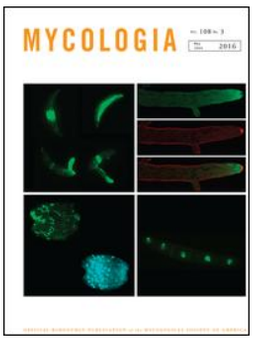

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Lichenicolous species of *Hainesia* belong to Phacidiales (Leotiomycetes) and are included in an extended concept of *Epithamnolia*

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ABSTRACT

The lichenicolous taxa currently included in the genus *Hainesia* were studied based on the nuclear rDNA (18S, 28S, and internal transcribed spacer [ITS]) genes. The authors found that lichenicolous taxa form a distinct lineage sister to *Epiglia gloeocapsae* (Phacidiales, Leotiomycetes), only distantly related to the type species of *Hainesia* (Chaetomellaceae, Helotiales). Owing to morphological similarities, the authors include the lichenicolous species into the previously monotypic genus *Epithamnolia*. A new species, *Epithamnolia rangiferinae*, is described, several names are reduced into synonymy, and a key to the species of *Epithamnolia* is provided. The incorporation of public environmental ITS sequences showed that the closest relatives of these lichenicolous taxa are various endophytic, endolichenic, and soil-inhabiting fungi.

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Asexual ascomycetes; environmental sequences; intraspecific variation; rRNA secondary structure

INTRODUCTION

Leotiomycetes, one of the most diverse classes of Ascomycota both in the sense of morphology and life styles, is characterized by inoperculate asci and mainly apothecioid ascomata (Jaklitsch et al. 2016). The class includes more than 5000 species (Kirk et al. 2011) currently divided among 10 orders (Jaklitsch et al. 2016). However, there is a lack of molecular data for a high proportion of taxa; therefore, the phylogenetic relationships within the class are far from being clear (Wang et al. 2006; Jaklitsch et al. 2016). Likewise, it is clear that in certain groups, the morphoanatomical characters do not reflect the origin and evolutionary relationships among taxa, owing to a high level of convergency (e.g., Han et al. 2014; Suija et al. 2015; Pärtel et al. 2017).

Recent molecular studies have shown that taxa of leotiomycetous fungi are related to many unsettled asexual taxa inherited from various environmental sources (e.g., Campbell et al. 2009; Tedersoo et al. 2009; Delgado et al. 2015). The genus *Hainesia* Ellis & Sacc. s. lat. comprises asexual ascomycetes occurring on plant material, soil (Seifert et al. 2011), and lichens (Diederich and van den Boom 2013). The genus is considered as belonging to Chaetomellaceae, a family with unclear affinities within Helotiales (Rossman et al. 2004; Wang et al. 2006; Jaklitsch et al. 2016). *Hainesia* is characterized by sporodochial or acervular cupulate-discoid conidiomata with hyaline enteroblastic phialidic acropleurogenous conidiophores and hyaline bacilli- to filiform conidia (Saccardo 1884; Seifert et al. 2011; Diederich and van den Boom 2013). The genus comprises approximately 30 species, many of them cosmopolitan, although species concepts are not always clear (Seifert et al. 2011).

The lectotype of *Hainesia* is *H. rhoina* (Sacc.) Ellis & Sacc., of which *H. lythri* (Desm.) Höhn. is an earlier synonym (Shear and Dodge 1921; Sutton 1980). Palm (1991) showed that *H. lythri* is a synanamorph of *Pilidium concavum* (Desm.) Höhn., sequences of which were included in a phylogenetic analysis by Rossman et al. (2004), and for which the correct name is now *Pilidium lythri* (Desm.) Rossman (Johnston et al. 2014). The generic type of *Pilidium* Kunze, *P. acerinum* (Alb. & Schwein.) Kunze, grouped with *P. concavum* in Rossman's phylogeny (Rossman et al. 2004); therefore, *Hainesia* s. str. has to be considered as a younger synonym of *Pilidium*. In that phylogeny, both *Pilidium* species were sister to *Chaetomella* Fuckel (Chaetomellaceae, Helotiales).

The first reported lichenicolous *Hainesia* species, *H. pertusariae*, growing on corticolous *Pertusaria* species,

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was described by Etayo and Diederich (1996). Seven additional lichenicolous species were recently added, viz., *Hainesia aeruginascens, H. lecanorae* (Brackel 2014), *H. brevicladoniae, H. longicladoniae* (Diederich and van den Boom 2013), *H. bryonorae* (Zhurbenko and Brackel 2013), *H. peltigerae* (Zhurbenko 2013), and *H. xanthoriae* (Brackel 2009). Although nonlichenicolous species of the genus have consistently aseptate conidia, conidia of most lichenicolous members are 1–5-septate (Zhurbenko 2013; Zhurbenko and Brackel 2013; Brackel 2014).

This study focuses on lichenicolous members of the genus *Hainesia*, aiming at (i) confirming their affiliation with Leotiomycetes; (ii) determining if they are congeneric with the type of *Hainesia*; (iii) clarifying whether they are phylogenetically allied to plant-pathogenic species of the genus; and (iv) identifying if these species are confined to single hosts or if they show a broader host spectrum.

MATERIALS AND METHODS

Taxon sampling and microscopy.—Altogether 15 specimens growing on five host genera (*Candelaria, Cladonia, Lecanora, Punctelia*, and *Xanthoria*) were used to create 40 new sequences of nuclear ribosomal DNA, including 15 internal transcribed spacer (ITS), 12 28S, and 13 18S sequences. Voucher information and GenBank accession numbers are given in TABLE 1.

The examined specimens are deposited in BR, G, LE, and TU and in the private collections of F. Berger, P. Diederich, P. van den Boom, and E. Zimmermann. Dry herbarium specimens were examined and measured under a binocular microscope Leica MZ 7.5 (magnification up to 50×; Wetzlar, Germany) and photographed using a binocular microscope Leica M165C, a Jenoptik ProgResC5 camera (Jena, Germany), and the free software CombineZM (available at https://combinezm.informer.com) for increasing the depth of field. Entire conidiomata were studied microscopically in water, 5% KOH, Melzer's reagent, Congo red, or Phloxine B, either with or without pressure on the coverslip. Microscopic photographs were prepared using a Leica DMLB microscope and a Leica EC3 camera, and Helicon Focus (Helicon Soft, Kharkiv, Ukraine) for increasing the depth of field, and those of *Epithamnolia karatyginii* using a Zeiss Axio Imager A1 microscope (Göttingen, Germany).

Conidial measurements are indicated as (minimum–)$\bar{X}-\sigma_X$–$\bar{X}+\sigma_X$(–maximum), followed by the number of measurements (N). A box-and-whisker plot illustrating the variation of conidial length of selected sequenced specimens of *Hainesia xanthoriae* was prepared using Microsoft Excel (Redmond, Washington, USA).

DNA extraction, amplification, and sequencing.—Genomic DNA was extracted from conidiomata (up to five per reaction) of freshly collected specimens. A single conidioma was removed from the lichen thallus, placed on a drop of the distilled water on a microscopical slide, and under the stereomicroscope remains of the lichen thallus were removed with a scalpel. The rest of the conidioma was placed into a 1.5-mL test tube. DNA was extracted using High Pure PCR Template Preparation Kit (Roche Applied Science, Penzberg, Germany) following the protocol provided by the manufacturer, except that in the final step only 75 µL of elution buffer was added.

Table 1. Voucher information and GenBank accession codes corresponding to sequences generated for this study.

Laboratory code	Collector and collection no.	Country	Herbarium	Host	18S	28S	ITS
HA85	van den Boom 50178	The Netherlands	van den Boom	*Punctelia subrudecta*	KY814521	KY814510	KY814528
HA86	van den Boom 49424	The Netherlands	van den Boom	*Punctelia subrudecta*	KY814520	KY814509	KY814527
HA87	van den Boom 48243	The Netherlands	van den Boom	*Punctelia subrudecta*	KY814523	KY814511	KY814530
HA88	van den Boom 48257	The Netherlands	van den Boom	*Punctelia subrudecta*	KY814522	—	KY814529
HA89	van den Boom 49369	The Netherlands	van den Boom	*Punctelia subrudecta*	—	KY814507	KY814525
HA90	van den Boom 50227	The Netherlands	van den Boom	*Candelaria concolor*	KY814524	KY814513	KY814532
HA92	Diederich 17562	Iceland	Diederich	*Lecanora symmicta*	KY814519	KY814508	KY814526
HA118	Berger 28669	Austria	Berger	*Cladonia squamosa*	—	KY814512	KY814531
HA127	Berger 28798	Austria	Berger	*Xanthoria parietina*	KY828443	KY814514	KY814533
HA131	van den Boom 52578	The Netherlands	van den Boom	*Xanthoria parietina*	KY828442	—	KY814534
HA132	van den Boom 52584	The Netherlands	van den Boom	*Punctelia subrudecta*	KY828441	KY814515	KY814535
HA156	Diederich 18191	Luxembourg	Diederich	*Xanthoria parietina*	KY828440	—	KY814536
HA172	Zimmermann 1257	Switzerland	G	*Cladonia rangiferina*	KY828439	KY814516	KY814537
HA182	Gardiennet 16031	France	TU 82109	*Punctelia subrudecta*	KY828445	KY814517	KY814538
HA183	Gardiennet	France	TU 82108	*Xanthoria parietina*	KY828444	KY814518	KY814539

Note. —, sequence not generated.

Three gene regions were selected for amplification: the internal transcribed spacer (ITS), 18S (SSU), and 28S ribosomal (LSU) RNA genes. ITS was amplified and sequenced using primers ITS0F and LA-W (Tedersoo et al. 2008), and ITS4 and ITS5 (White et al. 1990); 28S with primers LR5 and LR7 (Vilgalys and Hester 1990), LROR (Rehner and Samuels 1994), and CTB6 (Garbelotto et al. 1997); and 18S with primers PNS1 (Hibbett 1996) and NS41 (White et al. 1990) or with SSU1/SSU31R and SSU3/SSU42R (Pärtel et al. 2017). The polymerase chain reaction (PCR) reaction mix (25 µL) consisted of 5 µL 5× HOT FIREPol Blend Master Mix (Solis BioDyne, Tartu, Estonia), 0.5 µL of both primers (both in concentration of 20 µM), 0.8–3 µL of target DNA, and distilled water up to the total volume. For the purification of PCR products, 1 µL of FastAP and 0.5 µL of exonuclease I (Thermo Scientific, Waltham, Massachusetts, USA) were added per 20 µL of the product and the tubes were incubated at 37 C for 45 min; the enzymes were deactivated by heating at 85 C for 15 min. DNA sequencing of both complementary strands was performed by Macrogen Inc. (Amsterdam, the Netherlands). The sequence chromatograms were assembled, trimmed, and manually edited with Sequencher 4.10.1. (GeneCodes Corp., Ann Arbor, Michigan, USA). The resulting sequence contigs were used for analysis.

Phylogenetic analyses.—At first, we used the data set of 28S + 18S + 5.8S (134 taxa, 2024 bp) compiled originally by Wang et al. (2006) and supplemented by additional sequences derived from the National Center for Biotechnology Information (NCBI) database (see SUPPLEMENTARY TABLE 1) and created by us to detect (i) the position of lichenicolous *Hainesia* specimens within Leotiomycetes and (ii) to confirm or disprove congenerity of lichenicolous *Hainesia* with *Pilidium acerinum* and *P. lythri*. ITSx (Bengtsson-Palme et al. 2013) was used to extract variable ITS1 and ITS2 and conserved 5.8S subregions from full rDNA ITS sequences. The 28S + 18S + 5.8S set was aligned with MUSCLE (Edgar 2004) using default settings and followed by manual adjustment with SeaView 4.6 (Gouy et al. 2010). The best-fit substitution model according to Akaike information criterion (AIC), GTR+I+G, was calculated over 40 possible combinations using jModeltest 2.1.6. (Darriba et al. 2012). Maximum likelihood (ML) analyses using the best-fit model were implemented in RAxML 8.1.11 (Stamatakis et al. 2008) at the CIPRES Science Gateway (Miller et al. 2010). The best-scoring tree was selected based on the best log-likelihood score. The bootstrap support (BS) was calculated over 1000 pseudoreplicates. Bayesian phylogenetic analysis by Markov chain Monte Carlo (MCMC) sampling was performed with MrBayes 3.2 (Ronquist et al. 2012) using the same substitution model. Two parallel runs with four chain runs were performed over 5 000 000 generations starting from the random tree until the threshold 0.01 of the average standard deviation of split frequencies. Sampling was done after 200 steps; the first 25% of saved data were discarded as "burn-in", and the 50% majority-rule consensus tree and posterior probabilities (PPs) were calculated from the rest. The phylogenetic trees were visualized and edited using FigTree 1.4.2 (Rambaut 2014). Adobe Illustrator CS3 (Adobe Systems, San Jose, California, USA) was used for artwork. Clades with BS values ≥75 and PPs ≥0.95 were regarded as significantly supported.

We additionally compiled an rDNA ITS data set (45 sequences, 748 bp) including the most similar sequences (≥97%; GenBank accession numbers in FIG. 4) according to a BLAST search from the NCBI database. The data set was aligned with MUSCLE (Edgar 2004), then adjusted with SeaView 4.6, and analyzed with PhyML (Guindon et al. 2010) using GTR substitution model and bootstrapping over 500 replicates in SeaView 4.6 (Gouy et al. 2010). The ITS2 transcript folding pattern was performed with LocARNA (Will et al. 2012) at http://rna.informatik.uni-freiburg.de. Both alignment files used for analyses are available in TreeBASE repository under the reference number TB21327 (http://purl.org/phylo/treebase/phylows/study/TB2:S21327).

RESULTS

Phylogenetic positions within Leotiomycetes.—Maximum likelihood (ML) and Bayesian trees had no topological conflicts in the supported clades (FIG. 1), still throughout the phylogenetic trees the larger clades remained unsupported both by BS and PP values; also Erysiphales and Cyttariales were nested within Helotiales, indicating a possible paraphyly of the order (FIG. 1; see also Pärtel et al. 2017). Both analyses of the combined three-gene phylogeny (28S + 18S + 5.8S) suggested that lichenicolous *Hainesia* species belong to Leotiomycetes but represent a phylogenetically distinct lineage not related to Chaetomellaceae, including *Pilidium concavum*, a synanamorph of the type species of *Hainesia, H. lythri* (FIG. 1). The lichenicolous *Hainesia* species formed a highly supported (PP = 1, BS = 100) clade sister to the bryophilous *Epiglia gloeocapsae* (PP = 1, BS = 84),

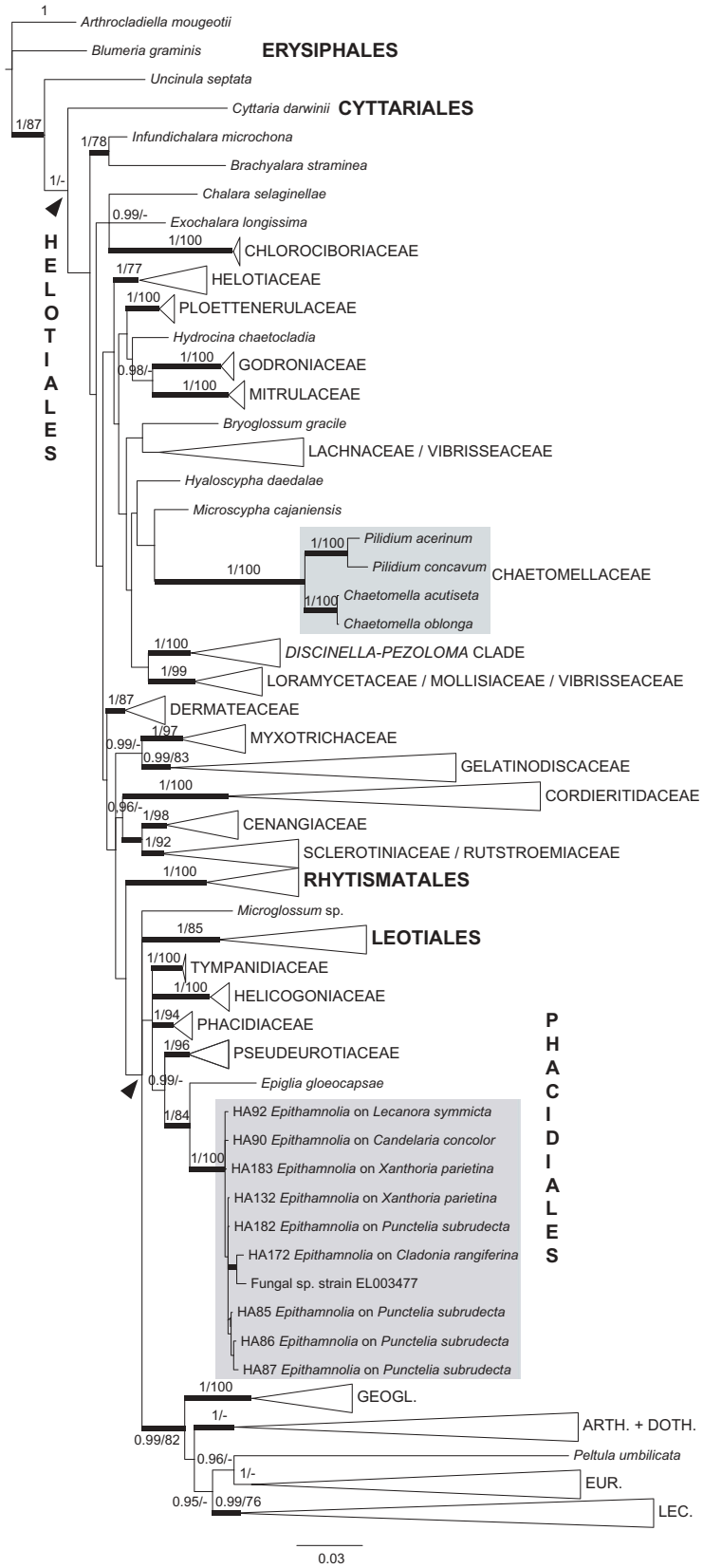

Figure 1. Three locus (28S + 18S + 5.8S)–based 50% majority-rule consensus tree based on Bayesian approach, representing relationships between Ascomycota and showing the position of lichenicolous *Epithamnolia* specimens within Leotiomycetes. The branches with posterior probabilities (PPs) ≥0.95 and bootstrap values (BS) ≥75 are marked with a thicker line; the clades corresponding to higher taxonomic units according to Jaklitsch et al. (2016) are collapsed (see SUPPLEMENTARY TABLE 1). Lichenicolous specimens and species from Chaetomellaceae are highlighted with gray box. The triangles at the nodes indicate separation of Helotiales and Phacidiales. Abbreviations of taxon names in outgroup: ARTH. = Arthoniomycetes; DOTH. = Dothideomycetes; EUR. = Eurotiomycetes; GEOGL. = Geoglossomycetes; LEC. = Lecanoromycetes.

and both were sister to Pseudeurotiaceae (PP = 0.99, BS = 67). The whole group was nested within the recently resurrected order Phacidiales (Crous et al. 2014) sensu Jaklitsch et al. (2016), which remained, however, unresolved. NCBI sequences marked as fungal sp. strain EL003477 and representing an endolichenic fungus from Antarctica were nested within the lichenicolous *Hainesia* clade, indicating that the distribution of this genus is wider as previously known. Another lichenicolous species, *Geltingia associata*, together with the fungicolous *Eleutheromyces subulatus*, had a basal, but unresolved position within the Phacidiales (FIG. 1).

Genetic variation of rDNA ITS sequences versus variation of conidial length.—The analysis of rDNA ITS nucleotide sequences clearly separated specimens growing on *Cladonia squamosa* (*Hainesia brevicladoniae*) and *C. rangiferina* (new species, described below). Both ITS sequences have an identical 187-bp intron at the beginning of ITS1 not detected in the rest of lichenicolous *Hainesia* sequences. The rest of 14 ITS sequences formed a supported (BS = 0.94) clade in which a single-nucleotide polymorphism (SNP) in position 48 of the conserved region of ITS2 was observed. The SNP correlated with the host choice separating specimens on *Punctelia subrudecta* from those on *Xanthoria parietina, Candelaria concolor*, and *Lecanora* spp. The secondary structure of the ITS2 transcript marker consisted of a three-looped structure; the difference between the two groups was in loop II, which was two nucleotide pairs longer in specimens on *Punctelia subrudecta* (FIG. 2A; 11 pairs) than in other specimens (FIG. 2B; 9 pairs).

Despite of a lack of variability of ITS sequences across specimens, the conidial length of a selection of sequenced specimens varies to a significant extent (FIG. 3). The longest mature conidia, 66.3–84.0 µm (average ± standard deviation) (N = 30), were from HA182 (Gardiennet 16031; host *Punctelia subrudecta*), whereas the shortest from the same host, 48.5–58.2 µm (N = 20), were from HA88 (van den Boom 48257). Within a single population (HA92, Diederich 17562, host *Lecanora symmicta*; Diederich 17563, host *L. saligna*), the conidial length varied from 23.4–32.4 µm (N = 14) in a young conidioma to 49.3–57.6 µm (N = 20) in a mature conidioma.

Extended rDNA ITS data set.—The extended data set of ITS sequences incorporated sequences obtained from different types of environmental samples (FIG. 4), including endophytic fungi on *Chorisodontium aciphyllum* (Dicranaceae, Bryophyta) and *Rhododendron ferrugineum* (Ericaceae, Angiospermae), fungus from soil horizon (KF617768), and house dust (AM901846). Moreover, the sequence cluster, which comprised endophytic fungi from *C. aciphyllum*, formed a supported clade (BS = 0.86) with two lichenicolous species on *Cladonia*, whereas sequences from the rest of lichenicolous taxa were genetically similar to sequences from various environmental sources. Sequence KF712231 annotated as *Tephromela*

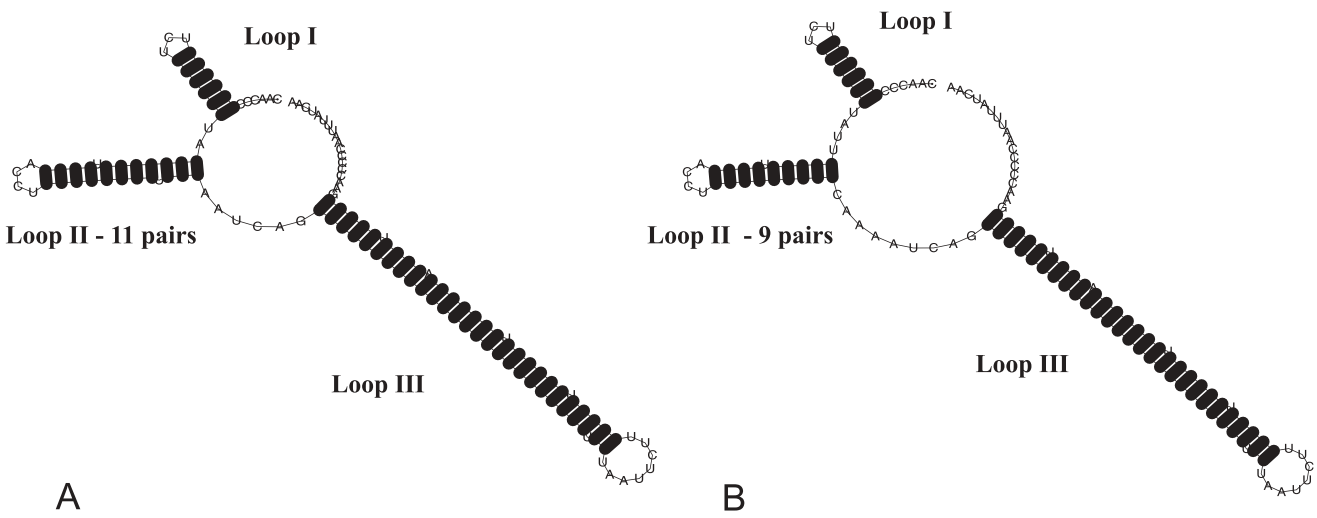

Figure 2. The secondary structures of ITS2 transcripts of *Epithamnolia* specimens. A. On *Punctelia*. B. On *Xanthoria, Lecanora*, and *Candelaria*. The loops are denoted with Roman numerals; the number in brackets at the loop II indicates the number of nucleotide pairs per loop.

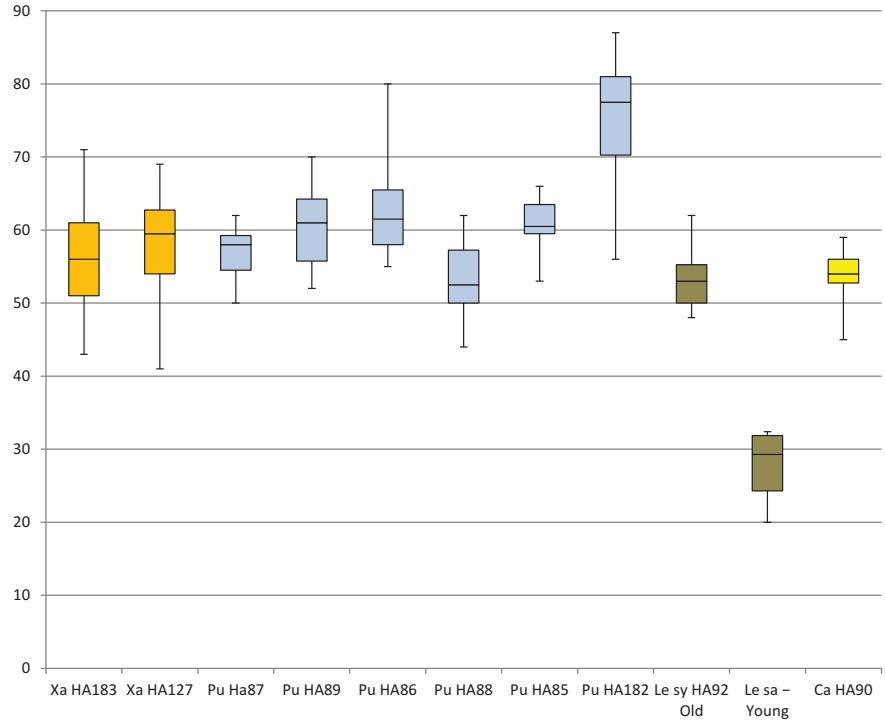

Figure 3. Box-and-whisker plot of conidial length of selected specimens of *Epithamnolia xanthoriae*. Each specimen is abbreviated by the host name, and those sequenced by a laboratory code (see TABLE 1). Abbreviations of hosts: Xa = *Xanthoria parietina*; Pu = *Punctelia subrudecta*; Le sa = *Lecanora saligna*; Le sy = *Lecanora symmicta*; Ca = *Candelaria concolor*; 'young' and 'old' denote conidia from young and mature conidiomata on *Lecanora*, respectively.

cf. *atra* in the NCBI database is obviously from a contaminant.

DISCUSSION

We have shown that lichenicolous *Hainesia* species are phylogenetically not related to *Pilidium concavum*, a synanamorph of the type species of *Hainesia*, and therefore cannot stay in *Hainesia*. The lichenicolous species differ from *Hainesia* s. str. by often much longer, in most species septate, conidia, and by the lichenicolous habitat. The only other lichenicolous genus producing similar cupulate conidiomata and hyaline, cylindrical, septate conidia is *Epithamnolia* Zhurb. (Zhurbenko 2012), which was supposed to differ by the missing conidiophores. Owing to morphological similarities, we include the lichenicolous *Hainesia* species into this previously monotypic genus (see Taxonomy).

The lichenicolous specimens obviously belong to the recently introduced order Phacidiales (Crous et al. 2014) that originally incorporated a single family, Phacidiaceae (Crous et al. 2014). Jaklitsch et al. (2016) extended the concept of the order based on yet unpublished results and showed genetic similarities between Phacidiaceae and two other families, Tympanidaceae and Helicogoniaceae. Our current selection of taxa and gene sequences is certainly inadequate and does not permit a further extension of the order's concept, as the clade remains unsupported based on one of two analyses performed. However, the asexual stages of Phacidiales are mostly characterized by a phialidic, acropleurogenous type of conidiogenesis and hyaline, mainly aseptate to 1-septate conidia (Crous et al. 2014; Jaklitsch et al. 2016), thus endorsing the placement of lichenicolous species into this order.

The incorporation of environmental and specimen-derived sequences suggested that lichenicolous species are phylogenetically close to some endophytic and endolichenic fungi (FIG. 4). Previous phylogenetic analyses have shown the relationship between these latter groups (e.g., Arnold et al. 2009). However, the link between lichenicolous and endolichenic-endophytic fungi has never been shown. Still, it is not always clear whether the environmental sequences are derived from true endophytes (or endolichenic fungi) or represent fungi for which fruit body–derived sequences are not yet available. Under certain environmental conditions, especially on senesced and decomposing leaves, some endophytes are able to

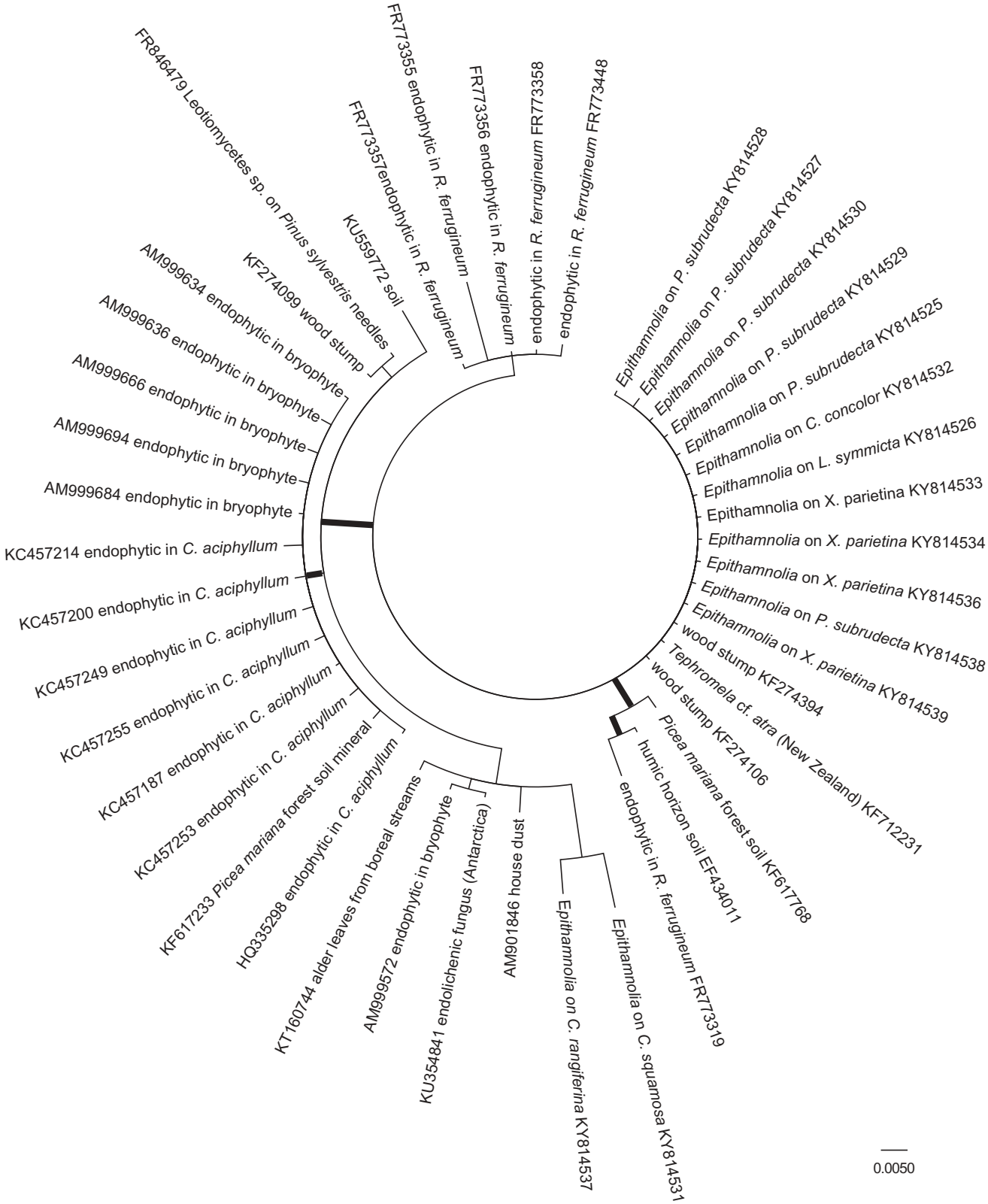

Figure 4. Maximum likelihood phylogeny including sequences generated for this study and NCBI-derived rDNA ITS sequences with sequence similarity ≥0.97. The branches marked in thicker line are supported by bootstrap values (BS ≥70). Abbreviations of taxon names: *C. aciphyllum* = *Chorisodontium aciphyllum* (Dicranaceae, Bryophyta); *R. ferrugineum* = *Rhododendron ferrugineum* (Ericaceae, Plantae); *C. concolor* = *Candelaria concolor; C. rangiferina* = *Cladonia rangiferina; C. squamosa* = *Cladonia squamosa; L. symmicta* = *Lecanora symmicta; P. subrudecta* = *Punctelia subrudecta; X. parietina* = *Xanthoria parietina* (all lichen-forming fungi).

form reproductive structures (e.g., U'Ren et al. 2010). Therefore, it is highly possible that the enlarged concept of the genus *Epithamnolia* introduced here (see Taxonomy) should be extended further to include fungi from other herbal substrates when their fruit body–derived sequences will be available. Already now, the inclusion of public ITS sequences allowed us to identify a few similar sequences (KF712231, KU354841) derived from lichen thalli, suggesting an extension of the known host choice (*Tephromela* cf. *atra*) and geographical distribution (New Zealand, Antarctica) of the genus.

The variability of rDNA ITS sequences, and especially the fast-evolving ITS2 region, provides valuable information about species delimitation (e.g., Müller et al. 2007), especially for closely related species. The ITS2 transcript folding pattern therefore may give an additional hint on species identification, as mutations, especially in the conserved motif of loop II, are indicative of changes in rRNA processing and protein binding (Good et al. 1997; Coleman 2009; Caisová et al. 2011). We interpret here the single-nucleotide polymorphism (SNP) in the conserved region of ITS2 (FIG. 2), separating specimens on *Punctelia subrudecta* from those on other epiphytic hosts, as an indication of an ongoing speciation process (cryptic speciation) based on the isolation by the environment (Wang and Bradburd 2014) and manifested here by host switching. Species delimitation of parasitic fungi using morphoanatomical characters is often challenging due to the scarcity of easily observable characters (Lawrey and Diederich 2003). This is obvious also here (see details in Taxonomy) as, for example, the intraspecific variability of one of the commonly used phenotypic traits, the conidial length (e.g., Zhurbenko and Brackel 2013; Brackel 2014), is high (FIG. 3). Therefore, we cannot delimit species in this lichenicolous genus by using a single morphological trait and host identity only.

TAXONOMY

Epithamnolia Zhurb., Lichenologist 44:158. 2012. Type: *E. karatyginii* Zhurb.

Notes: The genus *Epithamnolia* has been described for a lichenicolous fungus strongly resembling lichenicolous species of *Hainesia*, but distinguished by the missing conidiophores. In contrast, conidiophores in lichenicolous *Hainesia* species usually consist of a few short or more frequently elongate cells, from which conidia are produced acropleurogenously, and these conidiophores are often branched. As lichenicolous fungi included in *Hainesia* are phylogenetically not related to *Hainesia* (a younger synonym of *Pilidium*), two options exist: either a new genus is described for them or they are included in *Epithamnolia*, from which no sequences exist, despite the presence of distinct conidiophores. In our opinion, it should be avoided to describe new genera typified on asexual fungi when no molecular data are available, and instead such fungi should be included in existing genera that share most morphological characters. We reexamined all known specimens of the generic type, *Epithamnolia karatyginii*, and we were able to demonstrate the presence of some branched conidiophores consisting of more than one cell (FIG. 5), thus hardly differing from lichenicolous *Hainesia* species. Consequently, we include the former lichenicolous *Hainesia* species within *Epithamnolia*.

Several new lichenicolous *Hainesia* species have recently been described, differing from each other only by the conidial length and conidiomatal diameter. Examination of many specimens allowed us to conclude that the diameter of conidiomata is extremely variable within each species, and that specimens with smaller conidiomata may well be conspecific with specimens with mainly larger conidiomata. Similarly, conidial measurements obtained from many specimens suggest that specimens with mainly shorter conidia may be conspecific with other specimens with mainly longer conidia.

As an example, Brackel (2014) described the new *Hainesia aeruginascens* (on *Platismatia glauca*), differing from *H. xanthoriae* (on *Xanthoria parietina*) by distinctly longer conidia (no overlap in a statistical analysis, based on two specimens from each species), and the new *H. lecanorae* (on *Lecanora* s. lat.), with shorter conidia (again no overlap, based on two specimens from each). We have examined the conidial dimensions from two other specimens on *Lecanora* collected in the same locality (Diederich 17562 and 17563): in one conidioma, most conidia were aseptate and smaller than those of *H. lecanorae*, whereas in another conidioma most conidia were 5-septate and as long as those described from *H. xanthoriae*, i.e., much longer than those described from *H. lecanorae*. Phylogenetically, this specimen (Diederich 17562) groups with *H. xanthoriae* and almost surely belongs to that species. We conclude that conidial length strongly varies within some species, and even within a single specimen, and that new species should only be recognized when additional morphological characters separate them from known species, or when molecular data support the recognition of a new species.

Similarly, the new *Hainesia bryonorae* (on *Bryonora castanea*, conidia 18.1–24.5 µm long, conidiomata 30–100 µm diam) was described for material differing

from *H. pertusariae* (on *Pertusaria*, conidia 14–22 µm, conidiomata 80–150 µm) by slightly longer conidia and distinctly smaller conidiomata. Further specimens on *Cladonia* with conidia of an intermediate size (17.4–22.4 µm long) and relatively large conidiomata (100–150 µm) were assigned to *H.* cf. *bryonorae* (Zhurbenko and Pino-Bodas 2017), and this supports our assumption about the great variability of conidial length and conidiomatal diameter in these species. Another species, *Hainesia peltigerae*, described from *Peltigera rufescens*, has shorter conidia (10.6–14.6 µm long) and conidiomata 30–90 µm diam. A reexamination of the type of *H. pertusariae* revealed the presence of conidia that are smaller than in the original description and similar in size to those of *H. peltigerae* (down to 9.5 µm). Consequently, we prefer including all these specimens in an enlarged concept of *Hainesia pertusariae*.

Epithamnolia karatyginii Zhurb., Lichenologist 44:158. 2012. FIG. 5

Typification: CANADA. BRITISH COLUMBIA: Wells Gray Provincial Park, Raft Mountain, 51°44′N, 119°50′W, 2100 m, alpine tundra, on *Thamnolia vermicularis* var. *subuliformis*, 3 Aug 2002, *Zhurbenko 02343* (holotype LE 260498!).

Conidiomata when young pycnidioid, blackish, shiny, later opening to become broadly cupuliform, exposing the pale brown interior, subimmersed to finally superficial, arising singly, dispersed, 110–220 µm. Exciple in cross-section dark to medium brown, basally of 3–5 cell layers, in surface view resembling a *textura angularis/globulosa* (cells 3.5–10.0 µm across) or *textura porrecta*, K–. Conidiophores reduced to the conidiogenous cell, or composed of a few cells, occasionally branched. Conidiogenous cells 6.5–9 × 2–3 µm. Conidia cylindrical, slightly attenuated towards the apices, apically rounded to slightly truncate, (0–)1(–2?)-septate, (14–)18.5–27.0(–32) × (1.0–)1.5–2.0(–2.5) µm (N = 122; Zhurbenko 2012).

Distribution and hosts: Known from Canada (British Columbia) and Russia (Severnaya Zemlya, Northern Ural, Kola Peninsula, Taimyr Peninsula, and Yakutiya), always on the more or less damaged parts of the thallus of *Thamnolia vermicularis* (Zhurbenko 2012).

Notes: Conidia are reminiscent of those of *Epithamnolia pertusariae*, from which *E. karatyginii* differs by the missing or reduced conidiophores and the slightly wider conidia (1.5–2 vs. 1–1.5 µm). The study of additional specimens and/or molecular data should determine if both species are actually distinct.

Additional specimens examined: RUSSIA. SEVERNAYA ZEMLYA: Bol'shevik Island, Shokal'skogo Strait, 79°16′N, 101°40′E, 20 m, on *Thamnolia vermicularis* var. *subuliformis*, 1996, *Zhurbenko 96916* (LE 232933); NORTHERN URAL: headwaters of Pechora River, Yanypupuner Range, 62° 05′N, 59°06′E, 800 m, on *T. vermicularis* var. *subuliformis*, 1997, *Zhurbenko 97395a* (LE 260538a); YAKUTIYA: Laptev Sea coast, Tiksi, 71°40′N, 128°40′ E, 50 m, on *T. vermicularis* var. *vermicularis*, 1998, *Zhurbenko 98407b* (LE 260444b).

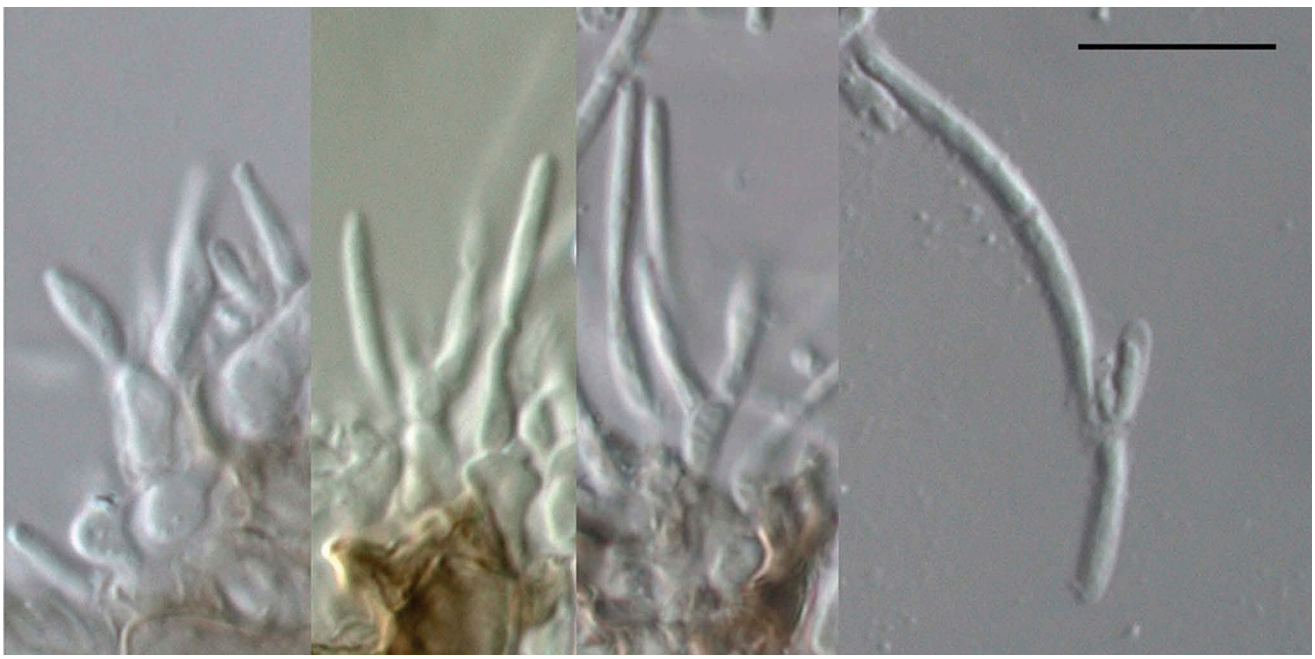

Figure 5. *Epithamnolia karatyginii* (LE 232933). Conidiophores. Bar = 10 µm

Epithamnolia brevicladoniae (Diederich & van den Boom) Diederich & Suija, comb. nov. FIG. 7A–D
MycoBank MB822789

≡ *Hainesia brevicladoniae* Diederich & van den Boom, Bull Soc Nat Luxemb 114:60. 2013 (basionym).

Typification: BELGIUM. W of Houffalize, 2 km SW of Nadrin, Le Hérou, forest along Ourthe, on *Cladonia* sp., 10 Sep 2010, *Diederich 17123* (holotype BR!; isotype hb Diederich!).

Conidiomata 40–220 µm. Conidiophores of 1–3 elongate cells. Conidiogenous cells 6.5–9 × 2–3 µm. Conidia bacilliform, basally truncate, apically broadly rounded to almost truncate, not attenuated towards the ends, (0–)1-septate (poorly visible), 12.5–17 × 1–1.5 µm (from Diederich and van den Boom 2013; Zhurbenko and Kobzeva 2014; Zhurbenko and Pino-Bodas 2017).

Distribution and hosts: Known from Austria, Belgium, France, Germany, the Netherlands, and Russia (NW Caucasus, Primorye Territory, Sakhalin Region), on the thallus of *Cladonia* species, including *C. coniocraea, C. gracilis, C. pocillum, C. polydactyla*, and *C. squamosa* (Diederich and van den Boom 2013; Brackel 2014; Zhurbenko and Kobzeva 2014; Zhurbenko and Pino-Bodas 2017).

Notes: Conidia were originally described as aseptate, (13.5–)15.3–17.5(–18.0) × (1.0–)1.0–1.1(–1.2) µm; a reexamination of the type material revealed the presence of a poorly visible septum in many conidia. Zhurbenko and Kobzeva (2014) noted that conidia in their material are aseptate or sometimes with a hardly visible septum, (9.6–)13.3–16.9(–18.6) × (1.1–)1.3–1.5(–1.6) µm. Zhurbenko and Pino-Bodas (2017) reported conidia as (0–)1(–3)-septate, (10.4–)12.4–16.0(–20.7) × (1.0–)1.2–1.6(–1.7) µm. In the sequenced specimen Berger 28669, conidia are (0–)1-septate, (13.2–) 14.0–15.8(–16.6) × (1.1–)1.2–1.4(–1.5) µm.

Conidia are bacilliform, with more or less the same thickness over the entire length, i.e., not broader in the center, not attenuated towards the apices, and broadly rounded to almost truncate at both apices.

Additional specimen examined: AUSTRIA. OBERÖSTERREICH: Donautal, Schlögener Schlinge, Steiner Fels-Hangwald bei km 2184.2, 48°26′25″N, 13°51′30″E, 320 m, on *Cladonia squamosa*, Nov 2014, *Berger 28669*.

Epithamnolia longicladoniae (Diederich & van den Boom) Diederich & Suija, comb. nov. FIG. 8F–G
MycoBank MB822790

≡ *Hainesia longicladoniae* Diederich & van den Boom, Bull Soc Nat Luxemb 114:62. 2013 (basionym).

Typification: LUXEMBOURG. À l'ouest de Steinfort, anciennes carrières, dans la réserve naturelle (West of Steinfort, old quarries, in nature reserve), 49.66753°N, 5.9053°E, on *Cladonia furcata* thallus, 15 Sep 2012, *Diederich 17486* (holotype BR!; isotype hb Diederich!).

Conidiomata 60–120 µm. Conidiophores of 0–3 elongate cells. Conidiogenous cells 7–10.5 × 1.7–2.3 µm. Conidia rod-shaped to vermiform, straight or irregularly bent, with the same width over the entire length, basally indistinctly truncate, apically rounded, not attenuated towards the ends, 0–6-septate, 35–71 × 1–2 µm (from Diederich and van den Boom 2013; Zhurbenko and Pino-Bodas 2017).

Distribution and hosts: Known from Italy, Luxembourg, the Netherlands, Russia (Krasnoyarsk Territory), and the USA (Alaska), on the thallus and apothecia of *Cladonia* species, including *C. coccifera, C. furcata, C. macilenta* subsp. *bacillaris, C. pyxidata*, and *C. rangiformis* (Diederich and van den Boom 2013; Brackel 2015; Zhurbenko and Pino-Bodas 2017).

Notes: Conidia were originally described as aseptate, (28.0–)40.1–63.5(–69.0) × (1.0–)1.1–1.4(–1.6) µm; a reexamination of specimen van den Boom 25449 from the Netherlands proved that they occasionally present an indistinct median septum (FIG. 7F). Conidia observed by Brackel (2015) were ca. 42–55 × 1.5 µm. Zhurbenko and Pino-Bodas (2017) reported conidia as 3–6-septate, ca. 35–50 × 1.5–2 µm in specimen LE 308594b, and (52.5–)58.0–71.0(–74.5) × (1.5–)1.6–1.8(–2.0) µm in LE 308828.

Conidia are rod-shaped to vermiform, with more or less the same thickness over the entire length, i.e., not distinctly broader in the center and not attenuated towards both apices, and this is the best character to distinguish the species from *Epithamnolia xanthoriae*.

Epithamnolia pertusariae (Etayo & Diederich) Diederich & Suija, comb. nov. FIG. 7E–F
MycoBank MB822792

≡ *Hainesia pertusariae* Etayo & Diederich, Mycotaxon 60:417. 1996 (basionym).

Typification: SPAIN. NAVARRA: Urroz de Santesteban, pantano de Leurtza, 900 m, on thallus of *Pertusaria* sp., 13 Feb 1994, *Etayo 12699* (holotype MA-Lich, isotypes hb Diederich!, hb Etayo).

= *Hainesia bryonorae* Zhurb., in Zhurbenko and Brackel, Herzogia 26:336. 2013.

NORWAY. SVALBARD: Spitsbergen, Nordenskiöld Land, W coast of Grønfjorden between Aldegondabreen glacier and the Brydebekken river mouth, 78°00′N, 14°12′E, 10 m, on apothecia of *Bryonora castanea*, 15 Jul 2003, *Zhurbenko* (holotype LE 261469!).

= *Hainesia peltigerae* Zhurb. & Davydov, in Zhurbenko, Graphis Scripta 25:41. 2013.

RUSSIA. ALTAI: Ulagan Region, N part of Kurai Range, 4 km W of Balyktukel Lake, 50°32′N, 87°39′E, 2200 m, on thallus of *Peltigera rufescens*, 14 Aug 1997, *Davydov 2204* (holotype LE 260889!).

Conidiomata 30–150 µm. Conidiophores septate, simple or branched, of 1–3 elongate filiform cells, each 5–11 × 1.3–2.8 µm. Conidiogenous cells 6–9.5 × 1.6–2 µm. Conidia bacilliform to filiform, straight or slightly bent, basally truncate, apically ± rounded, attenuated towards both ends, 0–1(–3)-septate, 10–24.5 × 1–1.5 µm (from Etayo and Diederich 1996; Zhurbenko 2013; Zhurbenko and Brackel 2013; Zhurbenko and Pino-Bodas 2017).

Distribution and hosts: Considered in the broad sense, this species is known from Canada (Ellef Ringnes Island), Norway, Portugal, Russia (Altai, Taimyr Peninsula), Spain (Navarra), Norway (Svalbard), and Switzerland, on the thallus of *Cladonia* species, including *C. macroceras* and *C. rangiferina, Peltigera rufescens*, and an unidentified corticolous *Pertusaria*, and on apothecia of *Bryonora castanea* (Etayo and Diederich 1996; Paz-Bermúdez et al. 2009; Zhurbenko and Brackel 2013; Zhurbenko and Pino-Bodas 2017).

Notes: Conidia of *Epithamnolia pertusariae* were originally described as 0(–1)-septate, 14–22 × 1–1.5 µm; a reexamination of the type specimen resulted in 0–1(–2)-septate conidia, (9.5–)12.6–16.5(–18) × (1.0–)1.1–1.4(–1.6) µm (N = 37), and conidiophores with cells of (4–)4.8–7.0(–7.7) × (1.2–)1.3–1.5(–1.6) µm (N = 20); conidiomata in the type specimen are 80–150 µm diam (Etayo and Diederich 1996). Conidia of *Hainesia bryonorae* were originally described as (0–)1-septate, (12.0–)18.1–24.5(–29.1) × (1.1–)1.3–1.5(–1.6) µm; conidiomata are 30–100 µm diam. Conidia of *Hainesia* cf. *bryonorae* on *Cladonia* from Norway and Russia are (0–)1-septate, (14.4–)17.4–22.4(–28.5) × (1.0–)1.1–1.3(–1.7) µm, and conidiomata (60–)100–150(–190) µm diam (Zhurbenko and Pino-Bodas 2017). Conidia of a Swiss specimen on *Cladonia rangiferina* (Zimmermann 810) are (0–)1-septate, (14.0–)15.0–17.7(–18.5) × (1.0–)1.0–1.2(–1.3) µm, and conidiomata 100–200 µm diam. These data suggest that both conidial length and conidiomatal diameter are variable within this material and, without further evidence, do not allow distinguishing several species.

Hainesia peltigerae strongly resembles *Epithamnolia pertusariae*. Conidia were described as (0–)1-septate, (8.3–)10.6–14.6(–16.5) × (1.0–)1.1–1.5(–1.7) µm, thus shorter than those observed in the type specimen of *E. pertusariae* by Etayo and Diederich (1996). However, during reexamination of the type of *E. pertusariae*, much smaller conidia were observed, down to 9.5 µm in length, and consequently, without further data, both species can hardly be distinguished.

Epithamnolia karatyginii also resembles *E. pertusariae* but differs by the missing or reduced conidiophores (often, but not always, reduced to the conidiogenous cell). Conidia were described as (0–)1(–2?)-septate, (14–)18.5–27.0(–32) × (1.0–)1.5–2.0(–2.5) µm (Zhurbenko 2012), slightly broader than those of *E. pertusariae*. Both names may be synonyms, but we provisionally keep them as distinct, awaiting the discovery of fresh material of *E. karatyginii* allowing molecular analyses.

Additional specimen examined: SWITZERLAND. BERN: Meiringen, Rosenlaui, Alpiglen, Swissgrid: 651′800–168′745, 1690 m, on *Cladonia rangiferina*, 2013, *Zimmermann 810* (hb Zimmermann).

Epithamnolia rangiferinae E. Zimm., Diederich & Suija, sp. nov. FIG. 6
MycoBank MB822794

Diagnosis: Conidiomata 60–150 µm. Conidiophores septate, simple or branched, of 1–3 cylindrical cells, each 5.5–6.5 × 1.5–2 µm. Conidiogenous cells 10.5–12.5 × 1.4–1.8 µm. Conidia short bacilliform, apically rounded, aseptate, 4.3–5 × 1.4–1.6 µm.

Typification: SWITZERLAND. VALAIS: Oberwald, Grimselpass, westl. Totesee, 2200 m, alpiner Rasen, Windkantenheide, silicate, on thallus of *Cladonia rangiferina*, 2015, ***Zimmermann 1257*** (holotype G, isotype hb Diederich).

Etymology: Referring to the host, *Cladonia rangiferina*.

Mycelium indistinct. Conidiomata cupulate, pale to dark brown, glossy, glabrous, sometimes with an undulating margin, 60–150 µm diam, superficial, dispersed to loosely aggregated; wall medium brown, thin, of loosely or densely interwoven hyphae 1.5–2.5 µm wide, K–. Conidiophores hyaline, arising from the base of the conidioma, branched, septate, composed of 1–3 cylindrical cells (5.2–)5.5–6.6(–6.9) × (1.6–)1.6–2.0(–2.2) µm (N = 15, in Congo red), each cell acting as conidiogenous cell. Conidiogenous cells hyaline, enteroblastic, phialidic, determinate, integrated, acropleurogenous, smooth-walled, apical cells narrowly lageniform or occasionally almost fusiform, (9.5–)10.4–12.4(–13.3) × (1.3–)1.4–1.8 µm (N = 15, in Congo red). Conidia hyaline, short bacilliform, straight, base ± truncate, apex rounded, aseptate, smooth-walled, often with two small guttules, (4.0–)4.3–4.9(–5.3) × (1.3–)1.4–1.6(–1.8) µm, length/breadth ratio (2.5–)2.7–3.3(–3.7) µm (N = 40, in Congo red).

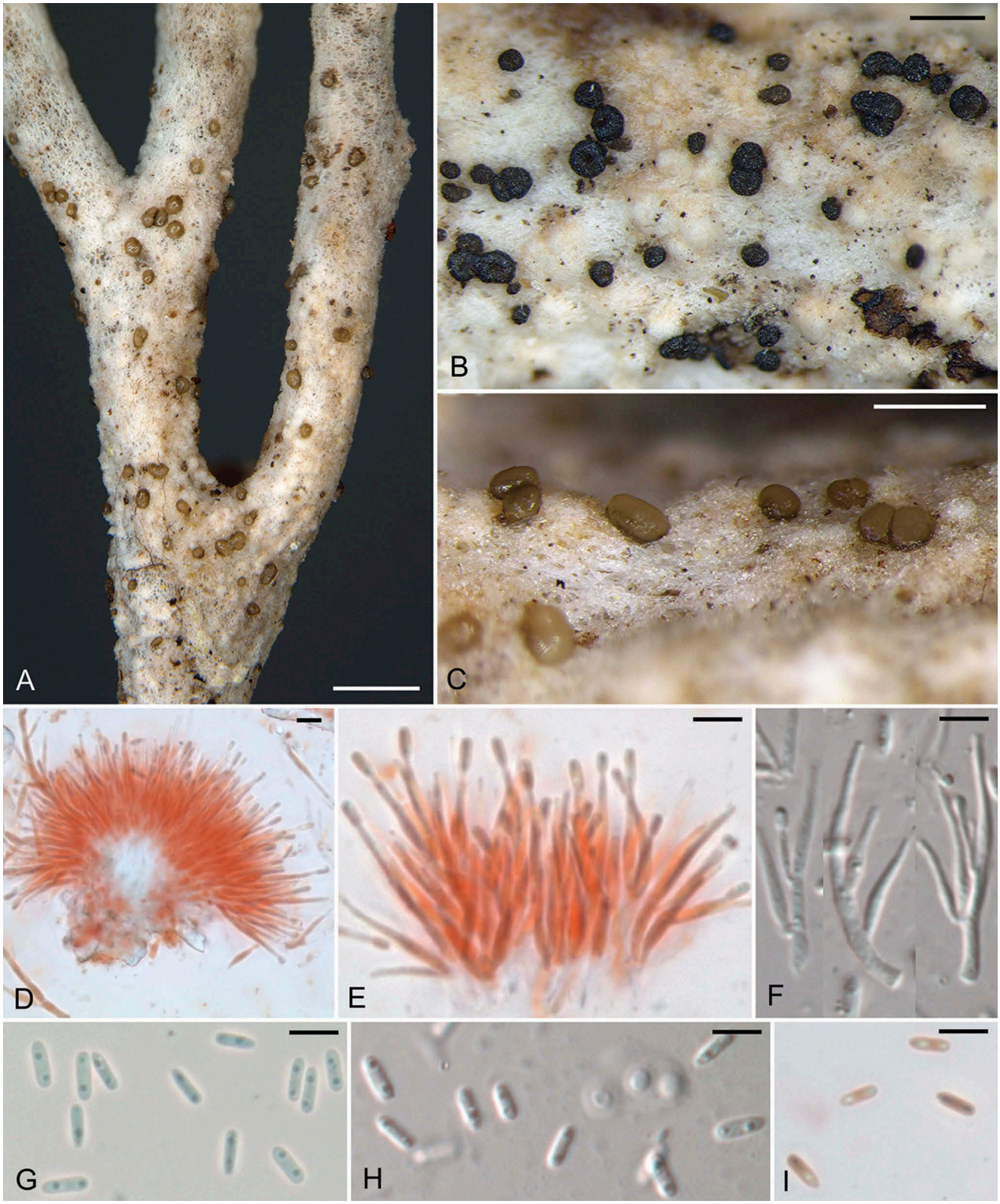

Figure 6. *Epithamnolia rangiferinae* (holotype). A–C. Conidiomata on the thallus surface of *Cladonia rangiferina;* note the pale brown color in the humid state (A and C) and the dark color when dry (B). D–E. Conidiogenous layer in a squash preparation (in Phloxine B). F. Branched conidiophores (in Melzer's reagent). G–I. Conidia in Melzer's reagent (G–H); in Phloxine B (I); using DIC optics (H). Bars: A = 1 mm; B–C = 200 µm; D–I = 5 µm.

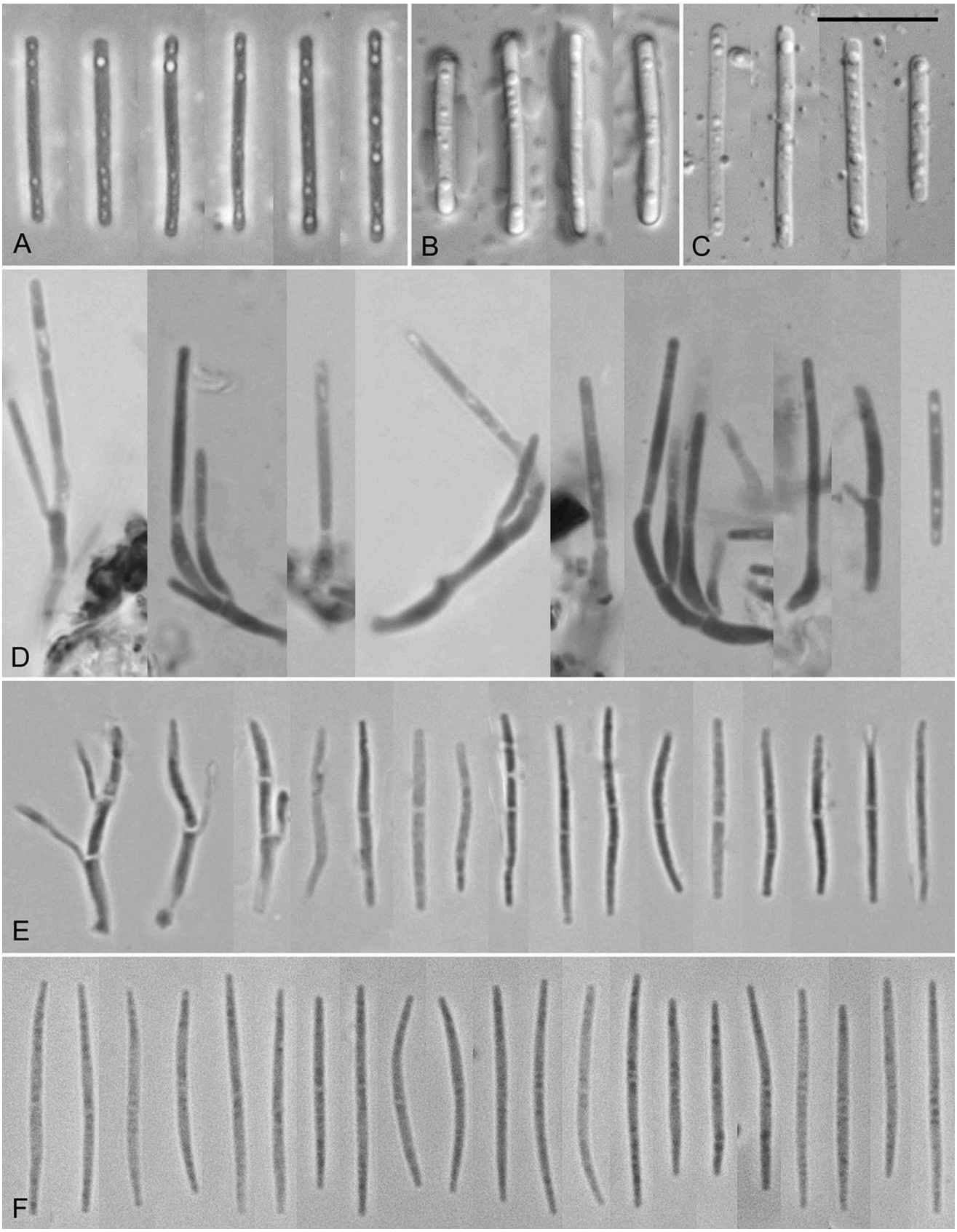

Figure 7. *Epithamnolia* species with medium-sized conidia. A–D. Conidia and conidiophores of E. *brevicladoniae* (A, isotype, hb Diederich; B, Zhurbenko 127; C, Zhurbenko 1216; details on the Zhurbenko specimens in Zhurbenko and Kobzeva 2014). E. Conidia and conidiophores of *E. pertusariae* (isotype, hb Diederich). F. Conidia of *E. pertusariae* on *Cladonia rangiferina* (Zimmermann 810). Bar = 10 µm (same for A–F).

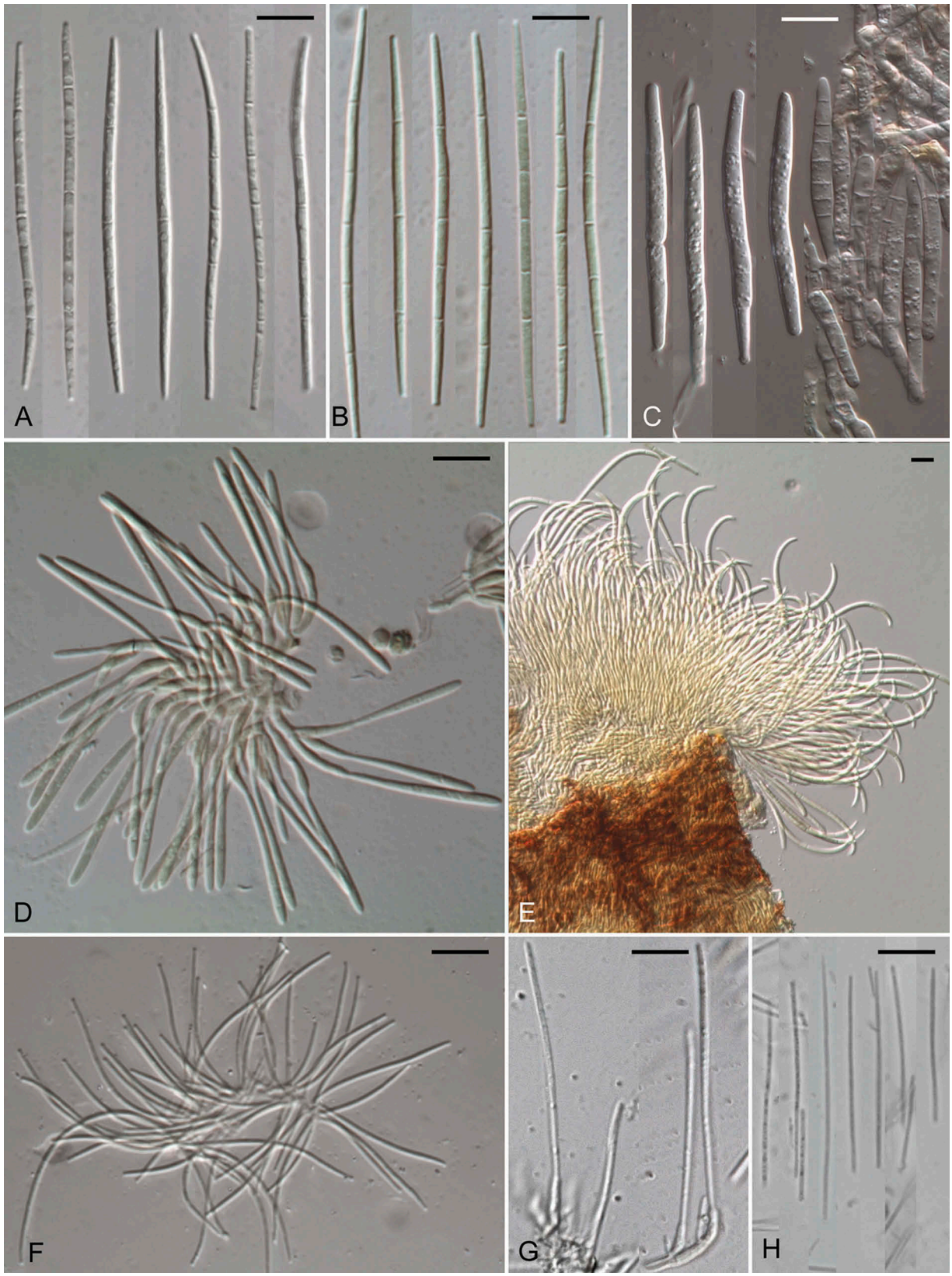

Figure 8. *Epithamnolia* species with long conidia. A–E. Conidia and conidiophores of *E. xanthoriae*. A. On *Xanthoria parietina* (van den Boom 53151, in Melzer's reagent). B. On *Punctelia subrudecta* (van den Boom 49424, in Melzer's reagent). C. Very broad and up to 7-septate conidia on *X. parietina* (Diederich 18191, in water). D. Immature conidia on *Lecanora saligna* (Diederich 17563, in Melzer's reagent). E. Strongly curved conidia on *X. parietina* (van den Boom 52578, in Melzer's reagent). F–H. Conidia and conidiophores of *E. longicladoniae* (F, van den Boom 25449, in Melzer's reagent; G–H, isotype, hb Diederich; G in water; H in Phloxine B); conidia in H are mostly fragments from broken old conidia in a squash preparation. Bars = 10 µm.

Distribution and hosts: Known from three Swiss localities, always on the thallus of *Cladonia rangiferina*. Host not visibly damaged by the presence of the fungus.

Note: This species is very similar to all other *Epithamnolia* species with developed conidiophores, from which it is distinguished by the short, bacilliform, aseptate conidia.

Additional specimens examined (both on *Cladonia rangiferina*): SWITZERLAND. GRAUBÜNDEN: Wergenstein, Caschgliun, Tguma, Swissgrid: 747′300–165′600, 2400 m, Windkantenheide über Kalk, 2015, *Zimmermann 1164*; La Punt, Albulapass, Swissgrid: 784′100–161′850, 2320 m, Windkantenheide, Jul 2016, *Zimmermann 1602* (hb Zimmermann).

Epithamnolia xanthoriae (Brackel) Diederich & Suija, comb. nov. FIG. 7A–E

MycoBank MB822793

≡ *Hainesia xanthoriae* Brackel, Ber Bayer Bot Ges 79:16. 2009 (basionym).

Typification: GERMANY. BAVARIA: Oberpfalz, Kreis Neustadt at Waldnaab, NW Hardt near Flos, 535 m, on thallus (rarely apothecia) of *Xanthoria parietina*, 26 Oct 2007, *Brackel* (holotype hb IVL 4566, isotype M, non vid.).

= *Hainesia aeruginascens* Brackel, Bibl Lichenol 109:131. 2014.

BAVARIA: Oberpfalz, Kreis Schwandorf, Hetschenlack im Neubäuer Forst, 390 m, on thallus of *Platismatia glauca*, 26 Oct 2007, *Brackel* (holotype hb IVL 4569, isotype M, non vid.).

= *Hainesia lecanorae* Brackel, Bibl Lichenol 109:134. 2014.

BAVARIA: Oberbayern, Stadt München, former stand of Riem airport, 48°07′52.7″N, 11°40′59″E, 530 m, on apothecia (rarely thallus) of *Protoparmeliopsis muralis*, 7 Nov 2006, *Brackel* (holotype hb IVL 4588, isotype M, non vid.).

Conidiomata 100–250 µm. Conidiophores septate, simple or branched, of 1–3 elongate filiform cells, each 7–11 × 2–2.2 µm. Conidiogenous cells 5–11 × 1.5–2.5 µm. Conidia filiform, straight or rarely bent, basally slightly truncate, apically rounded, distinctly attenuated towards both ends, 0–5(–8)-septate, (25–)40–84 × (1.8–)2–3(–4) µm (from Brackel 2009, 2014, and own observations).

Distribution and hosts: Epithamnolia xanthoriae, in the broad sense, has been collected in Austria, Belgium, France, Germany, Greenland, Iceland, Italy, Luxembourg, the Netherlands, and Russia, on the thallus or apothecia of *Candelaria concolor, Hypogymnia physodes, H. tubulosa, Lecanora chlarotera, L. saligna, L. symmicta, Melanohalea exasperatula, Parmelia sulcata, Phaeophyscia orbicularis, Physcia stellaris, P. tenella, Platismatia glauca, Polycauliona polycarpa, Protoparmeliopsis muralis, Pseudevernia furfuracea, Punctelia jeckeri, P. subrudecta, Rusavskia elegans*, and *Xanthoria parietina* (Brackel 2009, 2014, 2015; Eichler et al. 2010; Zhurbenko and Kobzeva 2014).

Notes: Conidia of *Epithamnolia xanthoriae* were originally described as 0(–5)-septate, (53–)57–63(–70) × (1.9–)2.1–2.8(–3.2) µm, and conidiomata 100–220 µm diam. Conidia of *Hainesia aeruginascens* as 1(–5)-septate, (65–)72–83(–90) × (1.7–)2.0–2.6(–2.8) µm, and conidiomata 200–250 µm. Conidia of *H. lecanorae* as (0–)3–5-septate, (36–)41–48(–54) × (2.2–)2.4–3.0(–3.8) µm, and conidiomata 100–160 µm. Our own observations suggest that there is a great variability of conidial length in populations resembling *E. xanthoriae*, and that it is not possible, based on this character, to distinguish several species in this group.

We also noticed that there is no correlation between conidial length and host: e.g., in most specimens on *Punctelia subrudecta*, the average conidial length is between 53 and 63 µm, but in specimen Gardiennet 16031 the average is 75.1 µm. In most specimens on *Xanthoria parietina*, the average conidial length is between 55 and 60 µm, but in specimen Diederich 18191 the average is 43.4 µm; conidia in this specimen are also particularly wide, 3.2–3.8 µm. Both specimens Gardiennet 16031 and Diederich 18191 have been included in our phylogenetical analysis, and their sequences group with those of *Epithamnolia xanthoriae*.

We found that conidial septation and length depends on conidiomatal maturity: e.g., in specimen Diederich 17563 on *Lecanora saligna*, aseptate conidia in a young conidioma are (20–)23.4–32.4(–32.4) µm long (N = 14), whereas conidia in a mature conidioma of specimen Diederich 17562, collected in the same locality on *L. symmicta* (sequences group with those of *E. xanthoriae*), are (48–)49.4–57.6(–62) µm long (N = 20).

Additional specimens examined: AUSTRIA. OBERÖSTERREICH: Bez. Schärding, Kopfing 130, 48.43972°N, 13.65667°E, 545 m, on *Xanthoria parietina*, Jan 2015, *Berger 28798*. FRANCE. CÔTE-D'OR: Til-Châtel, La Chalandrue, 47°31′13.48″N, 5°11′41.61″E, on *X. parietina*, Feb 2016, *Gardiennet* (TU 82108); Is-sur-Tille, on *Punctelia*, Feb 2016, *Gardiennet 16031* (TU 82109). ICELAND. 40 km E of Hvammstangi, 20 km S of Blöndulos, just NW of crossing of roads 1 and 721, 65.50494°N, 20.37849°W, on *Lecanora symmicta*, Aug 2013, *Diederich 17562*; ibid., on *L. saligna, Diederich 17563*. LUXEMBOURG. Weimerskirch,

Kuebebierg, 49.63029°N, 6.14069°E, on *X. parietina*, May 2015, *Diederich 18191*. THE NETHERLANDS. NOORD-BRABANT: Eindhoven, De Doornakkers, churchyard, on *P. subrudecta*, Apr 2013, *van den Boom 49369*; W of Soerendonk, Het Goor, on *P. subrudecta*, Apr 2013, *van den Boom 49424*; Oirschot, E side of the village, on *P. subrudecta*, Nov 2013, *van den Boom 50178*; Eindhoven (SW), Blaarthem, churchyard, on *P. subrudecta*, Sep 2012, *van den Boom 48257*; Aalst, W of center, on *P. subrudecta*, Sep 2012, *van den Boom 48243*; NE of Eindhoven, E of Amazonelaan, SW of Eckartdal, Eckartse Bos, on *Candelaria concolor*, Jan 2014, *van den Boom 50227*; SSE of Oirschot, Oirschotse Heide, on *X. parietina*, Jan 2015, *van den Boom 52578*; ibid., Mar 2015, *van den Boom 53151*; ibid., on *P. subrudecta*, Jan 2015, *van den Boom 52584*.

KEY TO THE KNOWN *EPITHAMNOLIA* SPECIES

1. Conidia 4.5–5 × ca. 1.4–1.6 µm, aseptate; on *Cladonia rangiferina*..... *E. rangiferinae*
1′. Conidia >10 µm long..... 2
2. Conidia 10–30 µm long, 0–1(–3)-septate..... 3
2′. Conidia > 35 µm long, 0–6(–8)-septate..... 5
3. Conidia at both ends abruptly truncate, of a similar diameter throughout almost the entire length, not attenuated towards the ends, 12.5–17 × 1–1.5 µm; on *Cladonia*..... *E. brevicladoniae*
3′. Conidia slightly to distinctly attenuated towards both more or less rounded ends..... 4
4. Conidiophores usually reduced to a single conidiogenous cell, rarely of several cells; conidia 18.5–27 × 1.5–2 µm; on *Thamnolia*..... *E. karatyginii*
4′. Conidiophores present, usually of several cells; conidia slightly smaller, 10–24.5 × 1–1.5 µm on various lichens..... *E. pertusariae*
5. Conidia rod-shaped or vermiform, of a similar diameter throughout almost the entire length, not attenuated towards the ends, 1–2 µm diam; on *Cladonia*..... *E. longicladoniae*
5′. Conidia elongate fusiform, distinctly attenuated towards both ends, 2–3 µm diam. on various lichens..... *E. xanthoriae*

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