

# **Epiphytic Lichen Diversity and Conservation in Armenia**

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*Consider the Lichen. Lichens are just about the hardiest visible organisms on Earth, but the least ambitious.*

Bill Bryson,

A Short History of Nearly Everything (2003)

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## Summary

The present doctoral thesis work reports the results of extensive lichenological research in the recent years and combines them with known literature data. The work presents the current state of knowledge on the diversity of epiphytic lichenized fungi and their conservation aspects in Armenia, as well as the research on the phylogeny of the *Ramalina pollinaria* aggregate of the genus *Ramalina*.

A total of 230 taxa of lichenized fungi are reported from epiphytic habitats in Armenia based on field studies from 2011 to 2015 and evaluation of the available literature. For each species, notes on taxonomy, chemistry, ecology, and local, regional and world distribution are presented, as well as presence in the protected areas of the country. An identification key for all species is added. Of the total, 219 are specialized epiphytes, rarely found on other substrata, while 11 species occur only incidentally on bark and more usually on rock. The epiphytic lichenized fungi of Armenia belong to 13 orders, 34 families and 88 genera. The most species-rich higher taxa are *Lecanorales* (*Parmeliaceae*, *Physciaceae*, *Teloschistaceae*), *Arthoniales*, *Peltigerales*, and *Pertusariales*. *Lecanora*, *Usnea* and *Phaeophyscia* are the most species-rich genera. 188 species (82% of the epiphytic lichen mycobiota) were found in the Specially Protected Nature Areas of Armenia. The conservation status of 74 species was evaluated following the IUCN Red List of Threatened Species categories and criteria. Among them, nine taxa were assessed as critically endangered (CR), five taxa as endangered (EN), two taxa as vulnerable (VU), four taxa as data deficient (DD), and 54 taxa as least concern (LC). Epiphytic lichens reported from Armenia showed predominantly holarctic distributional patterns. 187 species were found in the temperate deciduous and mixed forests, which dominate in northern and central Armenia, and 56 species in the open arid woodlands of southern Armenia. Overall, 196 taxa are new records of lichen fungi (including epiphytic, saxicolous, and terricolous taxa) and 12 are new records of lichenicolous fungi for Armenia. 37 genera are for the first time reported for the country.

In addition, two new corticolous lichen species, *Verrucaria juglandis* and *Megaspora cretacea*, are described as a new to science. *V. juglandis* grows on *Juglans regia* roots along river banks, infrequently submerged, in a riparian forest. It is associated with lichens that are normally saxicolous and it is characterized by its dark thallus, a dimidiate involucellum, and narrow ascospores. *Megaspora cretacea* species is characterized by a thick, cretaceous thallus and a pale bluish, rather coarse soredia covering most of the thallus. It grows on *Juniperus*

bark in open arid woodlands in Armenia. A key to the three species included in the genus *Megaspora* is also presented here.

As part of a doctoral study, the phylogeny of some species of *Ramalina* was studied, including *R. pollinaria* and allies and similar taxa. Here, the results for the *R. pollinaria* complex are presented in a broad phylogenetic framework, including samples from the Northern Hemisphere, which led to the recognition of two new species in North America and Europe, namely *Ramalina europaea* and *R. labiosorediata*. *Ramalina europaea* can be distinguished by small, punctiform, often terminal soralia starting out on small, spine-like branchlets, whereas *R. labiosorediata* differs from *R. pollinaria* and *R. europaea* in the almost exclusively terminal soralia formed on the tips of normal lobes, originating from the underside and becoming irregularly labriform. Morphological characters, chemistry, ecology and geographical distribution are discussed and an identification key is included. The topology of a maximum likelihood tree based on nuITS shows the presence of three well-supported clades, corresponding to the morphological differences of the three species.

## Zusammenfassung

Die vorliegende Doktorarbeit basiert auf intensiven Geländearbeiten des Autors in 2011–2015. Sie enthält eine umfassende Darstellung der Diversität der epiphytischen lichenisierten Pilze in Armenien und deren relevanter Naturschutzaspekte. Weiterhin werden Ergebnisse einer phylogenetischen Untersuchung der *Ramalina pollinaria*-Gruppe in Europa und Nordamerika dargestellt.

Insgesamt 230 Taxa von epiphytischen, lichenisierten Pilze wurden in Armenien durch eigene Geländearbeit nachgewiesen. Für alle Taxa werden Angaben über Taxonomie, Chemie, Ökologie, und lokale, regionale und weltweite Verbreitung, sowie das Vorkommen in Naturschutzgebieten in Armenien präsentiert. Dies wird ergänzt durch einen Bestimmungsschlüssel zu allen Taxa. 219 Taxa sind spezialisierte Epiphyten die nur ausnahmsweise auf anderen Substrate gefunden wurden. 11 Taxa wurden nur gelegentlich epiphytisch Substrate gefunden und wachsen normalerweise epilithisch. Die Taxa gehören zu 13 Ordnungen, 34 Familien und 88 Gattungen. Die am häufigsten vertretenen Gruppen sind *Lecanorales* (*Parmeliaceae*, *Physciaceae*, *Teloschistaceae*), *Arthoniales*, *Peltigerales*, und *Pertusariales*. *Lecanora*, *Usnea* und *Phaeophyscia* sind die artenreichsten Gattungen. 188 Arten (82% der gesamten epiphytischen Flechtendiversität) wurden in Naturschutzgebieten,

nach Armenischem Recht Specially Protected Nature Areas, festgestellt wodurch ihr Fortbestand bestmöglich gesichert scheint. Von 74 Arten wurde der Naturschutz-Status nach den Richtlinien der IUCN Red List of Threatened Species evaluiert. Neun Taxa erwiesen sich als "critically endangered" (CR), fünf als "endangered" (EN), zwei als "vulnerable" (VU), vier als "data deficient" (DD), und 54 als "least concern" (LC). Phytogeographisch überwiegen Arten mit holarktischen Verbreitungsmustern. 187 Arten sind in den temperaten Laub- und Mischwäldern verbreitet, die in Nord- und Zentral-Armenien vorherrschen. 56 Arten finden sich in den offenen, ariden Wäldern in Süd-Armenien. Insgesamt 196 Taxa von lichenisierten Pilzen und 12 von lichenikolen Pilzen sind neu für Armenien. 37 Gattungen sind neu für das Land.

Zwei rindenbewohnende Arten erwiesen sich als unbeschrieben und wurden neu beschrieben: *Verrucaria juglandis* and *Megaspora cretacea*. *Verrucaria juglandis* wächst auf freiliegenden, aber selten unter Wasser stehenden Wurzeln von *Juglans regia* an Bachufern, in bachbegleitenden Wäldern. Dort wird die Art von normalerweise gesteinsbewohnende Arten begleitet. Sie ist charakterisiert durch einen dunklen Thallus, ein halbiertes Involucellum, und schmale Ascosporen. Die Art *Megaspora cretacea* wird charakterisiert durch einen dicken, kreidigen Thallus mit hellblauen, eher groben Soredien die den größten Teil des Lagers bedecken. Sie wächst auf *Juniperus* in offenen, ariden Wäldern in Süd-Armenien. Ein Bestimmungsschlüssel für die drei zur Zeit in *Megaspora* unterschiedenen Arten ist hinzugefügt.

Für eine phylogenetische Studie wurde der *Ramalina pollinaria*-Artkomplex ausgewählt und basierend auf der nrITS Region ausführlich analysiert. Als Ergebnis wurden zwei neue Arten erkannt und beschrieben: *Ramalina europaea* und *R. labiosorediata*. *Ramalina europaea* unterscheidet sich durch kleine, punctiforme, oft endständige Sorale die sich auf kleinen, dorn-ähnlichen Thallusläppchen entwickeln. *Ramalina labiosorediata* unterscheidet sich von *R. pollinaria* und *R. europaea* durch die fast nur terminalen Sorale, die auf den Spitzen von normalen Thalluslappen an der Unterseite gebildet werden und unregelmäßig lippenförmig werden. Die morphologischen Merkmale, Chemie, Ökologie und geographische Verbreitung werden dargestellt und ein Bestimmungsschlüssel ist erstellt. Die Topologie des Maximum Likelihood-Stammbaums Baum basierend auf nrITS zeigt drei gut unterstützte Kladen, die mit den angegebenen Merkmalen korrespondieren.

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## **CHAPTER 1. General introduction**

### **1.1 Project background and aims**

The present work was implemented within the project “Developing Tools for Conserving the Plant Diversity of the Transcaucasus”, which is funded by the Volkswagen Foundation. The project, which started in 2011 and is now in its 6th year, is part of the “Caucasus Plant Biodiversity Initiative”, a joint initiative of the Botanic Garden and Botanical Museum Berlin-Dahlem (BGBM), Freie Universität Berlin, the Institute of Botany of the National Academy of Sciences of the Republic of Armenia and botanical research institutions from Georgia and Azerbaijan.

One of the target groups selected for more in-depth research within the project are the lichens. The lichenological research that ultimately led to this study was started in 2011 with research internships and study visits and continued in 2013 as a doctoral research project, the results of which are presented in this dissertation. The research presented here was focused on epiphytic lichens, a mostly unexplored ecological group which is threatened due to significant negative human impact on forest ecosystems in Armenia. Up to this project, lichenological studies and field collections in Armenia, although initiated in the 19th century, were not systematic and consequently only few comprehensive lichenological studies had been published so far.

The main goals of this doctoral research were to study epiphytic lichen diversity and conservation aspects in Armenia, as well as to implement more detailed phylogenetic and taxonomic studies on species of the genus *Ramalina*.

The thesis was prepared in cumulative format and consists of five chapters: General Introduction (Chapter 1), a monographic treatment published in the journal Phytotaxa, summarizing the results on studies of epiphytic lichen diversity and conservation in Armenia implemented from 2011-2015 (Chapter 2), several papers presenting lichen inventories throughout the country and new records of lichenized and lichenicolous fungi, published in the journals Willdenowia, Flora Mediterranea, Mycotaxon and Herzogia (Chapter 3), two research papers about lichen species new to science described from Armenia, published in the journals Willdenowia and Herzogia (Chapter 4), and a manuscript about the phylogeny of the *Ramalina pollinaria* group accepted for publication in the journal The Lichenologist (Chapter

5). At the end of this cumulative presentation, all references of the various chapters are provided in the required, unified format, and appendices for the chapters are added.

## **1.2 Lichens**

Lichens are composite organisms consisting of fungal and algae and/or cyanobacteria forming a symbiotic association. Recently, it has been shown that additional fungi embedded in the cortex of some lichens might play a role in the symbiosis (Spribille *et al.* 2016). Lichens being sensitive to changes in the environment, their occurrence depends on various factors, including climatic and microclimatic conditions, substrate properties (type of rock, bark etc.) and its chemical composition, as well as air quality (Armstrong 2015). According to the most recent classification of lichenized fungi in the Ascomycota and Basidiomycota, there are 19,387 accepted species in 995 genera, 115 families, 39 orders and eight classes (Lücking *et al.* 2017).

The focus group of this study were epiphytic lichens. They are an important component of forest ecosystems (Nascimbene *et al.* 2010) and contribute to nutrient, water cycling and food webs in forest ecosystems (Galloway 1992; McCune 2000, Ellis 2012, Li *et al.* 2015). Apart from a general inventory of all epiphytic lichens in the study region, I conducted taxonomic studies focused more profoundly on three genera introduced below.

### **1.2.1 The genus *Ramalina***

*Ramalina* Ach. is a genus of shrubby macrolichens with cosmopolitan distribution. About 150–170 taxa are known (Sérusiaux *et al.* 2010). The lichens of this genus have a green-grey to yellow-green, mostly fruticose thallus containing usnic acid, without a central cord, which separates them from the similar genus *Usnea*, ascospores with an apical apparatus of the so-called *Bacidia*-type, with eight, hyaline, colourless 1-septate ascospores per ascus. These lichens can be found on exposed rock-faces as well as epiphytic on trunks and twigs.

The genus is morphologically and chemically quite variable, but the taxonomic significance of this variation for species delimitation and the influence of environmental conditions on this variation are insufficiently understood. Only few articles have been published on the molecular phylogeny of the genus, particularly Ekman (2001) within phylogenetic studies of *Bacidiaceae* and Sérusiaux *et al.* (2010) studying the phylogenetic relationships between the genera *Niebla* and *Ramalina* in Macaronesia and the Mediterranean

region. In addition, there are several studies considering phylogenetic aspects of selected taxa in the genus *Ramalina* (LaGraeca, 1999, Ohmura *et al.* 2008, Timsina *et al.* 2012, Hayward *et al.* 2014, Perez-Vargaz & Perez-Ortega 2014, *etc.*).

Fifteen species of *Ramalina* were known so far from the South Caucasus (Barkhalov 1983). As part of the present work, we assembled a large amount of molecular phylogenetic data on the encountered species and here I concentrate on the molecular phylogeny and taxonomy of the *Ramalina pollinaria* group, which has substantial morphological polymorphism whereas it is chemically uniform.

### **1.2.2 The genus *Megaspora***

Until now, the genus *Megaspora* (Clauzade & Cl. Roux) Hafellner & V. Wirth included two species, namely *M. rimisorediata* Valadbeigi & A. Nordin and *M. verrucosa* (Ach.) Hafellner & V. Wirth (Valadbeigi *et al.* 2011). Both species are found in Armenia (Gasparyan & Sipman 2013, Harutyunyan *et al.* 2011).

### **1.2.3 The genus *Verrucaria***

Currently, the genus *Verrucaria* Schrad. incorporates about 300 species distributed across the world (Hawksworth 1995). Most of them are saxicolous (grow on rocks) and only 23 are epiphytic (corticolous or lignicolous; Aptroot 1991; Lendemer & Breuss 2009; Lumbsch *et al.* 2011). Overall, 14 species of the genus *Verrucaria* have been found in Armenia (Harutyunyan *et al.* 2011; Gasparyan *et al.* 2015, 2016).

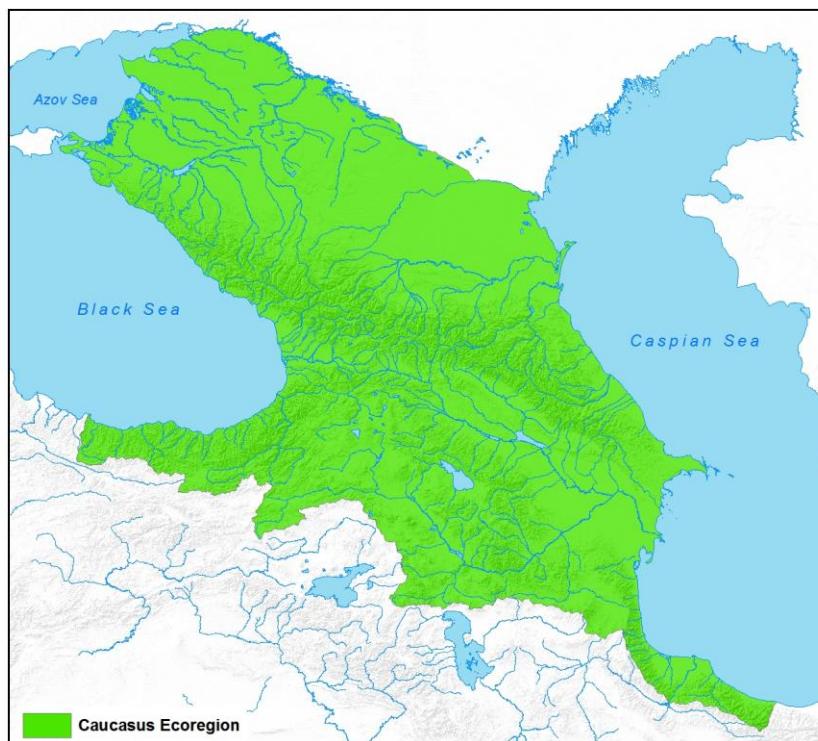
## **1.3 Caucasian ecoregion**

The Caucasus ecoregion covers an area of 580 000 km<sup>2</sup> and includes the territories of Armenia, Azerbaijan, Georgia, a part of the south-west of the Russian Federation, north-eastern Turkey, and the north-western part of Iran (Figure 1.1). Due to its rich biological diversity, a high number of endemic species, and an alarming rate of biodiversity loss, it is defined by the WWF as priority area for conservation and is listed among the "biodiversity hotspots" by Conservation International (CBC, 2012). The area is inhabited by over 7000 plant species, 153 mammals, 400 birds, and a rich diversity of other animal species, with a considerable proportion of endemics (CBC, 2012).

## **1.4 Biodiversity, threats and conservation challenges in Armenia**

Armenia is located in the Southern Caucasus (Transcaucasus). The country has an altitudinal variation from 375 m asl. near the Debed River, to 4,095 m asl. on the top of Mount Aragats, as well as a wide range of climatic zones. These and other environmental factors significantly contribute to the rich biological diversity at the species, landscape and ecosystem level. Various natural habitats, such as semi-deserts, steppes, juniper and xerophytic open and temperate forests, subalpine and alpine grasslands, as well as wetlands, can be found in the country. Armenia covers an area of 29,740 km<sup>2</sup> (6.7 % of the territory of the Caucasus). About half of the flora of the entire Caucasus is present in the country (c. 3,600 vascular plant species, including 125 endemic to Armenia). The fauna of the country includes about 17,500 species (including 316 endemics), the majority of which are invertebrates (Ministry of Nature Protection 1999).

The main threats for biodiversity in Armenia are habitat loss and degradation, deforestation, overgrazing, environmental pollution, and climate change Ministry of Nature Protection 1999).



**FIGURE 1.1.** The map of the Caucasus ecoregion (provided by WWF-Armenia).

### **1.4.1 Forest ecosystems in Armenia**

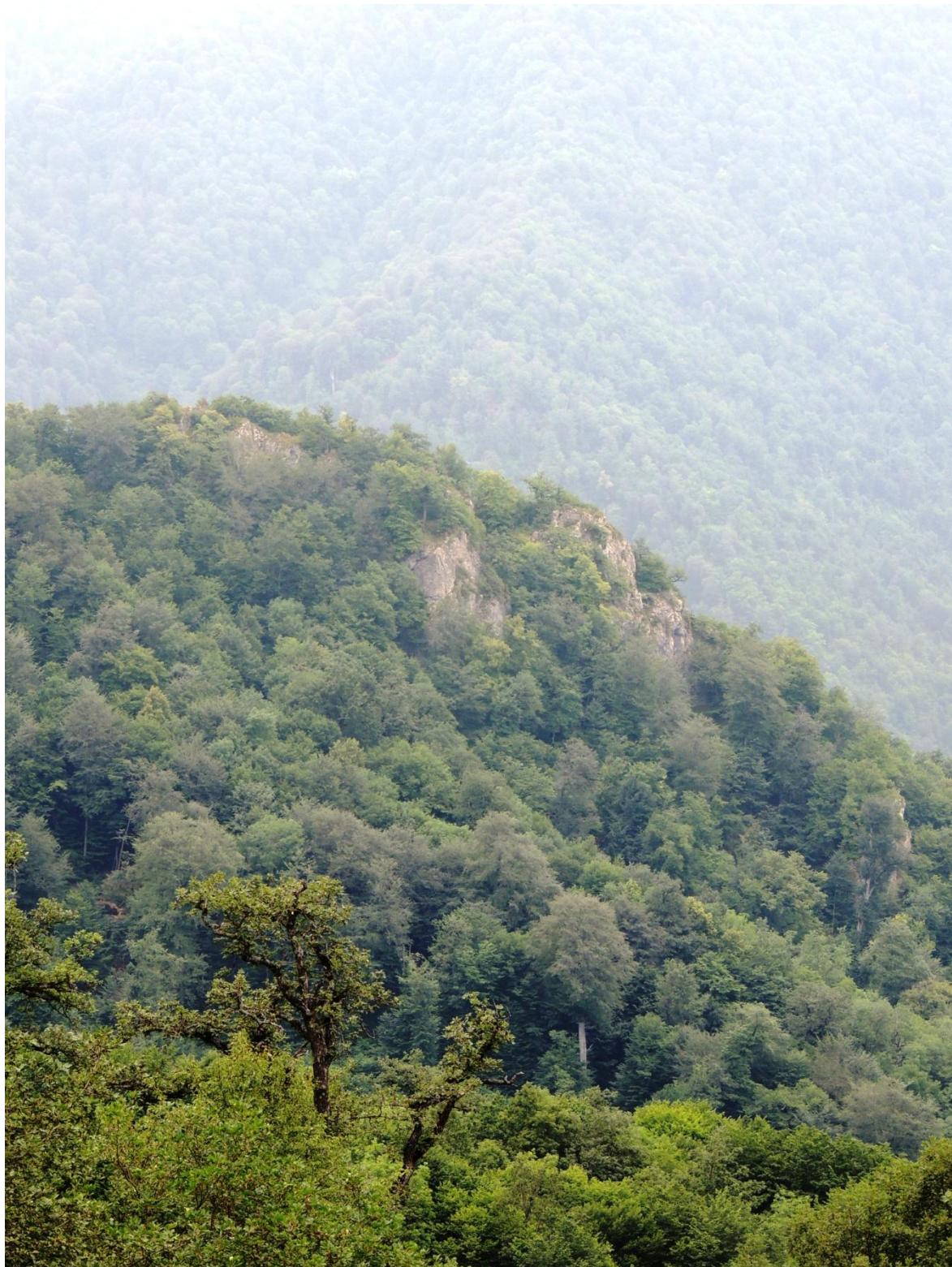
In Armenia, the forested areas occupy 332,333 ha or 11.17 % (Ministry of Nature Protection 2014). Forests out of the protected areas are managed by "Hayantar" SNCO, Ministry of Agriculture. Most of the forests are mostly dominated by temperate deciduous (mixed) forests (Fig. 1.2) and sparse juniper open woodlands (Fig. 1.3) and riparian forests (Vardanyan 2003). Oaks (*Quercus* spp.), oriental beech (*Fagus orientalis* Lipsky) and hornbeam (*Carpinus* spp.) are the main forest-forming species for deciduous forests, whereas juniper (*Juniperus* spp.) dominates in open arid woodlands (WWF-Armenia 2012).

### **1.4.2 Specially Protected Nature Areas of Armenia**

A network of specially protected areas was first established in Armenia in 1958 (Khanjyan 2004). It covers more than 387,054 ha, or 13,01 % of the total area of the country (Ministry of Nature Protection 2014). The biodiversity of the country is protected via a network of these Specially Protected Nature Areas (SPNA). It consists of three state reserves, four national parks, 232 nature monuments, and 27 state sanctuaries (Ministry of Nature Protection 2014). SPNAs are mainly managed by the Ministry of Nature Protection, but several sanctuaries belong to the Ministry of Agriculture and the Ministry of Economy and some nature monuments are managed by local communities. SPNAs have a status of State Non-Commercial Organizations (SNCO) and are regulated by the Law on “Specially Protected Nature Areas”.

According to the IUCN classification, state reserves are protected areas of category Ia, where human interventions are restricted and the area has a strict protection regime. The main goal of state reserves is to preserve ecosystems and their natural state. The national parks (category II of the IUCN classification) have multifunctional zones of protection (e.g., reserve, economic, recreational). They aim to protect natural biodiversity, as well as to promote education and recreation. Natural monuments (category III of the IUCN classification) conserve natural objects of scientific, ecological, geological or historical-cultural importance. State sanctuaries (category IV of the IUCN classification) aim to preserve threatened selected species of flora and fauna and their habitats.

In the frame of the present study, several protected areas, especially forested territories, were studied, and these are outlined below.



**FIGURE 1.2.** Temperate deciduous (mixed) forests in Tavush province of Armenia. (photo by A. Gasparyan)



**FIGURE 1.3.** Sparse juniper woodlands in the southern Armenia. (photos by A. Malkhasyan)

#### **1.4.2.1 Khosrov Forest State Reserve**

The Khosrov Forest State Reserve is considered the oldest protected area in Armenia. It is situated in the Ararat province. The reserve was founded in 334–338 AD as a hunting ground by the Armenian King Khosrov Kotak. In 1958, it was declared a state reserve (23,213 ha). More than 1,800 species of vascular plants are recorded from this area (Khanjyan 2004). The Khosrov Forest State Reserve is known for its open woodlands formed by the Tertiary relicts, Persian juniper (*Juniperus excelsa* subsp. *polycarpos*) and Caucasian oak (*Quercus macranthera*), accompanied by other species, such as ash (*Fraxinus excelsior*, *F. rotundifolia*), and different species of maples (*Acer* spp.) and pears (*Pyrus* spp.).

#### **1.4.2.2 Shikahogh State Reserve and Plane Grove State Sanctuary**

The Shikahogh State Reserve occupies a territory of 12,137 ha. It was also established in 1958 (Khanjyan 2004). The reserve is situated on the northern slopes of the Meghri ridge in Syunik province. Over 1,070 species of vascular plants are known from the reserve. The reserve is mostly covered by broad-leaved forests predominantly composed of oak (*Quercus* spp.) and hornbeam (*Carpinus* sp.) and other tree species. The Plane Grove State Sanctuary is also under the management of the Shikahogh State Reserve. It is a relict grove of the oriental plane (*Platanus orientalis*).

#### **1.4.2.3 Dilijan National Park**

The Dilijan National Park is famous for its temperate forest landscapes. The national park occupies an area of 33,765 ha. It is located in the north-eastern part of the country. In addition to temperate forest ecosystems, Tertiary relict species can also be found, particularly yew (*Taxus baccata*) and the Caucasian rhododendron (*Rhododendron caucasicum*) (Khanjyan 2004).

#### **1.4.2.4 Other Protected Areas**

Several other protected areas were studied here, particularly Arevik National Park, Arzakan-Meghradzor State Sanctuary, Gyulagarak State Sanctuary, Ijevan State Sanctuary,

Stepanavan Sochut Dendropark, Sevan National Park, and Zikatar State Sanctuary, which mostly include temperate forest ecosystems and open arid woodlands.

### **1.5 Red Data Book of Armenia**

The Red Data Book of Armenia is the official national document including information about the distribution, ecology, biology, current status and conservation of rare and endangered species (Tamanyan *et al.* 2010). The conservation status of the species covered by the Red Book has been assessed in compliance with IUCN categories and criteria. The Red Data Book of Plants consists of 452 species of vascular plants and 40 species of higher fungi (Tamanyan *et al.* 2010). However, it does not include lichens. Notably, the previous edition of the Red Book (published in the former USSR) listed three lichen species (*Leptogium burnetiae*, *Leptogium hildenbrandii* and *Usnea florida*) known from Armenia (Bannikov & Sokolov 1984).

The Red Data Book provides the practical basis and legal framework for the conservation of threatened species. The assessment of the conservation status of lichens and their inclusion in upcoming editions of the book is crucial for the conservation of threatened species and their habitats.

The present doctoral dissertation work is an important baseline study to provide the taxonomic and ecological data on epiphytic lichens to eventually filter out species that are potentially threatened.

## **CHAPTER 2. The epiphytic lichenized fungi in Armenia: diversity and conservation**

Gasparyan A. & Sipman H. J. M. 2016. The Epiphytic Lichenized Fungi in Armenia: Diversity and Conservation. – Phytotaxa, [S.l.], v. 281, n. 1, p. 1–68. ISSN 1179-3163.

DOI: <http://dx.doi.org/10.11646/phytotaxa.281.1.1>

## **CHAPTER 3. Inventory of lichens and new records for Armenia**

### **3.1 A contribution to the lichen-forming and lichenicolous fungi flora of Armenia**

Gasparyan, A., Sipman, H. J. M. & Brackel, W. von. (2014) A contribution to the lichen-forming and lichenicolous fungi flora of Armenia. *Willdenowia* 44: 263–267.

DOI: <http://dx.doi.org/10.3372/wi.44.44208>

### **3.2 Additions to the lichenized and lichenicolous mycobiota of Armenia**

Gasparyan A., Aptroot A., Burgaz A. R., Otte V., Zakeri Z., Rico V. J., Araujo E., Crespo A., Divakar P. K. & Lumbsch H. T. (2016) Additions to the lichenized and lichenicolous mycobiota of Armenia. *Herzogia* 29(2): 692-705.

DOI: <http://dx.doi.org/10.13158/heia.29.2.2016.692>

### **3.3 New lichen records from Armenia**

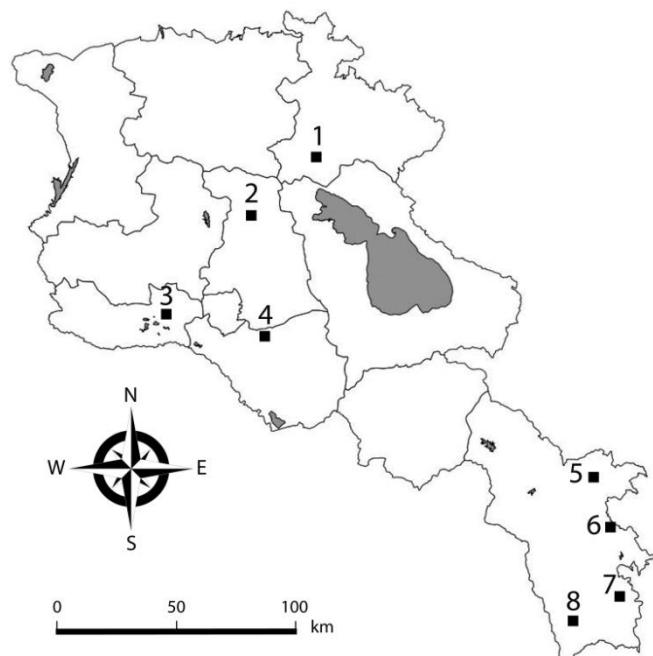
#### **3.3.1 Introduction**

The lichen diversity of Armenia has received considerable attention during recent decades, starting with Nikoghosyan (1963, 1964a, 1964b, 1965, 1966), Barchalov (1983) and Abrahamyan (1983). Harutyunyan *et al.* (2009) added 114 taxa and Harutyunyan *et al.* (2011) published a catalogue of all known lichenized fungi for Armenia, listing a total of 422 taxa.

As a contribution to the knowledge of the lichen diversity, we have started research in the forests. Armenia is one of the least forest-covered countries of the Caucasus. Only about 10% of its territory is covered by more or less intact woodlands (Anonymous 2009). About 97% of these are mixed deciduous broad-leaved forests with oriental beech (*Fagus orientalis*), hornbeam (*Carpinus betulus*, *C. orientalis*) and oak (*Quercus macranthera*, *Q. iberica*, *Q. araxina*) as the dominant species (Vardanyan 2003). Our first observations on the lichen mycota of the forests, presented here, show that this is still very incompletely known and that further exploration is very promising.

### 3.3.2 Material and methods

Specimens were collected in Armenia by the first author between October 2011 and September 2012 from one locality in the semi-desert area of Armavir province and 8 localities in forest areas of the provinces Ararat, Kotayk, Syunik, and Tavush (Fig. 3.4). The visited sites include Shikahogh State Reserve, “Khosrov Forest” State Reserve and “Dilijan” National Park. Morphological features and anatomy of the lichen thalli were studied by stereomicroscope and compound microscope. Secondary chemistry was investigated by thallus fluorescence under long-wave UV light (350 nm) and spot tests following Orange *et al.* (2010). Voucher specimens are deposited in the lichen herbarium of the Botanischer Garten und Botanisches Museum of the Free University in Berlin, Germany (B).



**FIGURE 3.4.** Map of the provinces of Armenia with our study areas. 1 – Tavush province, 2 – Kotayk province, 3 – Armavir province, 4 – Ararat province, 5–8 – Syunik province.

### 3.3.3 Results and discussion

The identified samples included 19 species of lichens (15 epiphytic, 3 saxicolous, and 1 terricolous) not known before from Armenia and reported here as new for the country. Especially notable are three species which, unlike most lichen species known from Armenia,

are rare or absent in Europe and so far mainly known from North America. These include *Lecanora wetmorei* Śliwa, described from numerous collections from North America (Śliwa 2007) and recently reported also from Iran (Valadbeigi *et al.* 2010); this appears to be common in Armenia in temperate mixed deciduous forests and in arid open forests, as well as being widespread on twigs of *Pyrus* sp. in xerophytic areas; identification confirmed by L. Śliwa (Kraków). Two saxicolous species of the *Lecanora dispersa* group described from North America are also among the new records, namely *Lecanora percrenata* H. Magn. (originally described from Central Asia) and *Lecanora flowersiana* H. Magn. (reported from Iran by Valadbeigi *et al.* 2010).

*Caloplaca monacensis* (Leder.) Lettau is an overlooked species in the *C. cerina* group, which has become more easily recognizable by the work of Šoun *et al.* (2011). It is known from temperate and Mediterranean Europe, and western Asia. In Armenia, it occurs in arid, open forest areas. Another unnamed taxon in the *C. cerina* complex, namely *C. cerina* s lat. - group B (Šoun *et al.* 2011), reported from Northern Iran, was also among our material but is not included in the list.

Two species in the *Graphis scripta* group were discovered, based on the recent study by Neuwirth *et al.* (2011), namely *G. betulina* (Pers.) Ach. and *G. pulverulenta* (Sm. & Sowerby) Leight., both occurring on *Carpinus* sp. *G. betulina* is widespread in mixed deciduous forests with hornbeam in the “Dilijan” National Park and the Shikahogh State Reserve, while *G. pulverulenta* was found only once.

Among the reported taxa is a new lichen genus for Armenia, *Gyalecta* Ach., represented by the widespread but inconspicuous species *G. truncigena* (Ach.) Hepp. *Melaspilea urceolata* (Fr.) Almb. is an unusual species known from scattered localities in central and southern Europe, namely Italy, Switzerland, Rumania, Croatia, France, and the surroundings of Münster in Germany (Redinger 1938, sub *M. arthonioides* (Fée) Nyl.; herb. B). Its substrate is mainly old *Quercus* spp. trunks. In Germany, it has not been observed for a long time (Wirth *et al.* 2011) and is probably extinct, but there are recent records from France and Italy in herb. B and it was recently reported from Iran (Valadbeigi *et al.* 2010). In Armenia it was found in the Syunik region (close to the Iranian border). The species is also known from North America (Esslinger 2012), but here the old name *M. arthonioides* is still used.

Two species the genus *Rinodina* are newly reported, *Rinodina furfuracea* H. Magn. and *R. oleae* Bagl. *R. furfuracea* is a distinctive species with an entirely blastidiate thallus and a Mediterranean-Atlantic distribution (Giralt *et al.* 1995). The species occurred in Southern

Armenia on *Quercus* sp., as is observed also in Europe. *R. oleae* is a common species in Southern Europe (Giralt 2001). In Armenia we observed this species in the mixed deciduous broad-leaved forests of the “Dilijan” National Park. H. Mayrhofer (Graz) kindly provided the identification for the first and confirmed the second species. Of the two epiphytic species reported before from Armenia (Harutyunyan *et al.* 2011), *R. pyrina* (Ach.) Arnold was also found by us, while the report of *R. exigua* (Ach.) Gray needs confirmation because the name has been much misapplied in the past. The sorediate species *Megaspora rimisorediata* Valadbeigi, recently described from Iran (Valadbeigi *et al.* 2011), was found on *Quercus* sp. in the Syunik region. This seems to be the first record of this species outside Iran. The further newly recorded species are *Acrocordia cavata* (Ach.) R.C. Harris, *Amandinea punctata* (Hoffm.) Coppins & Scheid., *Caloplaca flavocitrina* (Hoffm.) Th. Fr., *Lecanora valesiaca* (Müll. Arg.) Stizenb., *Leptogium hildenbrandii* (Garov.) Nyl., *Parmelina carporrhizans* (Taylor) Poelt & Vězda, *P. pastillifera* (Harm.) Hale and *Peltigera monticola* Vitik.

### 3.3.3.1 List of Species

For each species voucher information is given: study area number on the map (Fig. 1), locality, coordinates, elevation, substrate, herbarium number and date of collecting (DD/MM/YYYY).

*Acrocordia cavata* (Ach.) R.C. Harris – 1, Tavush: “Dilijan” National Park; 40°45'26"N 44°54'27"E; 1173 m; on tree; B 60 0189290; 17/08/2012

*Amandinea punctata* (Hoffm.) Coppins & Scheid. – 5, Syunik: Goris city, near Yerevanyan highway; 39°31' 07"N 46° 19' 31"E; 1533 m; on *Pinus sylvestris*; B 60 0189289; 02/05/2012

*Caloplaca flavocitrina* (Hoffm.) Th. Fr. – 3, Armavir: Tsatsan village, near cemetery; 40°11'31"N 44°15'56"E; 892 m; on rock; B 60 0189288; 09/11/2011

*Caloplaca monacensis* (Leder.) Lettau – 4, Ararat: “Khosrov Forest” State Reserve; 40°06'34"N 44°46'29"E; 1325 m; on tree; B 60 0189275; 16/05/2012 – 8, Syunik: Vahravar village, near road to Lehvaz; 38° 56'46"N 46°11'05"E; 1390 m; on tree; B 60 0189274; 03/05/2012

***Graphis betulina*** (Pers.) Ach. – 1, Tavush: “Dilijan” National Park; 40°45'12"N 44°56'13"E; 1521 m; on tree; B 60 0189273; 17/08/2012 – 7, Syunik: Shikahogh State Reserve; 39°05'22"N 46° 27'46"E; 1068 m; on *Carpinus betulus*; B 60 0189272; 25/07/2012

***Graphis pulverulenta*** (Sm. & Sowerby) Leight. – 1, Tavush: “Dilijan” National Park, near road to Haghartsin; 40°46'59" N 44°55'15" E; 1149 m; on tree; B 60 0189290; 17/08/2012

***Gyalecta truncigena*** (Ach.) Hepp – 1, Tavush: “Dilijan” National Park; 40°45'40"N 44°55'33"E; 1265 m; on tree; B 60 0189284; 17/08/2012

***Lecanora flowersiana*** H. Magn. – 1, Tavush: Dilijan city, near Dilijan Composers' Creativity House; 40°41'37"N 44°50'45"E; 1599 m; on tree; B 60 0189285; 07/10/2011

***Lecanora percrenata*** H. Magn. – 3, Armavir: Tsatsan village, near cemetery; 40°11' 31"N 44°15' 54"E; 895 m; on rock; B 60 0189287; 09/11/2011

***Lecanora valesiaca*** (Müll. Arg.) Stizenb. – 3, Armavir: Tsatsan village, near cemetery; 40°11' 30"N 44°15'56"E; 890 m; on rock; B 60 0189286; 09/11/2011

***Lecanora wetmorei*** Śliwa – 2, Kotayk: Artavaz village, near Yerevan State University summer practice camp; 40° 36'38"N 44°34'18"E; 1928 m; on tree; B 60 0189271; 07/07/2012 – 4, Ararat: “Khosrov Forest” State Reserve; 40° 06'16"N 44°45'25"E; 1292 m; on tree; B 60 0189270; 16/05/2012 – 1, Tavush: “Dilijan” National Park; 40°45'12"N 44°56'13"E; 1148 m; on tree; B 60 0189269; 17/08/2012 – 6, Syunik: near Tatev village; 39°21'23"N 46°14'59"E; 1757 m; on *Pyrus* sp; B 60 0189268; 03/05/2012 – 8, Syunik: Vahravar village, near road to Lehvaz; 38°56'46"N 46°11'05"E; 1390 m; on tree; B 60 0189267; 03/05/2012

***Leptogium hildenbrandii*** (Garov.) Nyl. – 7, Syunik: “Plane Grove” Sanctuary (Shikahogh State Reserve); 39°03'09"N 46°30'48"E; 694 m; on tree; B 60 0189279; 02/05/2012

***Megaspora rimisorediata*** Valadbeigi – 8, Syunik: Vahravar village, near road to Lehvaz; 38°56'46"N 46°11'05"E; 1390 m; on *Quercus* sp.; B 60 0189265; 03/05/2012

***Melaspilea urceolata*** (Fr.) Almb. – 6, Syunik: Shurnukh village, near access road; 39°22'35"N 46°23' 44"E; 1460 m; on tree; B 60 0189282; 04/05/2012

***Parmelina carporrhizans*** (Taylor) Poelt & Vězda – 7, Syunik: Shikahogh State Reserve; 39°05'11"N 46°27'30"E; 1119 m; on tree; B 60 0189278; 25/07/2012

***Parmelina pastillifera*** (Harm.) Hale – 1, Tavush: Ijevan city; on tree; B 60 0189276; 09/2012

***Peltigera monticola*** Vitik. – 7, Syunik: Shikahogh State Reserve; 39°05'10"N 46°27'27"E; 1125 m; on soil; B 60 0189277; 25/07/2012

***Rinodina furfuracea*** H. Magn. – 8, Syunik: Vahravar village, near road to Lehvaz; 38°56' 46"N 46°11'05"E; 1390 m; on *Quercus* sp.; B 60 0189265; 03/05/2012

***Rinodina oleae*** Bagl. – 1, Tavush: “Dilijan” National Park; 40°45'42"N 44°56'03"E; 1354 m; on tree; B 60 0189281; 17/08/2012

### **3.4 First inventory of lichens and lichenicolous fungi in the Khosrov Forest State Reserve, Armenia**

#### **3.4.1 Introduction**

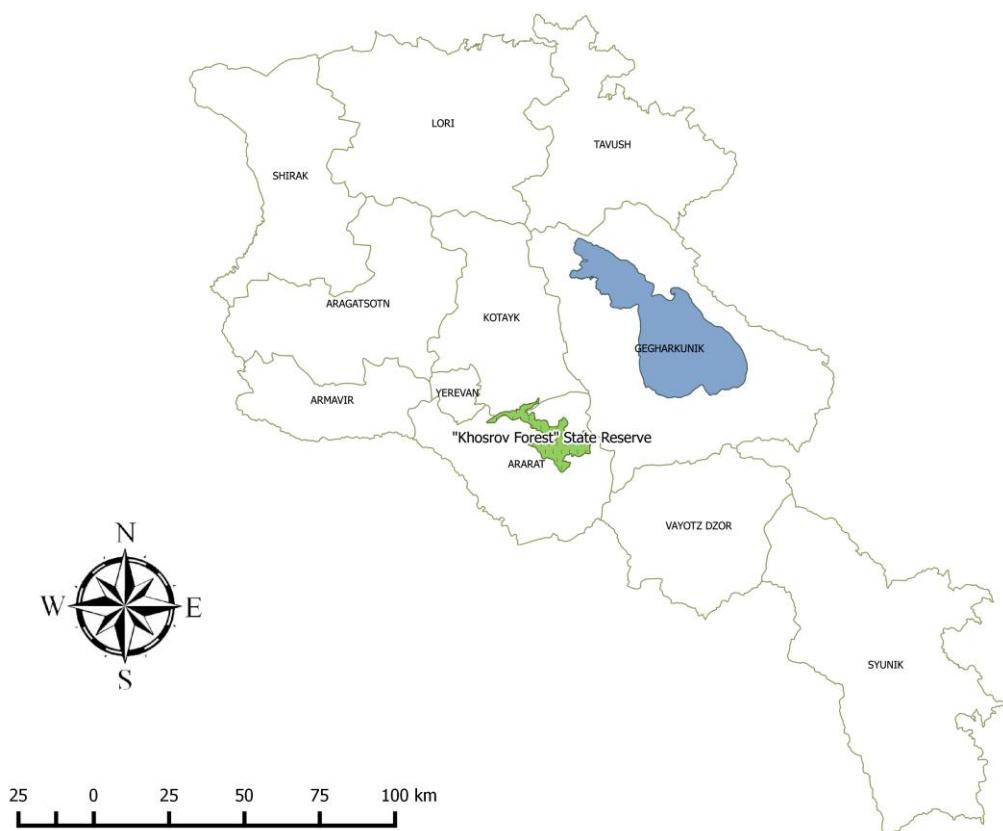
The area of the Khosrov Forest State Reserve has already been recognized as a protected area in the fourth century for hunting and conservation reasons by the Armenian king Khosrov Kotak. In 1958, the Khosrov Forest has officially been established as a State Reserve (Khanjyan 2004). The aim of this protected area is to preserve oak and juniper forest ecosystems and montane vegetation that supports numerous threatened species of plants and animals. The state reserve is under strict protection, corresponding to IUCN category I. The reserve area covers 29196 ha territory in the central part of the Ararat province in Southern Armenia (Fig. 3.5) (Ministry of Nature Protection 1999). It is situated between the slopes south of Geghama and north-west of Urts and north-east of the Yeranos mountain ranges, at

an altitude varying between 900 and 2500 m. The climate is dry continental with annual precipitation of 350 to 800 mm (Anonymous 2008).

The area is known by its exceptionally rich biodiversity and natural landscapes of frigid vegetation, open arid forests and montane steppes (Fig. 3.6). Currently 1849 species of vascular plants (including 24 endemic species) and 283 animal species are known from the reserve (Anonymous 2008). The semi-arid and frigid formations occur on foothills and the lower mountain belt, where dominated a vegetation of *Artemisia fragrans*, *Salsola ericoides*, *S. dendroides*, etc. The forest ecosystems (1400-2300 m) are generally dominated by oak trees (*Quercus macranthera*) and sparse juniper (*Juniperus polycarpos*, etc.) formations, accompanied by *Fraxinus excelsior*, *Sorbus aucuparia*, *Acer*, *Pyrus*, etc., species (Khanjyan 2004). The riverine forest vegetation is predominated by *Fraxinus*, *Populus*, *Salix*, etc., species. Various grasses, such as the feather grass (*Stipa stenophylla*, *S. capillata*, etc.), taragacanth (*Astragalus microcephalus* and *A. lagurus*) motley grass steppe, etc., dominate in the montane steppes (Anonymous 2008).

Currently, 464 lichens and 2 lichenicolous fungi species are known from Armenia (Harutyunyan *et al.* 2011, Gasparyan & Sipman 2013, Gasparyan *et al.* 2014). In the protected area, mainly higher plants, fungi and animals have been investigated and monitored. There are only a few previous lichen records known from the reserve. No comprehensive lichenological surveys have been carried out in the area till date, which could be important for our understanding of lichen diversity in Armenia.

This paper presents the results of the first inventory of lichens in the Khosrov Forest State Reserve, which has been carried out during an international lichenological excursion to Armenia. The excursion was organized in the frame of the OPTIMA Iter Lichenologicum initiative by the Young Biologists Association NGO (Armenia) and OPTIMA (Organization for the Phyto-Taxonomic Investigation of the Mediterranean Area).



**FIGURE 3.5.** The map of the Republic of Armenia including marzes (provinces), and location of the Khosrov Forest State Reserve.



**FIGURE 3.6.** Natural landscapes in the Khosrov Forest State Reserve (photo by Victor J. Rico).

### 3.4.2 Material and methods

During the excursion from 17 to 18 of June, 2015, a total of six localities (see list of localities in Appendix A.2) were visited by the participants of the excursion in the Khosrov Forest State Reserve. The specimens have been collected and identified with routine methods. Secondary metabolites have been studied by thin-layer chromatography (Orange *et al.* 2010). Additionally, in order to confirm the identification based on morphological features, internal transcribed spacer (ITS) of some *Parmeliaceae* and *Physciaceae* specimens have been sequenced (extracts numbered and kept in MAF-Lich.) and compared with available ITS sequences in GenBank (NCBI). Voucher specimens are deposited in the herbaria ABL, B, F, GLM, MACB, MAF-Lich. and PAL.

### 3.4.3 Results and discussion

Overall, 172 species of lichenized and four species of lichenicolous fungi were found in the Khosrov Forest State Reserve. Species are listed in alphabetic order. Species names are followed by the locality or localities numbers, letter corresponding to the substrate and in case of lichenicolous fungi the host lichen species in brackets. Some specimens of *Anaptychia*, *Melanelixia*, *Melanohalea*, *Parmelia*, *Parmelina* and *Physconia*, have been sequenced, in those cases the DNA extraction number is included, in brackets, after the letter corresponding to the substrate. Forty five taxa of lichens and 4 taxa of lichenicolous fungi are new records for Armenia and marked in the list with (\*), 14 of the new records represent new records for genera in Armenia and these have been marked using (\*\*). With these new records, the total number of lichens and lichenicolous fungi known from Armenia reaches 513 species, 507 lichens and 6 lichenicolous fungi.

#### 3.4.3.1 List of Species

*Acarospora assimulans* Vain. – 1c

*Acarospora cervina* (Ach.) A. Massal. – 1d

*Acarospora fuscata* (Nyl.) Th. Fr. – 5d

*Acarospora insolata* H. Magn. – 5d

*Acarospora veronensis* A. Massal. - 1c, 5d

- \**Acarospora versicolor* Bagl. & Car. - 3f, 5d  
\*\**Agonimia tristicula* (Nyl.) Zahlbr. – 2d, 3c  
*Alyxoria varia* (Pers.) Ertz & Tehler – 2a  
*Anaptychia ciliaris* (L.) Körb. ex A. Massal. - 2j (DNA 4965), 3g (DNA 4966)  
*Anaptychia roemeri* Poelt – 1d, 3c  
\*\**Anema decipiens* (A. Massal.) Forssell – 1d, 2d  
*Arctomia fascicularis* (L.) Otálora & Wedin – 2a  
*Arthonia intexta* Almq. (in *Lecidella elaeochroma* apothecia) - 2j  
*Arthonia phaeophysciae* Grube & Matzer (on *Phaeophyscia cf. hirsuta*) – 2a, 5b  
*Aspicilia cinerea* (L.) Körb. – 2c  
*Aspicilia contorta* subsp. *hoffmanniana* S. Ekman & Fröberg ex R. Sant. – 1d  
*Aspicilia desertorum* (Kremp.) Mereschk. – 1cd, 2c, 5d  
\**Aspicilia cf. glomerulans* (Poelt) Poelt – 5d  
\**Aspicilia intermutans* (Nyl.) Arnold - 2c  
*Aspicilia reticulata* Kremp. – 3c  
*Athallia pyracea* (Ach.) Arup – 1ae, 2ae, 3be, 4a  
\**Bacidina arnoldiana* V. Wirth & Vězda – 2e  
\*\**Bagliettoa calciseda* (DC.) Gueidan & Cl. Roux – 2d  
\**Bilimbia sabuletorum* (Schreb.) Arnold – 2d  
\**Blennothallia crispa* (Hudson) Otálora, P.M. Jørg. & Wedin – 3c  
*Calogaya biatorina* (A. Massal.) Arup, Froden & Sochting – 1d  
*Calogaya polycarpoidea* (J. Steiner) Arup, Froden & Sochting – 3a  
*Calogaya pusilla* (A. Massal.) Arup, Froden & Sochting – 2d  
*Caloplaca cerina* (Hedw.) Th. Fr. – 1a, 2ah  
*Caloplaca demissa* (Körb.) Arup & Grube – 3c  
*Caloplaca monacensis* (Leder.) Lettau – 1ae, 2a, 3b  
*Candelariella antennaria* Räsänen – 2e, 4a  
*Candelariella aurella* (Hoffm.) Zahlbr. – 2d  
*Candelariella vitellina* (Ehrh.) Müll. Arg. – 1d, 2d  
\*\**Chrysopsora testacea* (Hoffm.) Choisy – 1d  
*Circinaria calcarea* (L.) A. Nordin, S. Savić & Tibell – 1d, 2ad, 5d  
*Cladonia pocillum* (Ach.) O. J. Rich – 5a  
*Collema flaccidum* (Ach.) Ach. – 2a  
\**Collema subflaccidum* Degel. – 3b, 4a

- Dermatocarpon miniatum* (L.) W. Mann – 2c, 3f, 5d  
*Dimelaena oreina* (Ach.) Norman – 3c  
\**Diploschistes gypsaceus* (Ach.) Zahlbr. – 2f, 3f  
*Diploschistes muscorum* (Scop.) R. Sant. - 5h  
*Diploschistes scruposus* (Schreb.) Norman - 5d  
*Diplotomma hedinii* (H. Magn.) P. Clerc & Cl. Roux – 2d  
*Enchylium tenax* (Sw.) Gray – 2d  
\**Endocarpon pusillum* Hedw. - 3f, 5d  
*Evernia prunastri* (L.) Ach. - 3g  
*Flavoplaca flavocitrina* (Nyl.) Arup, Froden & Sochting – 2c  
*Glypholecia scabra* (Pers.) Müll. Arg. – 1d  
\*\**Gonohymenia nigritella* (Lettau) Henssen – 1d  
\**Gonohymenia schleicheri* (Hepp) Henssen – 3c  
*Gyalolechia flavovirescens* (Wulfen) Sochting, Froden & Arup – 5d  
\**Gyalolechia juniperina* (Tomin) Søchting, Frödén & Arup – 1b  
*Immersaria cupreolastra* (Nyl.) Calat. & Rambold – 1c, 2c  
\**Immersaria iranica* Valadb., Sipman & Rambold – 2c  
*Lathagrium cristatum* (L.) Otálora, P.M. Jørg. & Wedin – 1bd, 5d  
*Lecania cyrtella* (Ach.) Th. Fr. – 3a  
*Lecania fuscella* (Schaer.) A. Massal. - 2a, 3a  
\**Lecania rabenhorstii* (Hepp) Arnold – 2d  
*Lecanora argopholis* (Ach.) Ach. – 1c, 5d  
\**Lecanora barkmaniana* Aptroot & Herk – 3a  
*Lecanora bicincta* Ramond - 3c  
*Lecanora bolcana* (Pollinii) Poelt – 2c, 3c  
*Lecanora carpinea* (L.) Vain. – 1a  
*Lecanora chlarotera* Nyl. – 1a  
*Lecanora garovaglioi* (Körb.) Zahlbr. – 1c, 2c, 3c, 5d  
\**Lecanora juniperina* Śliwa – 1ae, 2ab, 3be  
*Lecanora muralis* (Schreb.) Rabenh. – 1d, 2d, 3c, 5d  
*Lecanora percrenata* H. Magn. – 1d, 5d  
*Lecanora rupicola* (L.) Zahlbr. – 2c, 3c  
\**Lecanora semipallida* H. Magn. – 2d, 5d  
*Lecanora wetmorei* Śliwa – 2a, 4ab, 5abj

- Lecidella elaeochroma* (Ach.) M. Choisy – 2aj, 3a, 5j
- Lecidella euphorea* (Flörke) Hertel – 1a, 2a, 3ag
- Lecidella patavina* (A. Massal.) Knoph & Leuckert – 3c
- Lepraria finkii* (de Lesd.) R. C. Harris – 3c
- Lepraria nivalis* J. R. Laundon – 2d, 3c, 5d
- \*\**Leprocaulon microscopicum* (Vill.) Gams – 3c
- Leptogium saturninum* (Dicks.) Nyl. – 2a, 3a
- \*\**Llimoniella phaeophysciae* Diederich, Ertz & Etayo (on *Phaeophyscia* cf. *hirsuta*) – 1a, 5b
- Lobothallia alphoplaca* (Wahlenb.) Hafellner - 3c, 5d
- Lobothallia praeradiosa* (Nyl.) Hafellner – 1cd, 2c
- Lobothallia radiososa* (Hoffm.) Hafellner – 3c, 5d
- \**Lobothallia recedens* (Taylor) A. Nordin, S. Savić & Tibell – 2c
- Megaspora rimisorediata* Valadb. – 1e, 2b
- Melanelixia glabra* (Schaer.) O. Blanco, A. Crespo, Divakar, Essl., D. Hawksw. & Lumbsch – 1af (DNA 5053), 2aij (DNA 5055), 3abeg (DNA 5054), 5g
- Melanelixia subargentifera* (Nyl.) O. Blanco, A. Crespo, Divakar, Essl., D. Hawksw. & Lumbsch – 1af (DNA 5065, 5067), 2abij (DNA 5068, 5070), 3aeg, 4ab (DNA 5069), 5fj (DNA 5063)
- Melanohalea elegantula* (Zahlbr.) O. Blanco, A. Crespo, Divakar, Essl., D. Hawksw. & Lumbsch – 2j, 3g (DNA 5170), 4ab (DNA 5059)
- Melanohalea exasperata* (De Not.) O. Blanco, A. Crespo, Divakar, Essl., D. Hawksw. & Lumbsch – 1f, 2agi, 4a, 6a
- Parmelia sulcata* Taylor - 3g (DNA 5003, 5004)
- Parmelina tiliacea* (Hoffm.) Hale – 2acj (DNA 4961, 4962, 4963, 4988, 4989, 4990, 4991, 5002), 3g (DNA 4992, 4993), 4b (DNA 4994, 4995, 4996), 6a
- \*\**Peccania coralloides* (A. Massal.) A. Massal. – 3c
- Peltigera canina* (L.) Willd. – 2c
- Peltigera elisabethae* Gyeln. – 3cd
- Peltigera ponjensis* Gyeln. – 2c
- Peltigera praetextata* (Flörke) Vain. – 2c
- Peltigera rufescens* (Weiss) Humb. – 3cd
- \*\**Peltula euploca* (Ach.) Poelt ex Ozenda & Clauzade – 5d
- Phaeophyscia ciliata* (Hoffm.) Moberg – 2g
- Phaeophyscia* cf. *hirsuta* (Mereschk.) Essl. – 1ae, 2ab, 5b

- Phaeophyscia nigricans* (Flörke) Moberg – 1ae, 3b, 5a, 6a  
*Phaeophyscia orbicularis* (Neck.) Moberg – 1a, 2aefi, 3a, 4ab, 5aef; 6a  
*Phaeophyscia sciastra* (Ach.) Moberg – 2f, 3c  
*Physcia adscendens* (Fr.) H. Olivier – 1a, 2ef, 3, 3a, 4b, 5ag  
*Physcia aipolia* (Ehrh. ex Humb.) Fürnr. – 1af, 2aej, 3ag  
*Physcia biziana* (A. Massal.) Zahlbr. – 1af, 2ai, 4a, 5ae  
*Physcia caesia* (Hoffm.) Fürnr. – 3c  
*Physcia dimidiata* (Arnold) Nyl. – 1ad, 2abf, 3e, 4a, 5f  
*Physcia dubia* (Hoffm.) Lettau – 2c  
*Physcia stellaris* (L.) Nyl. – 1af  
*Physcia tenella* (Scop.) DC. – 1f, 2f  
*Physconia detersa* (Nyl.) Poelt – 1f, 3af (DNA 5009)  
*Physconia distorta* (With.) J. R. Laundon – 1a, 2d, 3ab, 5bf  
*Physconia enteroxantha* (Nyl.) Poelt – 2i, 3bc, 5g  
*Physconia grisea* (Lam.) Poelt – 2a  
*Physconia perisidiosa* (Erichsen) Moberg – 2a, 3bg, 4a, 5j (DNA 4973)  
\**Physconia thorstenii* A. Crespo & Divakar - 1f (DNA 4972, 5010), 2j  
\*\**Piccolia ochrophora* (Nyl.) Hafellner – 1a  
\**Placidium lacinulatum* (Ach.) Breuss - 3f  
*Placidium rufescens* (Ach.) A. Massal. – 1d, 3cd  
*Placocarpus schaereri* (Fr.) Breuss – 1d, 5d  
\**Placopyrenium fuscellum* (Turner) Gueidan & Cl. Roux – 2d  
*Placynthium nigrum* (Huds.) Gray – 2d, 3c  
*Pleurosticta acetabulum* (Neck.) Elix & Lumbsch – 2a  
\*\**Psorotichia schaereri* (A. Massal.) Arnold – 1d  
*Punctelia borreri* (Sm.) Krog - 1f  
*Pyrenodesmia variabilis* (Pers.) A. Massal. – 1bc, 2d, 5d  
*Ramalina farinacea* (L.) Ach. – 3a  
*Ramalina pollinaria* (Westr.) Ach. - 3g  
*Rhizocarpon disporum* (Nägeli ex Hepp) Müll. Arg. – 2c, 3c  
*Rhizocarpon geographicum* (L.) DC. – 3c  
*Rhizoplaca chrysoleuca* (Sm.) Zopf – 3c  
*Rhizoplaca melanophthalma* (DC.) Leuckert – 1c  
*Rhizoplaca peltata* (Ramond) Leuckert & Poelt – 1c, 2c, 5d

\**Rinodina colobina* (Ach.) Th. Fr. – 1e, 2a

*Rinodina immersa* (Körb.) J. Steiner – 2cd

\**Rinodina obnascens* (Nyl.) H. Olivier – 2c

*Rinodina pyrina* (Ach.) Arnold – 3e

*Romjularia lurida* (Ach.) Timdal – 2d

*Rusavskia elegans* (Link) S.Y. Kondr. & Kärnefelt – 5d

*Sarcogyne regularis* Körb. – 2d

\**Scytinium gelatinosum* (With.) Otálora, P.M. Jørg. & Wedin – 2adf, 3ac, 5d

*Scytinium lichenoides* (L.) Otálora, P.M. Jørg. & Wedin – 2afj, 3e

\**Scytinium turgidum* (Ach.) Otálora, P.M. Jørg. & Wedin – 3c

\*\**Solenopsora holophaea* (Mont.) Samp. – 2d, 3c

*Squamaria cartilaginea* (With.) P. James – 2c



**FIGURE 3.7.** *Toninia cinereovirens* and *Thermutis velutina* on limestone in the Khosrov Forest State Reserve (photo by Maaike Vervoort).

*Staurothele areolata* (Ach.) Lettau – 3c

*Staurothele fuscocuprea* (Nyl.) Zschacke – 2d

*Tephromela atra* (Huds.) Hafellner – 5a

\*\**Thermitis velutina* (Ach.) Flot. – 1c, 1d, 2c (Fig. 3.7)

*Thyrea confusa* Henssen – 1d, 2d

\**Toninia candida* (Weber) Th. Fr. – 2d

*Toninia cinereovirens* (Schaer.) A. Massal. – 1d (Fig. 3.7)

*Toninia sedifolia* (Scop.) Timdal – 3d, 5d

\**Toninia squalida* (Ach.) A. Massal. – 3c

\*\**Tremella phaeophysciae* Diederich & M. S. Christ. (on *Pheophyscia orbicularis*) - 4b

\**Usnea lapponica* Vain. - 3g

*Usnea substerilis* Motyka - 3g

\**Usnea wasmuthii* Räsänen – 3g

\**Verrucaria dolosa* Hepp – 2e

*Verrucaria hochstetteri* Fr.- 2d

\**Verrucaria macrostoma* Dufour ex DC. – 1d

*Verrucaria muralis* Ach. – 2d

*Verrucaria nigrescens* Pers. – 2de, 5d

*Xanthocarpia lactea* (A. Massal.) A. Massal. – 2d

*Xanthomendoza fulva* (Hoffm.) Søchting et al. – 5b

*Xanthomendoza ulophyllodes* (Räsänen) Søchting, Kärnefelt & S.Y. Kondr. – 2a, 3b, 4a, 5b

*Xanthoparmelia conspersa* (Ach.) Hale - 5d

*Xanthoparmelia loxodes* (Nyl.) O. Blanco, A. Crespo, Elix, D. Hawksw. & Lumbsch – 3c

\**Xanthoparmelia protomatrae* (Gyeln.) Hale - 5d

*Xanthoparmelia pulla* (Ach.) O. Blanco, A. Crespo, Elix, D. Hawksw. & Lumbsch – 1c, 2c,

3c, 5dh

*Xanthoparmelia somloensis* (Gyeln.) Hale – 3c, 5dh

\**Xanthoparmelia tinctina* (Maheu & A. Gillet) Hale – 1c, 2c, 3cd, 5d

*Xanthoparmelia verruculifera* (Nyl.) O. Blanco, A. Crespo, Elix, D. Hawksw. & Lumbsch – 5d

*Xanthoria parietina* (L.) Th. Fr. – 5a, 5f, 5g

## **CHAPTER 4. New lichen species of the genera *Verrucaria* and *Megaspora* from Armenia**

### **4.1 *Verrucaria juglandis*, a new corticolous lichen species from Armenia**

Gasparyan, A. & Aptroot, A. (2016) *Verrucaria juglandis*, a new corticolous lichen species from Armenia. *Herzogia* 29: 103–107.

DOI: <http://dx.doi.org/10.13158/heia.29.1.2016.103>

**4.2 A new corticolous *Megaspora* (*Megasporaceae*) species from Armenia**

Zakeri, Z., Gasparyan A. & Aptroot A. (2016) A new corticolous *Megaspora* (*Megasporaceae*) species from Armenia. *Willdenowia* 46 (2): 245–251.

DOI: <http://dx.doi.org/10.3372/wi.46.46205>

## **CHAPTER 5. *Ramalina europaea* and *Ramalina labiosorediata*, two new species of the *Ramalina pollinaria* group (Ascomycota: Ramalinaceae), and new typifications for *Lichen pollinarius* and *Lichen squarrosus***

### **5.1 Introduction**

*Ramalina* s. lat. is a cosmopolitan genus with 150–170 taxa (Serusiaux *et al.* 2010). It is characterized by a fruticose thallus, almost exclusively with usnic acid, angular to flattened branches without a central cord, and hyaline 1-septate ascospores, features that separate the genus from similar lichens in *Alectoria*, *Evernia*, and *Usnea*. *Ramalina pollinaria* (Westr.) Ach. is one of the earliest described species in the genus (Westring 1795, as *Lichen pollinarius*). As with other species from this early period, the typification of the name poses problems. Howe (1913) supposed that the type was from Sweden, but noted that the location of Westring material was unknown. Krog & James (1977) designated a neotype based on a sheet containing eleven specimens, four of them (from Sweden) originally described as *R. pollinaria* var. *humilis* Ach., five (from Switzerland) as *R. pollinaria* var. *elatior*, and two (from France) identified as *R. pollinaria*, the designated neotype for *R. pollinaria* being H-ACH 1831D.

*Ramalina pollinaria* in its current definition is widespread in Eurasia and North America, but has also been reported from South America, Australia, New Zealand and Africa (GBIF 2016, Galloway 2007). However, the reports from South America and Australia are incorrect and correspond to taxa now known as *Ramalina chilena* (Nyl.) Kashiw. (Kashiwadani 1990) and *R. unilateralis* F. Wilson (Stevens 1987), respectively; reports from Africa need to be verified. *Ramalina pollinaria* has a rather broad ecological niche, although it is nowhere particularly abundant, growing within a wide elevation range and on various substrata including bark and wood of different phorophytes, as well as various rock types (Wirth *et al.* 2013). The species is characterized by an erect, shrubby thallus with narrow, solid, much branched lobes bearing laminal or terminal, or more rarely marginal, irregular soralia that may extend onto the lower side of the lobes (Andreev *et al.* 2008, Brodo *et al.* 2001, Smith *et al.* 2009). It is, however, rather polymorphic and can sometimes only be distinguished from similar taxa, such as *R. farinacea*, *R. intermedia*, or *R. obtusata*, by the presence of evernic acid as the major medullary substance (*R. farinacea*: protocetraric, hypoprotocetraric or norstictic/salazinic acid; *R. intermedia*: sekikaic acid; *R. obtusata*:

obtusatic and evernic acids). Obtusatic acid was reported for *R. pollinaria* (Brodo *et al.* 2001), but was not confirmed during the present study, nor were evernic acid-deficient specimens as reported from Great Britain (Smith *et al.* 2009).

The morphological plasticity found in *R. pollinaria* is apparent by the numerous infraspecific categories that have been described. Index Fungorum (2016) lists 36 basionym names of forms and varieties of *R. pollinaria*; none of these is formally used today and almost all have been subsumed into synonymy, with many not even belonging in *R. pollinaria* s.lat., which supports the notion that these names mostly did not reflect natural entities to begin with. A revision of about half of these names, including all that were at some point elevated to species level, is included in the present study.

The ITS barcoding locus has been recommended for Fungi (Schoch *et al.* 2012) and has been successfully employed in several studies of species delimitation and population genetics in the genus *Ramalina* (Groner & LaGreca 1997, LaGreca 1999, Stocker *et al.* 2004, Ohmura *et al.* 2008, Sérusiaux *et al.* 2010, Francisco de Oliveira *et al.* 2012, Timsina *et al.* 2012, Del Campo *et al.* 2013, Hayward *et al.* 2014). Thus, while some workers, such as Kashiwadani & Moon (2002), accept *R. pollinaria* in a broad sense, phylogenetic studies on North American representatives using ITS (Timsina *et al.* 2012) suggest that this is a polyphyletic entity. Ohmura *et al.* (2008), after revision of the three phenotypically similar species *R. pollinaria*, *R. yasudae* and *R. sekika*, confirmed the status of the latter two taxa as separate from *R. pollinaria* in East Asia, and they are actually not closely related to *R. pollinaria*. We therefore also employ the ITS fungal barcoding marker in the present study, combined with a broad assessment across the entire genus.

As part of a PhD research project on the epiphytic lichen diversity of Armenia by the first author, the phylogeny of selected species of *Ramalina* was studied, including *R. pollinaria* and allied taxa. Here, we present the results for the *R. pollinaria* complex in a broad phylogenetic framework, including samples from across the Northern Hemisphere, which led to the recognition of two new species in North America and Europe. We also reassess the status of historic names variously placed in synonymy of *R. pollinaria* or treated as infraspecific taxa under that species and provide four new typifications for critical names.

## **5.2 Material and methods**

### **5.2.1 Taxon sampling**

Among 58 specimens morphologically and chemically identified as *Ramalina pollinaria*, 33 were successfully sequenced and treated in this study. Among the sequenced specimens, 17 were collected by the first author in Armenia and Sweden. The 16 additional specimens sequenced, as well as further specimens not sequenced, were borrowed from New York Botanical Garden Herbarium (NY), the National Herbarium of Canada, Canadian Museum of Nature (CAN), the Herbarium of the Urals Federal University (UFU), the Lichen Herbarium of the Università degli Studi di Trieste (Herb. Nimis, TSB), the University of Manitoba Cryptogamic Herbarium (WIN), and the herbarium of Senckenberg Museum für Naturkunde Görlitz (GLM).

### **5.2.2 Morphological studies and thin-layer chromatography**

Morphological characters and features of the specimens were observed with a WILD M3A stereomicroscope (Heerbrugg, Switzerland) and a ZEISS Axioskop compound microscope (Jena, Germany). Lichen substances were examined with thin layer chromatography (TLC) using solvents A, B' and C (Orange *et al.* 2010).

### **5.2.3 Molecular techniques**

New sequences of the nuclear internal transcribed spacer (ITS) region were generated by using the NucleoSpin Plant II kit (Macherey Nagel) for extraction and Extract-N-Amp Plant PCR Kit (Sigma-Aldrich) for extraction and amplification according to the manufacturer's instructions, with minor modifications regarding the amount of extraction buffer (20 µl) and dilution buffer (20 µl) per sample. Dilutions of 1:5 of genomic DNA were added to the PCR mix solution. For amplification of fungal ITS rDNA, ITS1-forward and ITS4-reverse primers were used (White *et al.* 1990, Gardes and Bruns 1993). PCR cycling was performed with the following protocol: initial denaturation for 5 minutes 95°C; 35 cycles of 95°C for 45 sec, 54°C for 1 min and 72°C for 1 min and final elongation at 72°C for 5 min. PCR products were visualized by 1.5% agarose gel electrophoresis and cleaned with ExoSAP-IT PCR Product Cleanup (Affymetrix). The sequences were generated by Macrogen (Amsterdam, The Netherlands).

### **5.2.4 Alignment and phylogenetic analysis**

To delimit the study group, we first assembled a full data set of all available ITS sequences of *Ramalina* including *Niebla*, which in some studies has been shown to be nested within *Ramalina* from GenBank, including the newly generated sequences, using *Cliostomum griffithii* as an outgroup. A supported clade including the target group was then selected for subsequent analysis, using three sequences of *R. fastigiata* as an outgroup. In addition to the 33 newly generated sequences and the three outgroup sequences, 10 sequences were added from GenBank (see Appendix C1 Table 5.1), including eight from Europe, one from China, and one from Canada, out of four identified as *R. pollinaria* in the study by Timsina *et al.* (2012). Both data sets were first aligned with MAFFT (Katoh & Standley 2013) and then manually adjusted where necessary. Phylogenetic reconstruction was performed under the maximum likelihood (ML) criterion in RAxML 8.2.0. (Stamatakis 2014) using the GTR-Gamma model and 1000 pseudoreplicates for non-parametric bootstrapping.

## **5.3 Results and discussion**

### **5.3.1 Phylogeny**

The broad-scale phylogeny shows the target clade with good support clustering as unsupported sister to a clade containing sequences mostly labeled *Ramalina fastigiata* (see Appendix 3 Supplementary Fig. 5.1). Six sequences labeled *R. pollinaria* from GenBank cluster outside this clade and apparently represent misidentifications or conceptual problems, namely JN084055 from China clustering with *R. sinensis* s.lat., EU034670 from Hungary clustering with *R. fastigiata*, JF923602 from China clustering with *R. intermedia*, and three sequences (JQ003097, JQ003098, JQ003099) from Canada forming a separate clade as unsupported sister to *R. culbersoniorum* (see Appendix 3 Supplementary Fig. 5.1). These sequences are not further considered here as they are far outside the target clade. Especially the three samples from North America require further study, as they obviously represent an undescribed taxon morphologically and chemically similar to *R. pollinaria* but unrelated to the latter.

The *Ramalina pollinaria* clade shows three large, supported ingroup clades and three orphaned singleton sequences (Fig. 5.1). All specimens within this clade have the same

chemistry, namely usnic and evernic acids. Ingroup clade 1 contains all specimens from North America (Canada and USA) except the singleton sequence JQ003096, whereas the other two ingroup clades (2 and 3) include specimens from Europe, the Caucasus, and Russia (Eurasia); the remaining two singleton sequences originate from China (JF923612) and Switzerland (GU827324). A display of the variable positions in the ITS alignment shows that ingroup clades 1 and 2 are separated by seven fully diagnostic positions, ingroup clades 1 and 3 by eight, and ingroup clades 2 and 3 by seven fully diagnostic positions (see Appendix C4 Supplementary Fig. 2). In contrast, one or two positions are variable within each clade, resulting in a large barcoding gap as suggested by the long stem branches. The singleton sequence from China (JF923612) is an unsupported sister of clade 1, differing in six diagnostic positions. The singleton sequence from Switzerland (GU827324) is an unsupported sister of clade 3, with seven diagnostic differences. The singleton sequence from Canada (JQ003096) is found in an unresolved position basal position in the *R. pollinaria* clade. This sequence is very short and lacks the entire, diagnostic ITS2 region, whereas the ITS1 region contains three ambiguous base calls in diagnostic positions; it has one difference with clade 1 and no difference with clade 2 within the covered region.

The three larger ingroup clades correlate with morphological features: clade 1 from North America forms predominantly terminal soralia which originate from the underside and may become irregularly labriform and rather large. In contrast, the two Eurasian clades form smaller, subterminal to marginal-laminal soralia on the upper side of the lobes but partially continuing onto the underside marginally; in clade 3, the soralia start out as small, granular, punctiform soralia subterminally on the lobe surface, with the additional presence of numerous small, subterminal, thin branchlets, whereas in clade 2, the soralia are generally marginal and terminal and more elongate and farinose from the beginning, without additional small branchlets present. We conclude that the three clades represent three different taxa, with clade 2 in agreement with the newly selected neotype of *R. pollinaria* (see below), whereas no species-level names are available for the other two clades and these are formally described and discussed below, as *R. labiosorediata* (clade 1) and *R. europaea* (clade 3).



**FIGURE 5.1.** Maximum-likelihood phylogenetic tree of the target group inferred from ITS. Supported branches with bootstrap values 70% or higher are thickened and support values are given.

Two of the three singleton sequences, namely those from China and Switzerland, could represent additional species, based on the observed base differences, but more molecularly congruent specimens are needed to confirm this hypothesis; the possible existence of an additional species of this complex in Europe would be particularly intriguing, although the lectotype material of *R. humilis* from Sweden (see below) already suggests the existence of further taxa within the *R. pollinaria* complex in Europe. The singleton sequence

from Canada cannot be currently assessed due to the observed quality issues; it could either represent *R. pollinaria* s.str., then being the only confirmed specimen of that species in North America, or *R. labiosorediata*. We were able to examine the specimen, which is not well developed but agrees in morphology with *R. labiosorediata*, which would be consistent with its geographic origin.

Our results underline that the ITS barcoding locus works rather well for species delimitation within *Ramalina*, also because it provides by far the largest data set available in terms of taxonomic and geographic range and sampling density. We have therefore not attempted to add further loci in this particular case, since the species introduced here are well-resolved and supported and we are unaware of instances where a well-resolved ITS phylogeny at the species level would be contradicted by other markers. However, there are other cases within *Ramalina* that show a more complex pattern or limited resolution based on ITS, such as *R. farinacea*, and additional loci will be necessary in such cases.

### **5.3.2 Taxonomy**

#### ***Ramalina europaea* Gasparyan, Sipman & Lücking spec. nov. (Fig. 5.2)**

[MycoBank MB 819386]

**Diagnosis:** Differing from *Ramalina pollinaria* in the small, punctiform, often terminal soralia starting out on small, spine-like branchlets.

**Typus:** Sweden, Södermanland, Lerbo parish, 1.7 km NE Lerbo church, 600 m SW Dagöholm, the Natura 2000 habitat Dagöholmsbackarna, W of the road, wooded meadow with very old *Quercus robur*, on *Quercus robur*, 30 m, 58°59.120' N, 16°27.254' E, grazed, 07 May 2015, A. Gasparyan s.n. (B 60 0201019—holotypus).

**Description.** *Thallus* corticolous or saxicolous, fruticose, pale green to green; branches erect to subpendulous, about 1(–3) cm long, flattened, solid, up to 3 mm wide, several times dichotomously to palmately branched, often developing numerous tiny, irregular, sometimes spine-like proliferations with punctiform soralia on the top. *Soralia* lateral and terminal, in mature thalli terminally often on widened, somewhat vaulted lobes, but never becoming large and labriform; soredia granular, 50–70 µm in diam., easily abraded and leaving a bare, whitish surface of the soralia. *Apothecia* very rare, 1–3 mm wide, hymenium ca. 40 µm thick,

asci ca. 35-40 x 12 µm, clavate, *Bacidia*-type, ascospores (10-)12–14(-15) x 4-4.5(-5) µm (20 healthy-looking spores measured), straight or slightly curved.

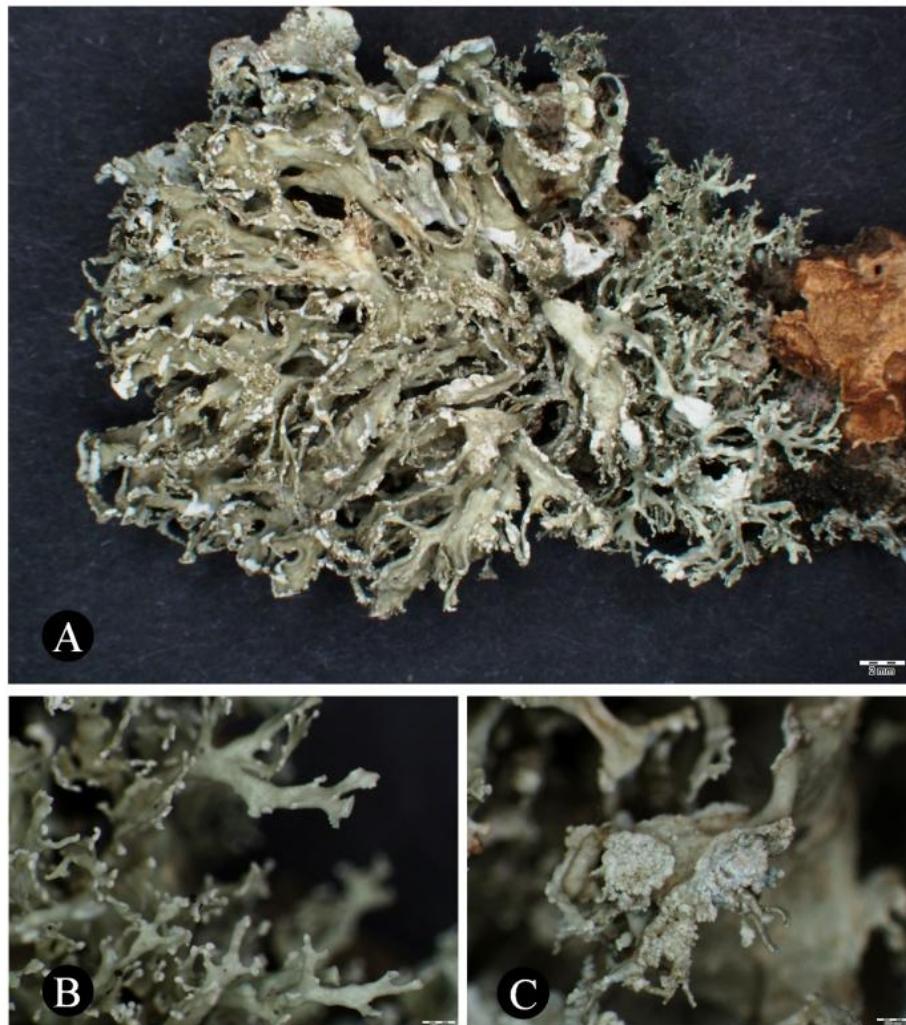
**Chemistry.** Medulla and soralia C-, K-, KC-, Pd-, UV-; TLC: usnic and evernic acids.

**Etymology.** The epithet refers to the wide European (to Eurasian) distribution of the species.

**Distribution and ecology.** The new species was found in broad-leaved and mixed forests. It grows on various rocky substrates (limestone, granite rocks) and various phorophytes (*Fagus* spp., *Carpinus* spp.). Currently it is known from Armenia, Austria, Finland, Russia (European part and North Caucasus) and Sweden, where it seems to be more common than *R. pollinaria* s.str.

**Notes.** This newly recognized species is phylogenetically distinct from *Ramalina pollinaria* s.str., and since both have a broadly overlapping, widely European to Eurasian distribution, they cannot be considered infraspecific lineages of a single species and hence are here recognized separately at the species level. Well-developed specimens show the morphological differences rather clearly, with *R. europaea* forming small, spine-like branchlets near the lobe tips on which the soralia start out as punctiform, granular structures, whereas in *R. pollinaria*, the soralia are more irregular, larger, and farinose from the beginning; older soralia are rather similar. Young or old or depauperate specimens might be difficult to identify with certainty.

We studied a number of historical names previously placed in synonymy with *Ramalina pollinaria* s.lat. or considered infraspecific taxa within the latter, to check whether a name was available for the two new species introduced here, particularly *R. europaea*. While part of the names were found to be conspecific with *R. pollinaria* s.str. (see below), others had already been placed in synonymy with other, unrelated species (see Appendix C2 Supplementary Table 1). The status of several infraspecific names remains uncertain (see Appendix C2 Supplementary Table 1), but these have no priority at the species rank. The only species-level name with uncertain status remaining and potentially predating *R. europaea* is *Lichen squarrosus* Pers. (1795). This name was variously treated as synonym of *R. pollinaria* s.lat. (Acharius 1810) and *R. polymorpha* (Lilj.) Ach. (Zahlbruckner 1930) and it would actually threaten the name *R. pollinaria* if synonymous, as it seems to have been published prior to *L. pollinarius* Westr. in the same year.



**FIGURE 5.2A-C.** A. Thallus of *R. europaea* (Gasparyan H-13-pl.03-06 [B 60 0201033]). B. Spine-like branchlets with small, punctiform soralia (Gasparyan H-13-pl.03-06 [B 60 0201033]). C. Granular soredia of *R. europaea* (Gasparyan s.n. [B 60 0201025]).

It has been overlooked that the name *Lobaria squarrosa*, usually attributed to Hoffmann (1796) and treated as taxon different from *Lichen squarrosus* (Zahlbrückner 1930), is actually a homotypic combination based on the latter and must be cited *Lobaria squarrosa* (Pers.) Hoffm. Likewise, the correct author citations for the names *Physcia squarrosa* and *Ramalina squarrosa* are *P. squarrosa* (Pers.) DC. and *R. squarrosa* (Pers.) Oxner. Persoon (1795) did not cite any material for *Lichen squarrosus*, and there is no material in L (M.

Brand, pers. comm. 23 November 2016), but from the protologue it is clear that the taxon was collected in Germany in the Solling forest (Lower Saxony), on the bark of beech trees (*Fagus sylvatica*). The description and ecology suggest that the name *Lichen squarrosus* refers to what is now known as *R. farinacea* (L.) Ach.; therefore, in order to stabilize nomenclature, we neotypify the name *Lichen squarrosus* with a well-developed, historical specimen collected in 1861 on beech in North Rhine-Westphalia, not far from the Solling, deposited in B (Fig. 5.3): *Lichen squarrosus* Pers., *Annalen der Botanick* 14: 35 (1795); *Lobaria squarrosa* (Pers.) Hoffm., *Deutschlands Flora* 2: 139 (1796); *Physcia squarrosa* (Pers.) DC. in Lamarck & de Candolle, *Flore Française, Edn. 3 (Paris)* 2: 398 (1805); *Ramalina squarrosa* (Pers.) Oxner, *Akademija Nauk Ukrainskoi RSR Institut Botaniki* (1937); type: Germany, North Rhine-Westphalia, 'Buchen in der Nähe der Bruchhauser Steinen', 14 April 1861, *Lahm s.n.* (B 60 0059371—neotype, here designated; MycoBank MBT374159). This neotypification makes *Lichen squarrosus* a synonym of *R. farinacea*.



**FIGURE 5.3.** Neotype of *Lichen squarrosus* Pers. (= *Ramalina farinacea*) in B. Scale = 1 cm (large divisions), 1 mm (small divisions).

*Additional specimens examined.* Armenia. Kotayk prov.: Aghavnadzor, mixed forest, on bark of deciduous tree, 1708 m., 40°34'22" N, 044°41'48" E, 08 September 2013, A. Gasparyan H-13-pl.03-06 [B 60 0201033]; id. [B 60 0201034]. Artavaz, mixed forest, on stone, 1767 m., 40°36.80" N, 044°34.11' E, 05 July 2015, A. Gasparyan s.n. [B 60 0201022]; id., [B 60 0201025]. Lori prov.: Jiliza forestry, road to the village Jiliza, coniferous stand in deciduous forest, on bark of *Pinus* sp., 1687 m., 41°10'81" N, 044°38'27" E, 05 August 2014. A. Gasparyan T-14-34g [B 60 0201030]; id., mixed forest, on bark of deciduous tree, 1688 m., 41°10'56" N, 044°37'85" E, 05 August 2014, A. Gasparyan T-14-35 [B 60 0201024]. Syunik prov.: Shikahogh State Reserve, deciduous forest, on *Quercus* sp., 1700 m., 39°00.77" N, 046°30.62" E, 09 September 2014. A. Gasparyan S-14-23 [B 60 0201031]. Tavush prov.: Dilijan National Park., 1218 m., 40°43'49" N, 044°58'26" E, 26 August 2013. A. Gasparyan D-13-pl.12.01 [B 60 0201018]. Koghb, Noyemberyan forestry, old beech forest, on *Fagus orientalis*, 1117 m., 41°07.585' N, 044°54.662' E, 26 July 2015. A. Gasparyan s.n. [B 60 0201027]; id., [B 60 0201028]; id., old beech forest, on *Fagus orientalis*, 1091 m., 41°08'11" N, 044°55'46" E, 23 August 2014, A. Gasparyan T-14-22 [B 60 0201021]. — Russia: Republic of Adygeja: Maikopskij Rajon, unterhalb der Poljana Firsowa in Richtung Tal des Mal. Sachrai., on *Carpinus* sp., 1270 m., 44°05'27" N, 040°24'33" E, 6 August 2012, V. Otte s.n. [GLM-L 34284]. Chelyabinsk region: Vicinity of Miass town, Ilmenskiy state reserve, 1.6 km to NW from Miasovo biological station, granite rocks under pine forest canopy, on granite rocks, 309 m., 55°08'14.7" N, 60°13.977' E, 13 August 2013, A. Paukov 2337 [UFU L-2517]. Ashinskiy district, appr. 3 km to 314 degrees (NW) from Biyanka village, mixed forest with *Picea abies*, *Populus tremula*, *Tilia* and *Quercus* sp., on *Tilia cordata*, 432 m. 55°12'00.9" N, 57°37'42.5" E, 04 July 2013, A. Paukov AGP20130704-69 [UFU L-1684]. Sverdlovsk region: Shalinskiy district, vicinity of Staroutkinsk settlement, "Reka Chusovaya" natural park, Vinokurennyi kamen' rock, limestone outcrops on left bank of Chusovaya river, on limestone, 55°08'14.7" N, 60°13.977' E, 18 July 2012, A. Paukov 2340 [UFU L-2520].

***Ramalina labiosorediata* Gasparyan, Sipman & Lücking spec. nov. (Fig. 5.4)**

[MycoBank MB 819387]

Differing from *Ramalina pollinaria* and *R. europaea* in the almost exclusively terminal soralia formed on the tips of normal lobes, originating from the underside and becoming irregularly labriform.

**Typus.** USA, Arizona, Coconino County, 10 miles S of Flagstaff in Kelly Canyon just upstream from junction with Pumphouse Wash, on shaded sandstone face, 35°03.533' N, 111°42.995' W, 6345 ft., 3 October 2012, David C. Thornburg 837 (NY 01808147, holotypus).

**Description.** *Thallus* corticolous or saxicolous, fruticose, pale green to green; branches erect to ascending, more rarely subpendulous, about 1(–3) cm long, flattened, solid, up to 1(–2) mm

wide, a few times dichotomously to palmately branched, at age apically often widened and producing terminal, lip-shaped, 1 to over 5 mm wide soralia, in addition with apical, irregular or granular, terete, spine-like proliferations producing granules in punctiform apical soralia. *Soralia* elongate to rounded, lateral and terminal, there often lip-shaped; soredia granular, 50–70(–90) µm in diam. *Apothecia* not seen.

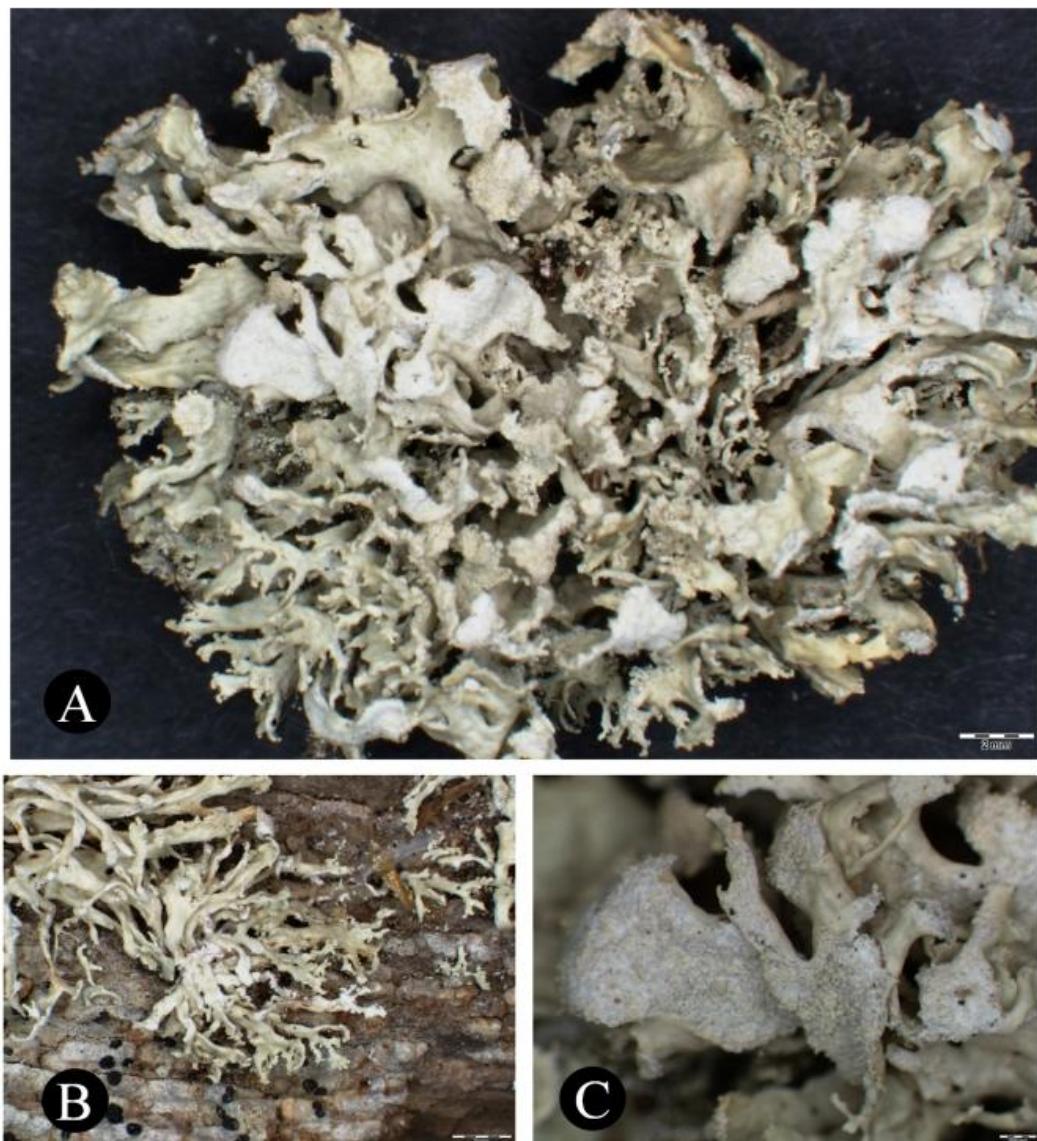
**Chemistry.** Medulla and soralia C–, K–, KC–, Pd–, UV–; TLC: usnic and evernic acids.

**Etymology.** The epithet is partially derived from the latin word "labium" and refers to the eventually lip-shaped soralia.

**Distribution and ecology.** The new species was found in North America (Canada and USA), where it seems widespread. It grows in deciduous, coniferous and mixed forests, on various phorophytes (*Acer saccharum*, *Liriodendron* sp., *Nyssa* sp., *Thuja* sp.), and also on rocky substrates.

**Notes.** In a previous study of North American *Ramalina*, *R. pollinaria* was already suggested to be polyphyletic (Timsina *et al.* 2012), forming two unrelated clades, with one sequence (JQ003096) clustering with European specimens and the other three (JQ003097, JQ003098, JQ003099) close to the western North American *R. menziesii*. We therefore initially expected the North American clade now separated from *R. pollinaria* s.str. to correspond to the separate clade identified in the previous study. However, three of the four specimens studied by Timsina *et al.* (2012) are phylogenetically unrelated to the *R. pollinaria* clade; in our large scale phylogeny, these three sequences clustered close to another North American species, *R. culbersoniorum* (Supplementary Fig. 1), although they are morphologically and chemically fairly similar to *R. pollinaria*. Only one specimen, Normore 3748 (JQ003096), belongs to the *R. pollinaria* clade and it morphologically resembles *R. labiosorediata*, but its ITS sequence is too short (216 base pairs) and has quality issues that prevent for it to be included in that species with certainty (see above).

The morphological differences of *Ramalina labiosorediata* with its terminal, labriform soralia originating from the underside versus predominantly laminal to marginal, elongate soralia in *R. pollinaria* s.str. are clearly visible in the studied and sequenced material, even in somewhat depauperate specimens.



**FIGURE 5.4A–B.** Thallus of *R. labiosorediata* (A: Lendemer 14495 [NY 00974764]; B: Tripp 671 [NY 01117934]). C. Labriform soralia formed on the tips of lobes, originating from the underside (Lendemer 14495 [NY 00974764]).

In treatments of *R. pollinaria* for North America, e.g. Brodo *et al.* (2001), the terminal, labriform soralia were mentioned or illustrated as diagnostic feature for the taxon, without recognizing that the soralia are differently shaped in European material of *R. pollinaria*. At this point, we presume that *R. pollinaria* s.str. is not present in North America. A confirmation of this was left out because it would require reexamination of a large amount of material. The CNALH portal (2016) alone has well over 600 specimens identified under that name from Canada and the USA.

No historic name was found based on material that would be conspecific with *Ramalina labiosorediata*.

*Additional specimens examined* (\* = not sequenced). **Canada:** *Ontario:* Prince Edward County, Sandbanks provincial park, mature deciduous forest along lake shore, on *Acer saccharum*, 76 m., 43°53'30.79" N, 077°16'49.59" W, 10 August 2009, C. Lewis 341 [CANL 124119]. Bruce Count., Fathom Five National Marine Park, Flowerpot Island, Flowerpot Loop Trail, *Thuja*-dominated forest cover Silurian dolomite, with extensive outcrops and costal bluffs, on *Thuja* and on cliffs, ca. 180 m. 45°18'14" N, 081°36'43" W, 28 September 2008, J. C. Lendemer 14495 [NY 00974764]. \*York County, Nashville, on elm tree, E. side, near base, 4 April 1959. R. F. Cain s.n. [NY 01544870]. — **USA:** *North Carolina:* Haywood County, Great Smoky Mountains National Park, E slopes above (E of) Caldwell Fork, along E portions of Boogerman Loop Trail, mixed rich cove forest and old growth homesteads with *Acer saccharum*, *Tsuga canadensis*, *Liriodendron* and *Hamamelis* grading to upland *Carya*, *Quercus montana*, mature *Liriodendron* forest with *Rhododendron* understory, on *Acer saccharum*, 3000-3600 ft., 35°36'20" N, 083°05.50' W to 35°37'30" N, 083°05.30' W. 4 August 2009, J. C. Lendemer 18826 & E. Tripp [NY 01118742]. \*Haywood County, Great Smoky Mountains National Park. Double Gap trail between Double Gap and Caldwell Fork, rich hardwoods (*Prunus*, *Betula*, *Aesculus*, *Acer*, *Carya*) with large trees, grading to acid *Tsuga* (large dead trees) - *Rhododendron* forest, on hardwood, 35°34'19" N, 83°06'03" W to 35°35'36" N, 83°07'03" W, 8 October 2011, Erin A. Tripp 2017 with James Lendemer, Naveed Davoodian, Zev Reuter, Andrei Moroz [NY 1685380]. *Pennsylvania:* Wayne County, Lacawa Sanctuary, Wallenpaupack Ledges, N shore of Lake Wallenpaupack, Paupack Township, massive sandstone ledges with maple (*Acer*) – hickory (*Carya*)—Oak (*Quercus*) forest, on rock, overhang, 1300-1450 ft., 41°23' N, 075°17' W, 01 July 2008. J. C. Lendemer 12329 [NY 00977236]. *Tennessee:* Blount County, Great Smoky Mountains National Park, Gregory Bald Trail, Parsons, Branch Rd. to Sheep Pen Gap (4 mi), acid uplands (*Pinus*, *Quercus*, *Kalmia*) and occasional rich cove forest (*Liriodendron*, *Quercus*, *Halesia*) with sparse old growth, on old *Liriodendron*, 2776-4616 ft., 35°31'11" N, 083°52'28" W to 35°32'34" N, 083°53'36" W. 12 October 2010, J. C. Lendemer 26793 & A. Moroz, E. Tripp [NY 01219716]. Cocke County, Great Smoky Mountains National Park, Gabes Mountain Trail, 0-2 mi E of jct with Maddron Bald Trail, acidic *Tsuga*, *Oxydendrum*, *Quercus montana*, *Acer saccharum* and *Nyssa* forest with *Rhododendron* understory, on *Nyssa*, 2400-3200 ft., 35°45'10" N, 083°16'30" W to 35°44'50" N, 083°14'50" W, 5 August 2009, E. A. Trip 671 & J. C. Lendemer [NY 01117934]. \*Sevier County, Brushyx Mt. trail, buckeye bark, 4500 ft., 29 March 1936, S. A. Cain s. n. [NY 02375823]

***Ramalina pollinaria* (Westr.) Ach., (Fig. 5.5–5.7)**

[MycoBank MB 356436]

Acharius, *Lichenographia Universalis*: 608 (1810); *Lichen pollinarius* Westring, *Kongliga Svenska Vetenskaps-Akademiens Handlingar* 16: 56 (1795); type (Fig. 5.6): Sweden, unknown locality, unknown date and collector but collected prior to 1810, no. 6 (BM 001107258—neotype!, here designated; MycoBank MBT374161); Sweden, Lerbo parish, the Natura 2000 habitat Dagöholmsbackarna, on *Quercus robur*, 7 May 2015, Gasparyan s.n. (B 60 0201017—epitype!, here designated; MycoBank MBT374163; ITS barcode sequence: KY362419).

*Ramalina pollinaria* f. *elatior* Ach., *Lichenographia Universalis*: 608 (1810); *R. elatior* (Ach.) Röhl., *Deutschlands Flora (Frankfurt)* 3(2): 140 (1813); type (Fig. 7): Sweden, unknown locality, unknown date and collector but collected prior to 1810 (H-ACH 1831E = H 9 502 934—lectotype!, here designated; MycoBank MBT374160).

*Ramalina farinacea* var. *bolcana* A. Massal., *Memorie Lichenographiche*: 67 (1853); *R. farinacea* f. *bolcana* (A. Massal.) Zahlbr., *Catalogus Lichenum Universalis* 6: 468 (1930); type: Italy (not seen, fide Nimis 2016).

*Ramalina fennica* Räsänen, *Annales Academiae Scientiarum Fennicae, Ser. A, 34(4)*: 27 (1931); type: Russia, 13 May 1923, Räsänen s.n. (H 9 500 246—holotype!).

*Ramalina pollinaria* var. *sortavalensis* Räsänen, *Annales Botanici Societatis Zoologicae-Botanicae Fenniae 'Vanamo'* 12(1): 40 (1939); type: Russia, 14 May 1923, Räsänen s.n. (H 9 500 274—holotype!).

**Description.** *Thallus* corticolous, fruticose, pale green to green; branches subpendulous, about 1–5 cm long, flattened, solid, a few times or more densely dichotomously to palmately branched, the tips up to 2(–3) mm wide, rarely with spine-like proliferations. *Soralia* elongate, marginal to subterminal, spreading over the underside, becoming more roundish, more crowded terminally, small soralia may develop laminally; soredia farinose to subgranular, 30–50 µm in diam. *Apothecia* not seen in the sequenced specimens.

**Chemistry.** Medulla and soralia C–, K–, KC–, Pd–, UV–; TLC: usnic and evernic acids.

**Distribution and ecology.** Specimens fitting this refined concept of the species were seen from Armenia, Belarus, the Netherlands, Russia and Sweden. It grows on various phorophytes (*Acer platanoides*, *Carpinus betulus*, *Fagus orientalis*, *Quercus robur*, *Tilia cordata*, *Robinia* sp.).

**Notes.** Differences between *Ramalina pollinaria* and the two newly described species are discussed under the previous two species entries and are outlined in the key below. The previously separated, somewhat related species, *R. yasudae* and *R. sekika*, form separate,

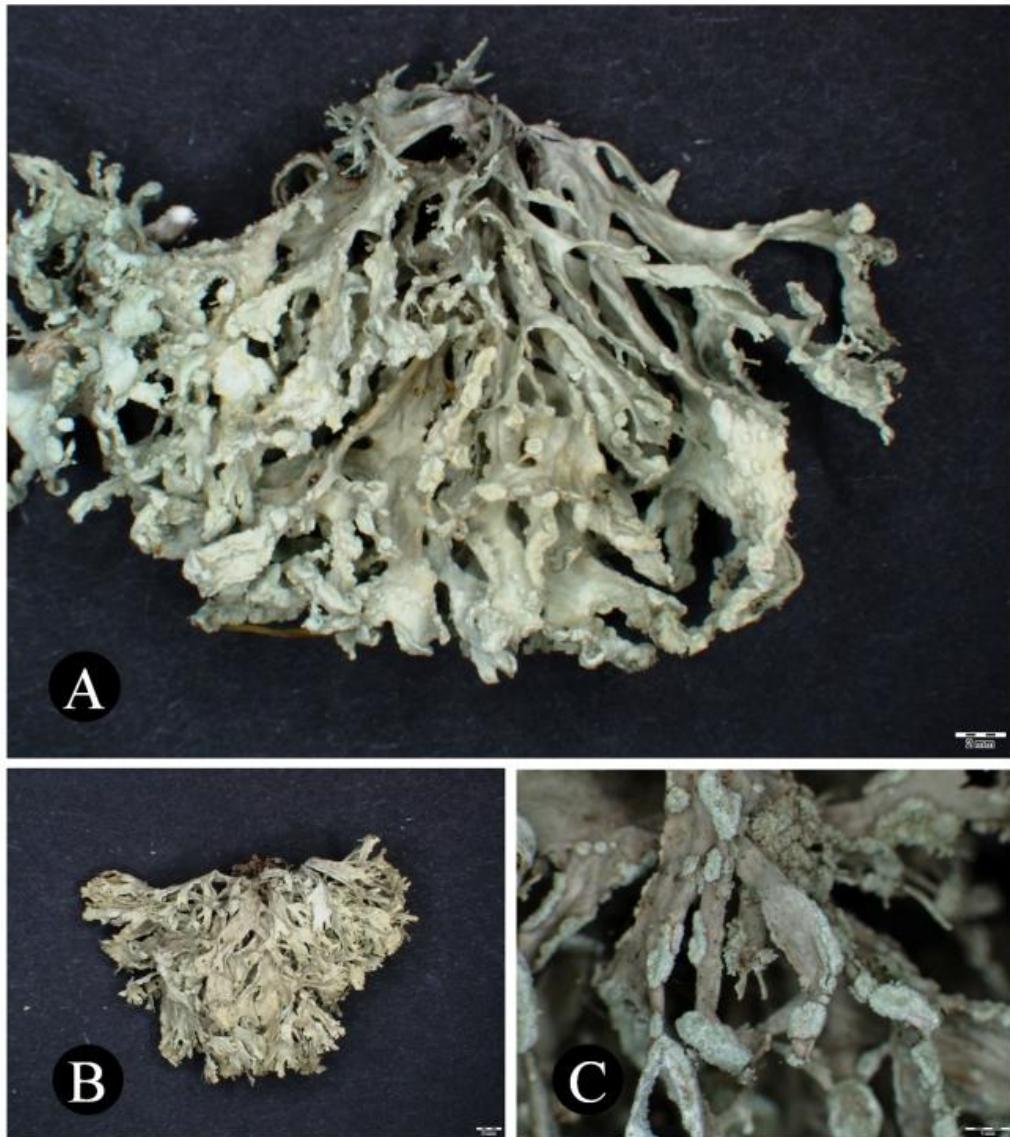
distinct, monophyletic clades out of the *R. pollinaria* clade (see Appendix 3 Supplementary Fig. 5.1) and differ from *R. pollinaria* also by the larger size of the soredia (100–150 µm in diam.), their distribution (*R. yasudae* and *R. sekika* are known from eastern Asia), and in the case of *R. sekika* also the chemistry (usnic, salazinic and sekikaic acids) (Ohmura *et al.* 2008, Kataeva 2014).

The identity of *Ramalina pollinaria* s.str. with the clade identified here under that name was assessed by comparison with the sheet containing the original neotype of *R. pollinaria* selected by Krog & James (1977). That specimen (H-ACH 1831D) forms part of the material which served as base for *R. pollinaria* var. *humilis* Ach., whereas according to Acharius (1810) and subsequent authors, typical *R. pollinaria* corresponds more to var. *elatior* Ach. Acharius (1810) divided *R. pollinaria* into the two varieties var. *elatior* and var. *humilis*, and technically, one of them would correspond to the autonym, var. *pollinaria* [ICN 26.1]. However, since the epithet *pollinarius* had been established 15 years prior and by a different author (Westring 1795), the autonym should be established based on that epithet, whereas the two varieties var. *elatior* and var. *humilis* are best considered separate, valid varieties, since at the time of their description (Acharius 1810), no type material had been established for any of these names [ICN 26.2, Ex. 4].

There are two sheets of material studied by Acharius on which he based his concept of *R. pollinaria*, namely the aforementioned H-ACH 1831(A–K) and also BM 001107258(5–11), the latter given by Acharius to the Linnean Society in London in 1808. While the H-ACH sheet reflects Acharius's (1810) concept of dividing the species into var. *elatior* and var. *humilis*, the BM sheet, which also bears the number '804', was apparently assembled prior, as Acharius likewise divided the species into two varieties but used unpublished names (var. 'major' and var. 'minor'). The specimen 1831D from the H-ACH sheet, selected as neotype for *R. pollinaria* by Krog & James (1977), is morphologically the most aberrant material, due to its unique, tree-like branching pattern, and it corresponds to another specimen illustrated by Dillenius (1741: plate XXI, fig. 56A) that Acharius (1810) had also included in var. *humilis*.

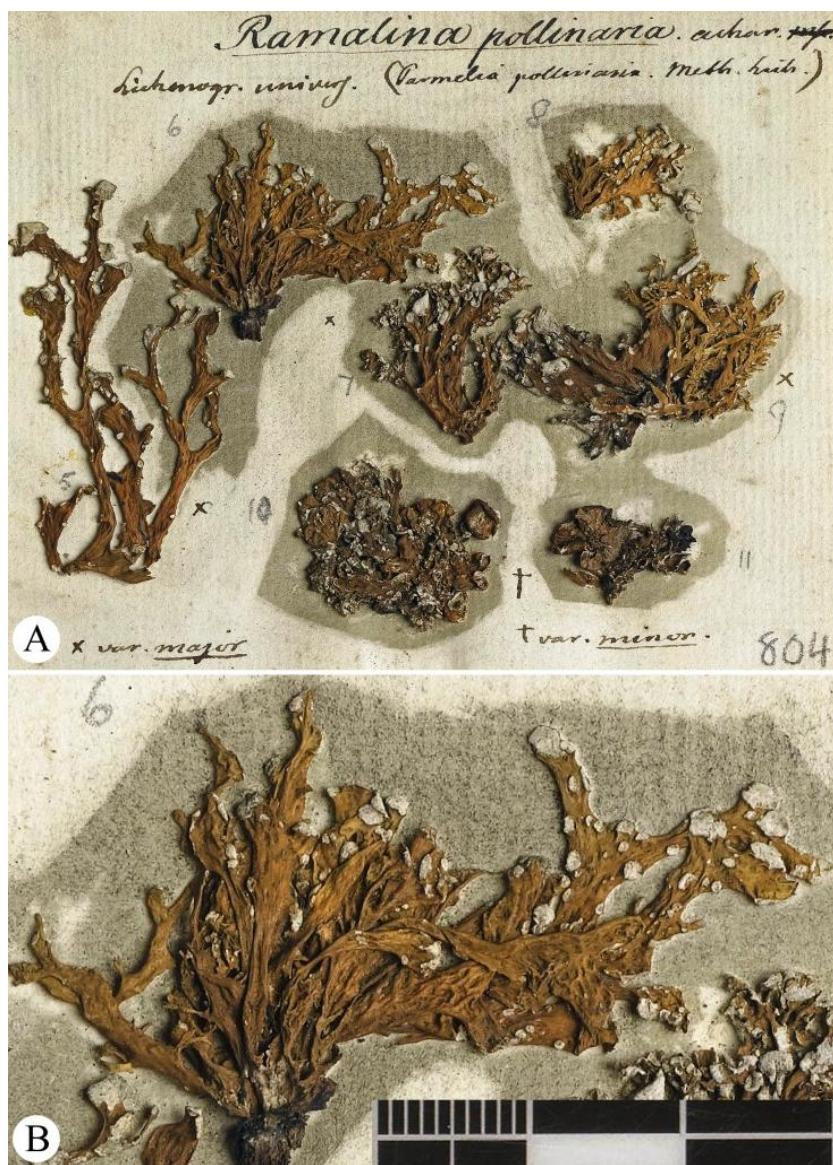
All remaining specimens on the H-ACH sheet and all specimens on the BM sheet except no. 10 and 11 (which belong to *R. baltica* Lettau) represent *R. pollinaria* s.str. both in historical and modern treatments and in our revised phylogenetic concept. Since the neotype selected for *R. pollinaria* by Krog & James (1977) does not correspond neither to the historical nor the modern concept of *R. pollinaria* (≡ f. *pollinaria*, var. *pollinaria*), it is here superseded by a

new neotype [ICN. 9.14, 9.19b, 9.19c] from the BM sheet (Fig. 6), as the H-ACH sheet covers material from which the lectotypes of var. *elatior* and var. *humilis* should be selected.

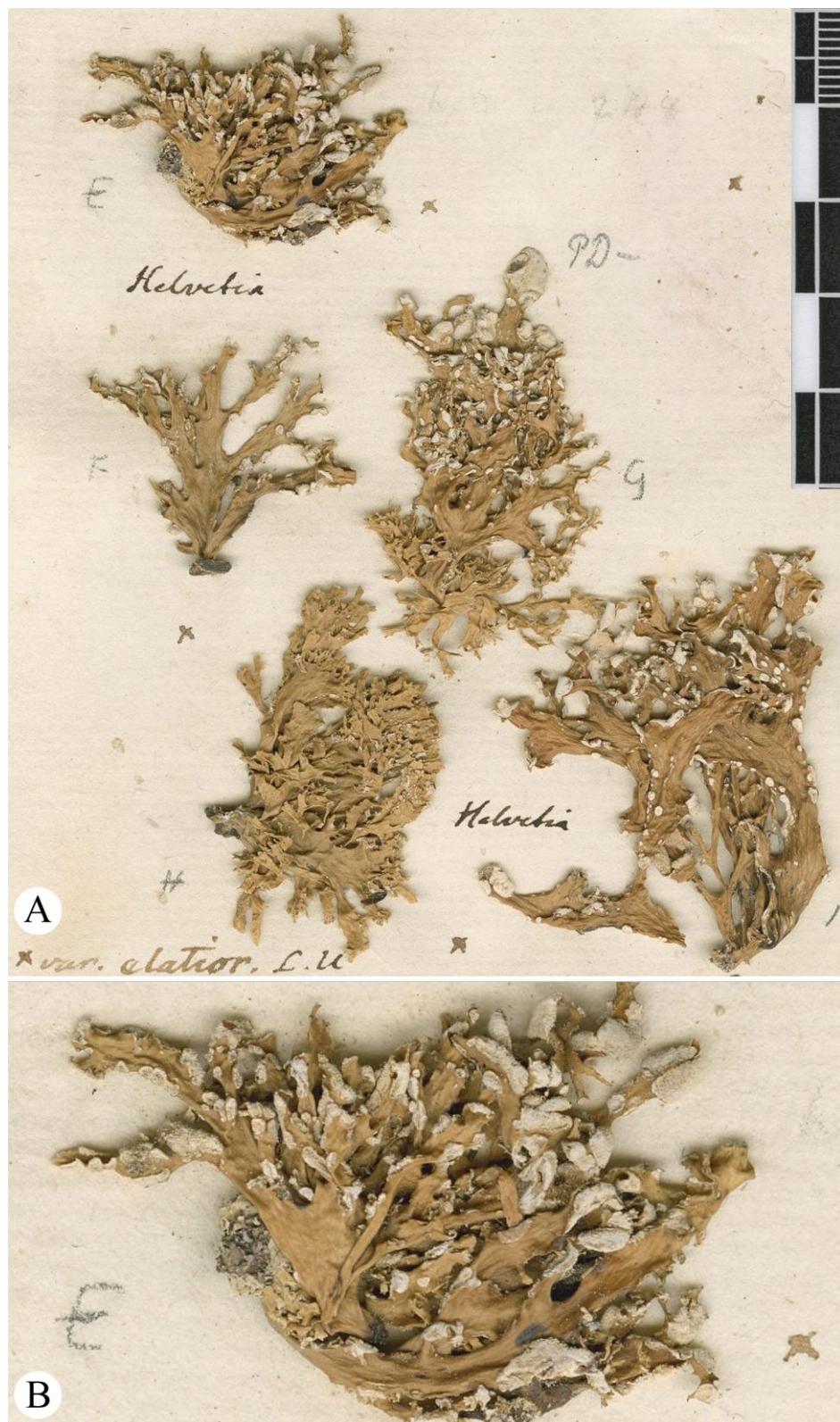


**FIGURE 5.5A–B.** Thallus of *R. pollinaria* (A: Gasparyan s.n. [B 60 0201023]; B: Yatsina MSK-L 12660 [B 60 0201032]). C. Irregular elongate soralia of *R. pollinaria* (Gasparyan S-14-21 [B 60 0201026]).

The lectotype of the former is indicated above (Fig. 5.7); the lectotype of var. *humilis* is here designated as identical with the former neotype of *R. pollinaria* (Krog & James 1977), specimen 1831D (Fig. 5.8): *R. pollinaria* var. *humilis* Ach., *Lichenographia Universalis*: 609 (1810); *R. humilis* (Ach.) Röhl., *Deutschlands Flora (Frankfurt)* 3(2): 140 (1813); Sweden, unknown locality, unknown date and collector but collected prior to 1810 (H-ACH 1831D = H 9 502 933—lectotype!, here designated; MycoBank MBT374162). This lectotypification is made since this specimen represents an aberrant morphology resembling the illustration in Dillenius (1741) to which Acharius (1810) refers, and it might represent a yet unrecognized taxon, for which the name *R. humilis* is then available.



**FIGURE 5.6.** New neotype of *Lichen pollinarius* Westr. (= *Ramalina pollinaria*) in BM. A. Entire set of specimens from sheet no. 804. B. Neotype specimen no. 6. Scale = 1 cm (large divisions), 1 mm (small divisions). Image reproduced from the *Global Plants* project with permission by Holger Thüs.



**FIGURE 5.7.** Lectotype of *Ramalina pollinaria* var. *elatior* Ach. (= *Ramalina pollinaria*) in H. A. Entire set of specimens of var. *elatior* from sheet no. 1831. B. Lectotype specimen E. Scale = 1 cm (large divisions), 1 mm (small divisions). Image reproduced from the *Global Plants* project with permission by Soili Stenroos.



**FIGURE 5.8.** Lectotype of *Ramalina pollinaria* var. *humilis* Ach. (= *Ramalina humilis*) in H. A. Entire set of specimens of var. *humilis* from sheet no. 1831. B. Lectotype specimen D (this specimen had previously been designated as neotype of *R. pollinaria*). Scale = 1 cm (large divisions), 1 mm (small divisions). Image reproduced from the *Global Plants* project with permission by Soili Stenroos.

As outlined above, several synonyms and numerous infraspecific names have been established for *Ramalina pollinaria*, which are potentially available for new species distinguished within this complex. We revised all names available at the species level and many of the infraspecific names. Of the latter, about half could be studied and only three of them belong in the *R. pollinaria* clade (see Appendix C2 Supplementary Table 5.1), all being synonyms of *R. pollinaria* s.str. and different from *R. europaea*, namely *R. pollinaria* f. *elatior* Ach., *R. pollinaria* var. *bolcana* (A. Massal.) Zahlbr., and *R. pollinaria* var.

*sortavalensis* Räsänen (see above). Surprisingly, 14 of the 17 studied names (82%) are synonyms of or represent other species (see Appendix C2 Supplementary Table 5.1), which underlines the notion that these historical, infraspecific names largely are not based on a sound biological understanding of species and infraspecies level taxonomy. We therefore follow the philosophy that such historical, infraspecific names should not receive attention and be subsumed in synonymy whenever possible, except where their infraspecific status is based on biological reasoning, for instance when it was established for a morphologically distinct, geographically restricted population. An example would be *R. pollinaria* f. *chilena* Nyl., subsequently recognized as separate species, *R. chilena* (Nyl.) Kashiw.

An additional name that was partially based on material originally identified as *Ramalina pollinaria* is *R. fallax* Motyka (1960; the paratype from Sweden is Malme's *Lichenes Suecici Exsiccati* 578). The holotype material of that name was later synonymized with *R. farinacea* by Krog & James (1977), as it agrees morphologically and contains protocetraric acid. The names *R. maciformis* (Delise) Bory and *R. evernioides* Nyl. have sometimes been discussed as possibly conspecific with *R. pollinaria*; however, both are now considered synonymous under *R. maciformis*, which is a distinct species differing from *R. pollinaria* in its morphology, chemistry (bourganic and salazinic or norstictic acid), and ecology, being a Mediterranean species (Krog & Østhagen 1980).

*Additional specimens examined.* **Armenia:** Syunik prov.: Shikahogh State Reserve, mixed deciduous forest, on bark of deciduous tree, 1454 m., 39°01.20' N, 046°30.81' E, 09 September 2014, A. Gasparyan S-14-21 [B 60 0201026]. Road to the city Berd, mixed forest, on bark of deciduous tree, 1382 m., 40°52'65" N, 045°16'27" E, 18 July 2014, A. Gasparyan T-14-01 [B 60 0201020]. Tavush prov.: Dilijan National Park, road to lake Gosh, deciduous forest, on *Fagus orientalis*, 1423 m., 40°43.26' N, 045°00.88' E, 18 July 2014, A. Gasparyan T-14-08 [B 60 0201029]. — **Belarus:** Minsk prov.: Cherevenskiy district, Rovanichi, in the park, on *Acer platanoides*, 53°53'05.1" N, 28°36'34.6" E, 17 October 2013, A. P. Yatsina MSK-L 12660 [B 60 0201032]. — **Russia:** Bashkortostan: Beloretskiy district, Revet', 250 m., *Tilia* stand along small brook, on *Tilia cordata*, 54°12'90.7" N, 57°34'32.0" E, 08 August 2015, A. Paukov 2335 [UFU L-2515]. — **Sweden:** Södermanland: Stora Malm parish, Stora Malm church yard with scattered old deciduous trees, on deciduous tree bark, 40 m., 58°58.062' N, 16°21.658' E, 07 May 2015. A. Gasparyan s.n. [B 60 0201023]. Lerbo parish, 1.7 km NE Lerbo church, 600 m SW Dagöholm, the Natura 2000 habitat Dagöholmsbackarna, W of the road, grazed wooded meadow with very old *Quercus robur*, on *Quercus robur*, 30 m., 58°59.120' N, 16°27.254' E, 07 May 2015, A. Gasparyan s.n. [B 60 0201017]. — **The Netherlands:** Utrecht prov.: Amersfoort, Bergkwartier, on *Robinia* bark in park, 52°09' N, 05°21' E, 22 January 2016, A. Aptroot 74529 [ABL].

### 5.3.3 Key to the species of the *Ramalina pollinaria* group

- 1 Soralia terminal but originating from the underside, irregularly lip-shaped and rather large; North America ..... ***Ramalina labiosorediata***
- Soralia marginal to laminal, elongate to rounded, if terminal then starting out as small, punctiform structures on small, spine-like branchlets; Europe (Eurasia) ..... 2
- 2(1) Soralia starting out terminally as small, punctiform structures on small, spine-like branchlets; soredia granular, 50–70 µm in diam..... ***Ramalina europaea***
- Soralia from the beginning subterminal to laminal-marginal, irregularly elongate, not starting out on small branchlets and such branchlets absent or rare; soredia farinose, < 50 µm in diam. .... ***Ramalina pollinaria* s.str.**

Note. If thallus very small and branching in tree-like rather than shrub-like fashion, i.e. with lateral branches branching off consecutively towards the tip of the thallus rather than mainly from close to the base, compare *Ramalina humilis* (see above).

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## **List of publications and own contributions**

### **Contribution to Chapter 2**

Gasparyan, A. & Sipman, H. J. M. (2016) The Epiphytic Lichenized Fungi in Armenia: Diversity and Conservation. *Phytotaxa* 281 (1): 1–68. ISSN 1179-3163.

DOI: <http://dx.doi.org/10.11646/phytotaxa.281.1.1>

URL: <http://biotaxa.org/Phytotaxa/article/view/phytotaxa.281.1.1>

**Own contribution:** Implemented the field work, identified the lichens and carried out laboratory work, data analysis, wrote the manuscript together with co-author.

### **Contribution to Chapter 3**

Gasparyan, A. & Sipman, H. J. M. (2013) New lichen records from Armenia. *Mycotaxon* 123: 491–492.

DOI: <http://dx.doi.org/10.5248/123.491>

URL: <http://www.mycotaxon.com/resources/checklists/Gasparyan-v123-checklist.pdf>

**Own contribution:** Implemented the field work, identified the lichens and carried out laboratory work, data analysis, wrote the manuscript together with co-author.

Gasparyan, A., Sipman, H. J. M. & Brackel, W. von. (2014) A contribution to the lichen-forming and lichenicolous fungi flora of Armenia. *Willdenowia* 44: 263–267.

DOI: <http://dx.doi.org/10.3372/wi.44.44208>

URL: <http://www.bioone.org/doi/pdf/10.3372/wi.44.44208>

**Own contribution:** Implemented the field work, identified the lichens and carried out part of laboratory work, data analysis, wrote the manuscript together with co-authors.

Gasparyan, A., Aptroot, A., Burgaz, A. R., Otte, V., Zakeri, Z., Rico, V. J., Araujo, E., Crespo, A., Divakar, P. K. & Lumbsch, H. T. (2015) First inventory of lichens and lichenicolous fungi in the Khosrov Forest State Reserve, Armenia. *Flora Mediterranea* 25: 105–114.

DOI: <http://dx.doi.org/10.7320/flmedit25.105>

## List of publications and own contributions

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URL: <http://147.163.105.223/flora/25-105.pdf>

**Own contribution:** Together with co-authors implemented the field work, identified the lichens and carried out part of laboratory work, data analysis, wrote the manuscript.

Gasparyan A., Aptroot A., Burgaz A. R., Otte V., Zakeri Z., Rico V. J., Araujo E., Crespo A., Divakar P. K. & Lumbsch H. T. (2016) Additions to the lichenized and lichenicolous mycobiota of Armenia. *Herzogia* 29(2): 692-705.

DOI: <http://dx.doi.org/10.13158/heia.29.2.2016.692>

URL: <http://www.bioone.org/doi/abs/10.13158/heia.29.2.2016.692>

**Own contribution:** Together with co-author implemented the field work, identified the lichens and carried out laboratory work, data analysis and wrote the manuscript.

## **Contribution to Chapter 4**

Gasparyan, A. & Aptroot, A. (2016) *Verrucaria juglandis*, a new corticolous lichen species from Armenia. *Herzogia* 29: 103–107.

DOI: <http://dx.doi.org/10.13158/heia.29.1.2016.103>

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**Own contribution:** Together with co-author implemented the field work, identified the lichens and carried out laboratory work, data analysis and wrote the manuscript.

Zakeri, Z., Gasparyan A. & Aptroot A. (2016) A new corticolous *Megaspora* (*Megasporaceae*) species from Armenia. *Willdenowia* 46 (2): 245–251.

DOI: <http://dx.doi.org/10.3372/wi.46.46205>

URL: <http://www.bioone.org/doi/abs/10.3372/wi.46.46205>

**Own contribution:** Together with co-authors implemented the field work, identified the lichens and carried out part of the laboratory work (excluding phylogenetic research), data analysis and wrote the manuscript.

## **Contribution to Chapter 5**

List of publications and own contributions

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Gasparyan, A., Lücking, R. & Sipman, H. J. M. *Ramalina europaea* and *Ramalina labiosorediata*, two new species of the *Ramalina pollinaria* group (Ascomycota: *Ramalinaceae*), and new typifications for *Lichen pollinarius* and *Lichen squarrosus*. *The Lichenologist*. (accepted)

**Own contribution:** Implemented the field work, identified the lichens and carried out laboratory work, together with co-authors conducted data analysis and wrote the manuscript.

## **Appendices**

### **Appendix A. Supplementary material for Chapter 3**

#### **Appendix A.1 List of visited localities**

Gasparyan A., Aptroot A., Burgaz A. R., Otte V., Zakeri Z., Rico V. J., Araujo E., Crespo A., Divakar P. K. & Lumbsch H. T. (2016) Additions to the lichenized and lichenicolous mycobiota of Armenia. *Herzogia* 29(2): 692-705.

DOI: <http://dx.doi.org/10.13158/heia.29.2.2016.692>

**Appendix A.2** List of visited localities

**1.- ARMENIA: ARARAT:** Vedi, Urtsadzor, Khosrov Forest State Reserve, 39°59'07"N 44°53'51"E, 1390 m, 17-VI-2015, **a)** on *Ulmus* sp., **b)** on calcareous soil, **c)** on siliceous volcanic rocks, **d)** on limestone, **e)** on *Juniperus* sp., **f)** on bark, mixed forest of *Quercus*, *Juniperus* and *Acer*.

**2.- ARMENIA: ARARAT:** Vedi, Urtsadzor, Khosrov Forest State Reserve, 40°00'42"N 44°54'41"E, 1600 m, 17-VI-2015, **a)** on *Fraxinus excelsior*, **b)** on *Juniperus* sp., **c)** on siliceous volcanic rocks **d)** on limestone, **e)** on aquatic siliceous rocks, **f)** on rocks, **g)** on *Salix* sp., **h)** on *Quercus* sp., **i)** on *Acer monspessulanum*, **j)** on bark, mixed forest of *Quercus*, *Juniperus* and *Acer*.

**3.- ARMENIA: ARARAT:** Vedi, Urtsadzor, Khosrov Forest State Reserve, around a stream, 40°01'14"N 44°55'00"E, 1700 m, 17-VI-2015, **a)** on *Fraxinus excelsior*, **b)** on *Quercus macranthera*, **c)** on siliceous volcanic rocks, **d)** on soil, **e)** on *Juniperus* sp., **f)** on limestone, **g)** on bark, mixed forest of *Quercus*, *Juniperus* and *Acer*.

**4.- ARMENIA: ARARAT:** Vedi, Urtsadzor, Khosrov Forest State Reserve, top of the hill, 40°01'20"N 44°54'33"E, 1850 m, 17-VI-2015, **a)** on *Quercus macranthera*, **b)** on bark, mixed forest of *Quercus*, *Juniperus* and *Acer*.

**5.- ARMENIA: ARARAT:** Goght, Khosrov Forest State Reserve, entrance from Garni, riverside Azat, 40°06'25"N 44°45'16"E, 1300 m, 18-VI-2015, **a)** on *Quercus macranthera*, **b)** on *Populus* sp., **c)** on quartzitic rocks, **d)** on siliceous volcanic rocks, **e)** on *Fraxinus excelsior*, **f)** on *Juglans regia*, **g)** on *Cornus* sp., **h)** on soil, **i)** on limestone, **j)** on bark, mixed forest of *Quercus*, *Juniperus* and *Acer*.

**6.- ARMENIA: ARARAT:** Vedi, Urtsadzor, Khosrov Forest State Reserve, abandoned village, 40°01'10.7"N, 44°54'46.3"E, 1760 m, 17-VI-2015, **a)** on *Malus* sp.

**Appendix B. Supplementary material for Chapter 4**

**Appendix B1.** Table 4.1. Voucher specimens and NCBI GenBank accession numbers of the ITS sequences used in the phylogenetic analyses.

Gasparyan, A. & Aptroot, A. (2016) *Verrucaria juglandis*, a new corticolous lichen species from Armenia. *Herzogia* 29: 103–107.

DOI: <http://dx.doi.org/10.13158/heia.29.1.2016.103>

## Appendix C. Supplementary material for Chapter 5

**Appendix C1.** Table 5.1. The list of species, collectors and collection numbers (if available), herbarium numbers and GenBank accession numbers of the ITS sequences used in this study.

Species	Collector, collection number, herbarium number	GenBank accession number
<i>R. aff. pollinaria</i>	07-29001	JF923612
<i>R. aff. pollinaria</i>	P. van den Boom LG 41227 R501	GU827324
<i>R. aff. pollinaria</i>	Normore3748	JQ003096
<i>R. europaea</i>	V. Otte s.n. [GLM-L 34284]	KY362397
<i>R. europaea</i>	A. Gasparyan T-14-22 [B 60 0201021]	KY362398
<i>R. europaea</i>	A. Gasparyan T-14-34g [B 60 0201030]	KY362399
<i>R. europaea</i>	A. Gasparyan T-14-35 [B 60 0201024]	KY362400
<i>R. europaea</i>	A. Gasparyan D-13-pl.12.01 [B 60 0201018]	KY362401
<i>R. europaea</i>	A. Gasparyan H-13-pl.03-06 [B 60 0201034]	KY362402
<i>R. europaea</i>	A. Gasparyan s.n. [B 60 0201028]	KY362403
<i>R. europaea</i>	A. Gasparyan s.n. [B 60 0201022]	KY362404
<i>R. europaea</i>	A. Gasparyan S-14-23 [B 60 0201031]	KY362405
<i>R. europaea</i>	A. Gasparyan s.n. [B 60 0201025]	KY362406
<i>R. europaea</i>	A. Paukov 2340 [UFU L-2520, dupl. in B]	KY362407
<i>R. europaea</i>	A. Paukov 2337 [UFU L-2517, dupl. in B]	KY362408
<i>R. europaea</i>	A. Paukov AGP20130704-69 [UFU L-1684, dupl. in B]	KY362409
<i>R. europaea</i>	TNS:VH:25409	AB362795
<i>R. europaea</i>	Tuerk 40707	EF432560
<i>R. europaea</i>	A. Gasparyan s.n. [B 60 0201019]	KY362410
<i>R. europaea</i>	A. Gasparyan s.n. [B 60 0201027]	KY362499
<i>R. europaea</i>	A. Gasparyan s.n. [B 60 0201033]	KY362500
<i>R. fastigiata</i>	-	AY462055
<i>R. fastigiata</i>	Hur H060127	EU034669
<i>R. fastigiata</i>	V. Otte 31160	KY362411
<i>R. farinacea</i>	Ekman s.n.	AM292707
<i>R. pollinaria</i>	TNS:AA:67802	AB362794
<i>R. pollinaria</i>	-	KX132955

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<i>R. pollinaria</i>	A. Aptroot 74529 [ABL]	KY362412
<i>R. pollinaria</i>	A. P. Yatsina MSK-L 12660 [B 60 0201032]	KY362413
<i>R. pollinaria</i>	A. Gasparyan S-14-21 [B 60 0201026]	KY362414
<i>R. pollinaria</i>	A. Gasparyan T-14-01 [B 60 0201020]	KY362415
<i>R. pollinaria</i>	A. Gasparyan s.n. [B 60 0201023]	KY362416
<i>R. pollinaria</i>	A. Paukov 2335 [UFU L-2515, dupl. in B]	KY362417
<i>R. pollinaria</i>	A. Gasparyan T-14-08 [B 60 0201029]	KY362418
<i>R. pollinaria</i>	A. Gasparyan s.n. [B 60 0201017]	KY362419
<i>R. labiosorediata</i>	Chris Lewis 341 [CANL 124119]	KY362420
<i>R. labiosorediata</i>	Chris Lewis 341 [CANL 124119]	KY362421
<i>R. labiosorediata</i>	E. A. Tripp 671 [NY 01117934]	KY362422
<i>R. labiosorediata</i>	J. C. Lendemer 12329 [NY 00977236]	KY362423
<i>R. labiosorediata</i>	J. C. Lendemer 14495 [NY 00974764]	KY362424
<i>R. labiosorediata</i>	J. C. Lendemer 18826 [NY 01118742]	KY362425
<i>R. labiosorediata</i>	J. C. Lendemer 26793 [NY 01219716]	KY362426
<i>R. labiosorediata</i>	D. C. Thornburg 837 [NY 01808147]	KY362427

**Apéndice C2.** Supplementary Table 5.1. Infraspecific names in *Ramalina* and their current status, as far as known (either published or assigned in this paper).

Resolved names:

- R. pollinaria* f. *cetrariooides* Bagl. = ***R. requienii* (De Not.) Jatta**  
*R. pollinaria* f. *chilena* Nyl. = ***R. chilena* (Nyl.) Kashiw.**  
*R. pollinaria* f. *elatior* Ach. = ***R. pollinaria* (Westr.) Ach. s.str.**  
*R. pollinaria* f. *levasiensis* Räsänen; *R. pollinaria* var. *levasiensis* (Räsänen) Motyka = ***R. balatonica* Gyeln.**  
*R. pollinaria* f. *sublacerella* Räsänen 1939; *R. pollinaria* var. *sublacerella* (Räsänen) Räsänen = ***R. farinacea* (L.) Ach.**  
*R. pollinaria* f. *ventricosa* Eitner = ***R. baltica* Lettau**  
*R. pollinaria* var. *anceps* (Nyl.) Trevis. = ***R. anceps* Nyl.**  
*R. pollinaria* var. *balatonica* (Gyeln.) Motyka = ***R. balatonica* Gyeln.**  
*R. pollinaria* var. *boleana* (A. Massal.) E. Szatala = ***R. pollinaria* s.str.**  
*R. pollinaria* var. *dilacerata* (Hoffm.) Mong. = ***R. dilacerata* (Hoffm.) Hoffm.**  
*R. pollinaria* var. *duriaei* De Not. = ***R. lacera* (With.) J. R. Laundon**  
*R. pollinaria* var. *evernioides* (Nyl.) H. Olivier = ***R. maciformis* (Delise) Bory**  
*R. pollinaria* var. *humilis* Ach.; *R. pollinaria* f. *humilis* (Ach.) Anders = ***R. humilis* (Ach.) Röhl**  
*R. pollinaria* var. *insularis* Vain. = ***R. exilis* Asah.**

*R. pollinaria* var. *phycooides* Mont. = ***Niebla crispatula* (Despr. ex Nyl.) Bowler & J.E. Marsh**

*R. pollinaria* var. *pulvinata* Anzi = ***R. breviuscula* (Nyl.) Nyl.**

*R. pollinaria* var. *sortavalensis* Räsänen = ***R. pollinaria* (Westr.) Ach. s.str.**

Unresolved names:

*R. pollinaria* f. *ampullacea* (Wallr.) Flot.

*R. pollinaria* f. *cariosa* Laurer

*R. pollinaria* f. *conglobata* Mereschk.

*R. pollinaria* f. *cucullata* Harm.

*R. pollinaria* f. *elegantella* Mereschk.

*R. pollinaria* f. *gracilis* H. Magn.

*R. pollinaria* f. *hemisphaerica* Tomin

*R. pollinaria* f. *minor* Arnold; *R. pollinaria* var. *minor* (Arnold) Motyka

*R. pollinaria* f. *monophylla* Coem.

*R. pollinaria* f. *multipartita* Hepp; *R. pollinaria* var. *multipartita* (Hepp) Motyka

*R. pollinaria* f. *nitidiuscula* Zahlbr.

*R. pollinaria* f. *prolifera* (Wallr.) Flot.

*R. pollinaria* f. *rupestris* Flörke

*R. pollinaria* f. *torulosa* Erichsen

*R. pollinaria* var. *globosa* Motyka

*R. pollinaria* var. *intermedians* H. Olivier; *R. pollinaria* f. *intermedians* (H. Olivier) E. Szatala

*R. pollinaria* var. *marginata* Motyka

*R. pollinaria* var. *multipartita* f. *compacta* Motyka

*R. pollinaria* var. *scobinosa* Motyka

**Apendix C3.** Supplementary Figure 5.1. Maximum-likelihood phylogenetic tree of the genus *Ramalina* inferred from ITS. Supported branches are thickened and support values are given.

You can view or download the file from here: [goo.gl/0J2wRx](http://goo.gl/0J2wRx)

**Appendix C4.** Supplementary Figure 5.2. Variable alignment positions extracted from the target group data set to show positions that are diagnostic between recognized clades.

- R. pollinaria* (Sweden Gasparyan 60 0201023)
- R. pollinaria* (Russia Paukov 2335)
- R. pollinaria* (Nederlands Aptroot 74529)
- R. pollinaria* (Belarus Yatsina 12660)
- R. pollinaria* (Armenia Gasparyan T 14 01)
- R. pollinaria* (Armenia Gasparyan T 14 08)
- R. pollinaria* (Sweden Gasparyan 60 0201017)
- R. pollinaria* (Netherlands AB362794)
- R. pollinaria* (Armenia Gasparyan S 14 21)
- R. pollinaria* (Sweden AM292707)
- R. pollinaria* (Switzerland KX132955)
- R. aff. pollinaria* (Canada JQ003096)
- R. labiosorediata* (Canada Lewis 341)
- R. labiosorediata* (Canada Lewis 341a)
- R. labiosorediata* (USA Lendemer 12329)
- R. labiosorediata* (USA Lendemer 671)
- R. labiosorediata* (USA Lendemer 18826)
- R. labiosorediata* (USA Lendemer 26793)
- R. labiosorediata* (USA Thornburg 837)
- R. labiosorediata* (Canada Lendemer 14495)
- R. aff. pollinaria* (China JF923612)
- R. europaea* (Russia Paukov 2337)
- R. europaea* (Armenia 60 0201033)
- R. europaea* (Armenia Gasparyan T 14 35)
- R. europaea* (Armenia Gasparyan 60 0201027)
- R. europaea* (Armenia Gasparyan S 14 23)
- R. europaea* (Russia Paukov 20130704 69)
- R. europaea* (Russia Paukov 2340)
- R. europaea* (Sweden Gasparyan 60 0201019)
- R. europaea* (Armenia Gasparyan T 14 22)
- R. europaea* (Armenia Gasparyan 60 0201022)
- R. europaea* (Armenia Gasparyan 60 0201025)
- R. europaea* (Finland AB362795)
- R. europaea* (Austria EF432560)
- R. europaea* (Armenia Gasparyan 60 0201034)
- R. europaea* (Russia Otte 34284)
- R. europaea* (Armenia Gasparyan 60 0201028)
- R. europaea* (Armenia Gasparyan T 14 34g)
- R. europaea* (Armenia Gasparyan 60 0201018)
- R. aff. pollinaria* (Switzerland GU827324)

