphaerica</i>	MIADLIKOWSKA <i>et al.</i> (2006)		DQ973000
<i>Varicellaria hemisphaerica</i>	SCHMITT <i>et al.</i> (2003)		AF381563
<i>Varicellaria velata</i>	PRIETO <i>et al.</i> (2012)	JX000109	
<i>Varicellaria velata</i>	SCHMITT <i>et al.</i> (2010)		GU980981
<i>Varicellaria velata</i>	LUMBSCH <i>et al.</i> (2004)		AY300906

On the basis of phylogenetic analysis based on the nuclear large subunit (nu LSU) and the mitochondrial small subunit (mt SSU) DNA sequences (LUMBSCH *et al.* 2004, SCHMITT *et al.* 2006, 2010) three monophyletic branches were found, which correspond to the following three genera, *Pertusaria* DC. s. str., *Varicellaria* Nyl. and “*Variolaria*” Pers.. However, there are still no data on the ITS1/ITS2 portion of nuclear ribosomal DNA in GenBank for the same taxa, so not all of these taxa could be included in the combined phylogenetic analysis (Fig. 1).

According to our data “*Pertusaria*” *dactylina* is positioned in the *Ochrolechia* branch based on combined nuclear and mitochondrial data, which is quite different from data of previous authors (JAMES *et al.* 2006, MIADŁIKOWSKA *et al.* 2006).

It should be mentioned that our data do not confirm data of previous authors that genera *Marfloraea* and *Varicellaria* belong to the family Ochrolechiaceae. These genera are positioned in separate clade according to our combined analysis based on ribosomal nuclear and mitochondrial DNA data.

Species of the genus *Icmadophila* form a sister group to the branch of the genus *Dibaeis* based on ITS1/ITS2 portion of nuclear ribosomal DNA sequences. However, there is problem with the inclusion of the members of the genus *Icmadophila* in the combined phylogenetic analysis, since there are no data on both molecular and mitochondrial DNA sequences on the same taxa of this genus in GenBank. So data are available only on ITS1/ITS2 nuclear ribosomal DNA sequences for *Icmadophila japonica* (Zahlbr.) Rambold et Hertel (OHMURA 2011), whereas only mitochondrial data are present for *Icmadophila ericetorum* (L.) Zahlbr. in GenBank (MIADŁIKOWSKA *et al.* 2006, PLATT and SPATAFORA 1999, WEDIN *et al.* 2005).

There is a similar situation with the species of the genus *Parasiphula* Kantvilas et Grube, i.e. *P. complanata* (Hook. f. et Taylor) Kantvilas et Grube, *P. elixii* (Kantvilas) Kantvilas et Grube and *P. foliacea* (D. J. Galloway) Kantvilas et Grube. These species form a sister clade to the genus *Coccotrema* Müll. Arg. (Coccotremataceae, Pertusariales) and to the lichens of the genera *Aspicilia*, *Circinaria*, *Megaspora*, and *Lobothallia* of the Megasporaceae based on ITS1/ITS2 portion of nuclear ribosomal DNA. Our results confirm in part the results of previous investigations on the genus *Parasiphula* (GRUBE and KANTVILAS 2006). However, there are hitherto no data on mitochondrial DNA sequences of the same species of the genus *Parasiphula*.

Our data confirm the results of previous investigations on the polyphyletic origin of the former genus *Pertusaria*. There are two additional clades to the genus *Pertusaria* s. str., i.e.: *Varicellaria* and *Marfloraea* (as “*Variolaria*” (sensu LUMBSCH *et al.* 2004, SCHMITT and LUMBSCH 2004, SCHMITT *et al.* 2006, 2010)). However, it should be mentioned that the following species of the genus *Varicellaria*, i.e. *V. rhodocarpa* (Körb.) Th. Fr., *V. culbersonii* (Vězda) I. Schmitt

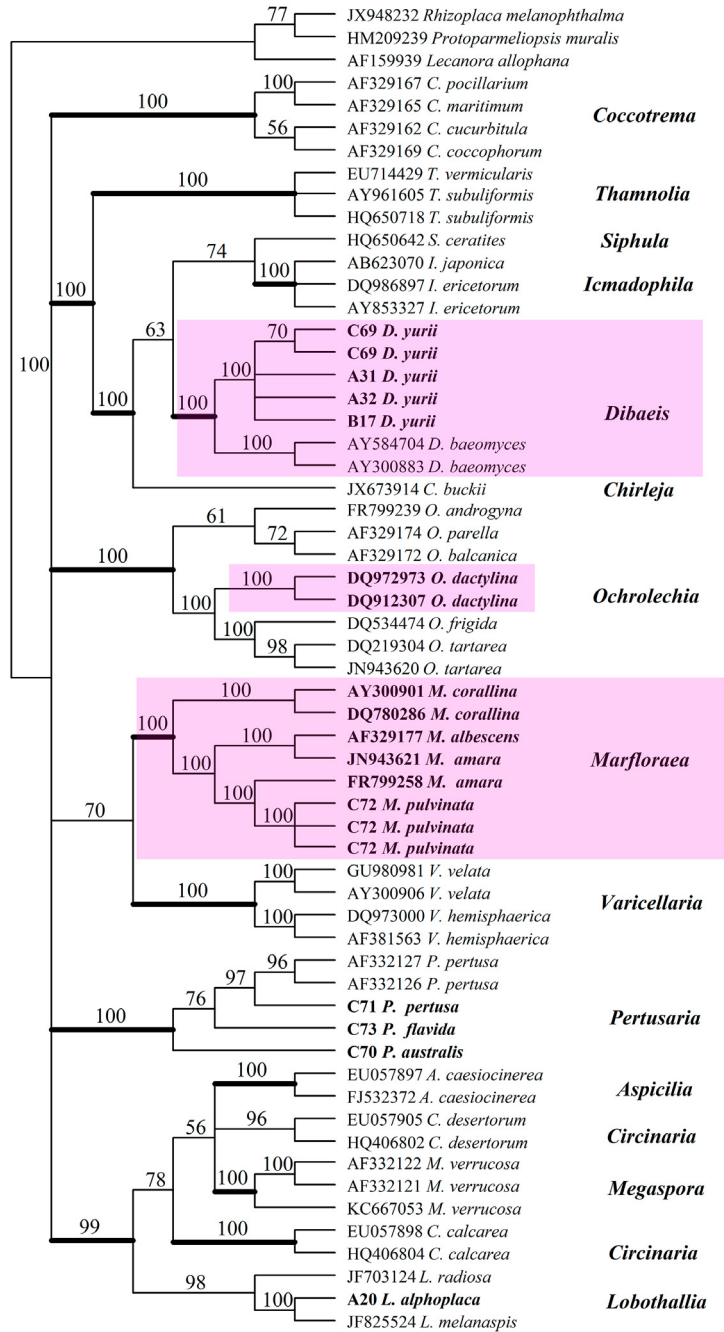


Fig. 1. Phylogenetic tree of the pertusarian lichens based on combined data set.

et Lumbsch, and *V. lactea* (L.) I. Schmitt et Lumbsch, could not be included in the combined data set, because there are no data on their ITS1/ITS2 portion of nuclear ribosomal DNA.

Based on our data *Siphula fastigiata* (Nyl.) Nyl. is positioned now as a sister branch to *Chirleja buckii* Lendemer et B. P. Hodk., while *Siphula ceratites* (the type species of the genus *Siphula*) is positioned in the same clade together with *Icmadophila ericetorum* (type species of the genus *Icmadophila*) based on mitochondrial data. However, data on nuclear ribosomal DNA (ITS1/ITS2 portion) are missing for *Siphula fastigiata*.

NEW TAXA

Marfloraea S. Y. Kondr., L. Lökös et J.-S. Hur, *gen. nova*

Mycobank no.: MB 814663.

Similar to the genus Ochrolechia, but differs in having 1-spored asci, in having weakly amyloid or non-amyloid hymenial gel, in having picrolichenic and thamnolic acids and in the lack of gyrophoric acid.

Diagnosis: Thallus crustose, often with soredia or isidia in some species, apothecia disciform, asci strongly amyloid without recognisable apex structures, the hymenial gel weakly amyloid or non-amyloid, asci 1-spored, ascospore with thin and single-layered wall.

Chemistry: Picrolichenic and thamnolic acids present.

Etymology: The genus *Marfloraea* is named after an outstanding Ukrainian lichenologist Maria Florianivna Makarevych (1907–1982), who worked in Kyiv, Ukraine (i.e. after her nick-name “MarFlor”, well-known among Eurasian lichenologists of the 20th century), and made important contributions to the revision of the Pertusariaceae, Lecanoraceae and other families of crustose North Eurasian lichens as well as to the geography of lichens in general (see BLUM *et al.* 2008, KONDRATYUK 2007a, b, MAKAREVICH 1963, 1971, MAKAREVICH *et al.* 1982).

Species diversity and distribution: so far 12 species are confirmed for this genus based on both nuclear and mitochondrial DNA data sets. Other species of the former *Pertusaria* s. l. are in urgent need of confirmation using molecular data.

Type species: *Marfloraea amara* (Ach.) S. Y. Kondr., L. Lökös et J.-S. Hur.

(We avoided to select *Marfloraea albescens* as type of this genus, since *Variolaria discoidea* Pers. was selected as type species of the genus *Variolaria* Gray, Nat. Arr. Brit. Pl. (London) 1: 490 (821) (see also Dictionary of the Fungi, ed. 10 (KIRK *et al.* 2008))).

Taxonomic notes: the new name *Marfloraea* is proposed for the *Pertusaria amara* group because the name *Variolaria* Pers. (Ann. Bot. (Usten) 7: 23 (1794)

proposed by Persoon in 1794 is a later homonym of *Variolaria* Ball., Bull. Hist. Champ. Fr. (Paris) 1: 181 (1791) (Fungi, Insertae sedis). Persoon's name is rejected (Code, Art. 14.7). Since we have not found appropriate published name for this group we have proposed the new name *Marfloraea*.

The genus *Marfloraea* is similar to the genus *Pertusaria* s. str., but differs in having only disciform apothecia (*v.* disciform or poriform apothecia in *Pertusarias*), in having strongly amyloid without recognisable apex structures (*v.* amyloid ascus with a distinctive ocular chamber), in having weakly amyloid or non-amyloid reaction of the hymenial gel (*v.* non-amyloid in *Pertusarias*), in having 1-spored asci (*v.* 2–8-spored asci in *Pertusarias*), in having only thin and single-layered ascospore walls (*v.* thick or thin as well as double-layered or single-layered ascospore wall), in having picrolichenic and thamnolic acids, and in the lack of chlorinate xanthenes, gyrophoric and planaic acids.

The genus *Marfloraea* is similar to the genus *Ochrolechia*, but differs in having 1-spored asci (*v.* 2–8-spored asci in *Ochrolechias*), in having weakly amyloid or non-amyloid hymenial gel (*v.* strongly amyloid in *Ochrolechias*), in having picrolichenic and thamnolic acids and in the lack of gyrophoric acid.

The genus *Marfloraea* is similar to the genus *Varicellaria*, but differs in having 1-spored asci (*v.* 1–2-spored asci in *Varicellarias*), in having weakly amyloid or non-amyloid hymenial gel (*v.* non-amyloid in *Varicellarias*), in having thin ascospore walls (*v.* more or less thick ascospore walls), in having picrolichenic and thamnolic acids and in the lack of lecanoric acid.

Based on our combined phylogenetic analysis data the genera *Marfloraea* and *Varicellaria* are positioned in separate clades (Fig. 1).

The genus *Marfloraea* is probably polyphyletic, because after Schmitt and colleagues (SCHMITT *et al.* 2006) the *Marfloraea* clade has rather low level of bootstrap support (only 90). Furthermore the *Marfloraea* clade includes at least three species groups, i.e. the *M. amara*, the *M. erythrella* and the *M. ophthalmiza* groups based on data of the authors mentioned above. Species of the genus *Marfloraea* are under special study by several scientific groups (SCHMITT *et al.* 2006, 2010). Hopefully, the status of the *Marfloraea erythrella* and the *M. ophthalmiza* groups will be clarified in the nearest future.

Molecular data on *M. corallophora*, *M. erythrella*, *M. mammosa* and *M. aspergilla* were provided by SCHMITT *et al.* (2006). However, the position of these species within the genus *Marfloraea* is in need of checking after getting data on ITS1/ITS2 nr DNA.

Marfloraea pulvinata is found to be different from *M. amara* according to molecular data. Thus, *M. pulvinata* and *M. amara* are considered as two separate species.

New combinations

Dibaeis yurii (S. Y. Kondr., L. Lökös, S.-O. Oh et J.-S. Hur) S. Y. Kondr., L. Lökös et J.-S. Hur, *comb. nova* [MycoBank no.: MB 814677] – Basionym: *Ochrolechia yurii* S. Y. Kondr., L. Lökös, S.-O. Oh et J.-S. Hur, *Acta Bot. Hung.* 56(1–2): 132 (2014).

Marfloraea albescens (Huds.) S. Y. Kondr., L. Lökös et J.-S. Hur, *comb. nova* [MycoBank no.: MB 814664] – Basionym: *Lichen albescens* Huds., *Fl. Angl.*: 445 (1762). ≡ *Pertusaria albescens* (Huds.) M. Choisy et Werner, *Cavanillesia* 5: 165 (1932).

Marfloraea amara (Ach.) S. Y. Kondr., L. Lökös et J.-S. Hur, *comb. nova* [MycoBank no.: MB 814665] – Basionym: *Variolaria amara* Ach., *K. Vetensk.-Acad. Nya Handl.* 30: 163 (1809). ≡ *Pertusaria amara* (Ach.) Nyl., *Bull. Soc. linn. Normandie, sér. 2* 6: 288 (1872).

Marfloraea aspergilla (Ach.) S. Y. Kondr., L. Lökös et J.-S. Hur, *comb. nova* [MycoBank no.: MB 814666] – Basionym: *Lichen aspergillus* Ach., *Lich. suec. prodr.* (Linköping): 28 (1799) [1798]. ≡ *Pertusaria aspergilla* (Ach.) J. R. Laundon, *Taxon* 41(4): 745 (1992).

Marfloraea corallina (L.) S. Y. Kondr., L. Lökös et J.-S. Hur, *comb. nova* [MycoBank no.: MB 814667] – Basionym: *Lichen corallinus* L., *Mant. Pl.* 1: 131 (1767). ≡ *Pertusaria corallina* (L.) Arnold, *Flora, Jena* 49: 658 (1861).

Marfloraea corallophora (Vain.) S. Y. Kondr., L. Lökös et J.-S. Hur, *comb. nova* [MycoBank no.: MB 814668] – Basionym: *Pertusaria corallophora* Vain., *Résult. Voy. Belgica, Lich.*: 22 (1903).

Marfloraea erythrella (Müll. Arg.) S. Y. Kondr., L. Lökös et J.-S. Hur, *comb. nova* [MycoBank no.: MB 814669] – Basionym: *Pertusaria erythrella* Müll. Arg., *Bull. Herb. Boissier* 1: 41 (1893).

Marfloraea excludens (Nyl.) S. Y. Kondr., L. Lökös et J.-S. Hur, *comb. nova* [MycoBank no.: MB 814670] – Basionym: *Pertusaria excludens* Nyl., *Flora, Jena* 68: 296 (1885).

Marfloraea mammosa (Harm.) S. Y. Kondr., L. Lökös et J.-S. Hur, *comb. nova* [MycoBank no.: MB 814671] – Basionym: *Pertusaria mammosa* Harm., *Lich. de France* 5: 1141 (1913).

Marfloraea ophthalmiza (Nyl.) S. Y. Kondr., L. Lökös et J.-S. Hur, *comb. nova* [MycoBank no.: MB 814672] – Basionym: *Pertusaria multipuncta* var. *ophthalmiza* Nyl., *Lich. Scand. (Uppsala)*: 180 (1861). ≡ *Pertusaria ophthalmiza* (Nyl.) Nyl., *Flora, Regensburg* 48: 354 (1865).

Marfloraea panyrga (Ach.) S. Y. Kondr., L. Lökös et J.-S. Hur, *comb. nova* [MycoBank no.: MB 814673] – Basionym: *Urceolaria panyrga* Ach., *Method.*

Lich.: 146, tab. IV, fig. 2 (1803). ≡ *Pertusaria panyrga* (Ach.) A. Massal., Framm. Lichenogr.: 53 (1855).

Marfloraea pulvinata (Erichsen) S. Y. Kondr., L. Lökös et J.-S. Hur, *comb. nova* [Mycobank no.: MB 814674] – Basionym: *Pertusaria pulvinata* Erichsen, Rabenh. Krypt.-Fl., Ed. 2 (Leipzig) 9 (5.1): 524 (1936). ≡ *Pertusaria amara* f. *pulvinata* (Erichsen) Almb., Bot. Notiser, Suppl. 1(no 2): 77 (1948).

Marfloraea scaberula (A. W. Archer) S. Y. Kondr., L. Lökös et J.-S. Hur, *comb. nova* [Mycobank no.: MB 814678] – Basionym: *Pertusaria scaberula* A. W. Archer, Mycotaxon 41(1): 240 (1991).

Marfloraea subventosa (Malme) S. Y. Kondr., L. Lökös et J.-S. Hur, *comb. nova* [Mycobank no.: MB 814675] – Basionym: *Pertusaria subventosa* Malme, Ark. Bot. 28A(no. 9): 7 (1936).

Ochrolechia dactylina (Ach.) S. Y. Kondr., L. Lökös et J.-S. Hur, *comb. nova* [Mycobank no.: MB 814676] – Basionym: *Lichen dactylinus* Ach., Lich. succ. prodr. (Linköping): 89 (1799) [1798]. ≡ *Pertusaria dactylina* (Ach.) Nyl., Acta Soc. Sci. fenn. 7: 447 (1863).

Phylogenetic analysis of the Pertusariales based on combined nuclear ribosomal and mitochondrial DNA data proved the position of the species of the genus *Marfloraea*, *Dibaeis yurii* and *Ochrolechia dactylina*. Future collections of the sterile soledious, recently described terricolous lichen, *Dibaeis yurii* will allow to compare and complete data on its apothecia and other characters.

* * *

Acknowledgements – We are thankful to Arne Thell (Lund, Sweden) and Edit Farkas (Vácrátót, Hungary) for critical review and valuable comments to this paper, as well as for correction of English. We are thankful to Inna V. Shibneva, Yuri Z. Vaseyev and Igor M. Kashnikov (Primorsky settlement, Khasan district, Russia) for warm hospitality and help during field work in Kedrovaya Pad State Nature Biosphere Reserve, to Elena I. Salmanova (Vladivostok, Russia) for help with getting permits for collecting lichens, as well as to Yuri Ya. Kondratyuk (Vladivostok) for warm hospitality and all kinds of help during our field trip to Khasan district of the Primorsky region. The study was supported by the State Agency on Science, Innovations and Information of Ukraine (M317-2011-409, M111-2012-409, and M40-2013-409), the Korean National Research Foundation (2012-0005582), the Korean National Research Resource Center Program (NRF, 2012 M3A9B8021726) and the Korean Forest Service Program (KNA2013) through the Korea National Arboretum, and partly (for LL) by the Hungarian Scientific Research Fund (OTKA K81232).

Összefoglaló: Kombinált filogenetikai analízis (ITS1/ITS2 riboszomális DNS, 12S SSU mitokondriális DNS) lehetővé tette egy új zuzmónemzetség (*Marfloraea*) lehatárolását a “*Variolaria*” *amara* fajcsoportból, továbbá két rokonfaj taxonómiai státusának megállapítását a *Dibaeis* és *Ochrolechia* nemzetségekben. Erre alapozva 15 új kombinációt hajtottunk végre: *Dibaeis yurii* (bazionim: *Ochrolechia yurii* S. Y. Kondr., L. Lökös, S.-O. Oh et J.-S. Hur), *Ochrolechia dactylina* (bazionim: *Lichen dactylinum* Ach.), *Marfloraea albescens* (bazionim: *Lichen albescens* Huds.),

Marfloraea amara (bazionim: *Variolaria amara* Ach.), *Marfloraea aspergilla* (bazionim: *Lichen aspergillus* Ach.), *Marfloraea corallina* (bazionim: *Lichen corallinus* L.), *Marfloraea corallophora* (bazionim: *Pertusaria corallophora* Vain.), *Marfloraea erythrella* (bazionim: *Pertusaria erythrella* Müll. Arg.), *Marfloraea excludens* (bazionim: *Pertusaria excludens* Nyl.), *Marfloraea mammosa* (bazionim: *Pertusaria mammosa* Harm.), *Marfloraea ophthalmiza* (bazionim: *Pertusaria multipuncta* var. *ophthalmiza* Nyl.), *Marfloraea panyrga* (bazionim: *Urceolaria panyrga* Ach.), *Marfloraea pulvinata* (bazionim: *Pertusaria pulvinata* Erichsen), *Marfloraea scaberula* (bazionim: *Pertusaria scaberula* A. W. Archer), és *Marfloraea subventosa* (bazionim: *Pertusaria subventosa* Malme). A *Dibaeis yurii* zuzmófajt első ízben jelezzük Dél-Koreából.

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(submitted: 02.07.2014, accepted 04.11.2015)