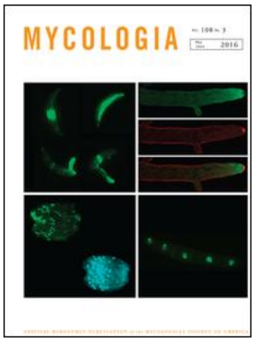

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New species and reports of *Cuphophyllus* from northern North America compared with related Eurasian species

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ABSTRACT

This study describes four gray or brown species of *Cuphophyllus* (Hygrophoraceae, Agaricales), two of them new species, restricted to arctic-alpine and northern boreal zones of North America, and relates them morphologically and phylogenetically using multigene and nuc rDNA internal transcribed spacer ITS1-5.8S-ITS (ITS barcode) analyses to their similar, known counterparts. *Cuphophyllus cinerellus*, epitypified here, is shown to be a pan-palearctic species with sequence-confirmed collections from Fennoscandia and easternmost Asia. Occupying a similar habitat in the Nearctic is its sister species, the morphologically similar but novel *C. esteriae*, so far known only from eastern North America, including Greenland. Sister to the *C. cinerellus*–*C. esteriae* lineage, and known only from boreal raised *Sphagnum* bogs in Newfoundland, is a new medium-sized light cinereous brown species, *C. lamarum*. It has a yellow stipe but is phylogenetically distant from the yellow-stiped European *C. flavipes* and its North American sister species, *Hygrophorus pseudopallidus*. As cryptic speciation was discovered within *C. flavipes*, we lecto- and epitypify the name and transfer *H. pseudopallidus* to *Cuphophyllus* based on ITS analysis of the holotype. We also transfer the small European *Hygrocybe comosa* to *Cuphophyllus* based on morphology. *Cuphophyllus hygrocyboides* is reported from North America with the first sequence-confirmed collections from arctic-alpine British Columbia and Greenland. In addition, sequencing the holotype of *C. subviolaceus* identifies it as the sister species to the putative *C. lacmus*. Both species seem to have an intercontinental distribution. In total, we add new sequences to GenBank from 37 *Cuphophyllus* collections, including the holotypes of *C. hygrocyboides* and *C. subviolaceus*, the two new epitypes, and the two novel species.

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INTRODUCTION

Cuphophyllus (Donk) Bon is a globally distributed genus of Hygrophoraceae Lotzy with species common in diverse ecosystems of the Northern and Southern Hemispheres. Most species in the genus have broad distribution ranges and occur from the nemoral to the arctic-alpine zone (Hesler and Smith 1963; Bon 1985; Borgen and Arnolds 2004; Boertmann 2010). Members of the genus have been documented from boreal, temperate, and tropical regions of North America (Hesler and Smith 1963), but the few arctic-alpine species, well known in Europe (Kühner 1977), have so far not been described in North American texts or monographs.

Cuphophyllus species produce mostly clitocyboid, rarely mycenoid, basidiomata with thick decurrent lamellae and a white spore print. Microscopically, they have interwoven (or rarely almost subregular) lamellar trama, with or without a regular or subregular central strand; smooth, hyaline, inamyloid basidiospores; very long basidia relative to spore length (usually 7–8, rarely 5–6 times spore length); and basal clamps on the basidia (Lodge et al. 2013). The typically interwoven lamellar trama, together with the long basidium-to-spore length ratios, forms the most reliable characters for separating *Cuphophyllus* from other white-spored agaric genera. In a systematic review of Hygrophoraceae, Lodge et al. (2013) noted that *Cuphophyllus* occupied an early-diverging phylogenetic position in the family, but the exact placement relative to *Cantharocybe* H.E. Bigelow & A.H. Sm. and *Ampulloclitocybe* Redhead, Lutzoni, Moncalvo & Vilgalys was not resolved.

Species of *Cuphophyllus* have a biotrophic mode of nutrition, but the nature of the fungus-plant association is largely unknown (Seitzman et al. 2011; Halbwachs

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Supplemental data for this article can be accessed on the publisher's Web site.

et al. 2018). *Cuphophyllus virgineus* (Wulfen) Kovalenko was shown to be a root endophyte of *Plantago lanceolata* and vertically transmitted via seeds (Tello et al. 2014). Other plant associates have not been identified conclusively (Halbwachs et al. 2018).

This study focuses on gray and brown new species and new records of *Cuphophyllus* from arctic-alpine regions and *Sphagnum* bogs of northern regions of North America (Canada, Greenland) and compares them with closely related species from elsewhere in the Northern Hemisphere.

MATERIALS AND METHODS

Selection of species.—Investigating a new species of *Cuphophyllus* in the Canadian province of Newfoundland and Labrador (NL) led to the discovery that it is closely related to the arctic-alpine *C. cinerellus* (Kühner) Bon. This study was initially designed to investigate *C. cinerellus* in eastern Canada and Europe and define its relationship to the new species. During these investigations, we learned of another arctic-alpine species in western Canada (British Columbia) that resembled *C. hygrocyboides* (Kühner) Bon. We enlarged our study to include both arctic-alpine species and their relatives in Canada and compare these with similar species from North America and Europe. We provide a key to the gray, gray-brown, violet-brown, and orange-brown *Cuphophyllus* species in the Northern Hemisphere but have excluded *C. radiatus* (Arnolds) Bon because it is a taxonomic synonym of *C. flavipes* (Britzelm.) Bon (Boertmann 2010). We also excluded *Hygrocybe cinerea* (Pers.) P.D. Orton & Watling and *C. berkeleyi* (P.D. Orton & Watling) Bon because they are poorly circumscribed and *Hygrophorus pallidus* Peck because it is an ambiguous species (Hesler and Smith 1963; Boertmann 2010). Note also that the name *C. lacmus* (Schumach.) Bon, a species with a whitish to cream buff stipe base, was misapplied by Hesler and Smith (1963) to a European taxon with a yellow stipe base that is currently recognized as *C. flavipes* (Boertmann 2010).

Collections and descriptions.—TABLE 1 summarizes data of collections from which new sequences were generated for this study. Additional details about these collections can be found in SUPPLEMENTARY TABLE 1. Specimens were collected using routine field techniques and air dried with heat <20 C. Macroscopic descriptions are based on in situ specimens. Extreme (outlier) measurements are enclosed in parentheses and represent the uppermost and lowermost quartiles based on frequency.

Microscopic examinations were carried out on squash mounts in 2% KOH of both fresh and dried specimens, as well as thin sections of the pileipellis. Basidiospore sizes reported in the descriptions were from multiple observers. A separate single-observer spore size study was carried out on sequence-confirmed specimens of *C. cinerellus*, *C. esteriae*, sp. nov., and *C. lamarum*, sp. nov. Spore length measurements of the two macroscopically most similar species, *C. cinerellus* and *C. esteriae*, were analyzed statistically using the Student's *t*-test and Fisher's test with Yates' correction. The latter test was also used to analyze the significance of differences in basidial length of these two species.

Molecular methods.—DNA extraction and sequencing was carried out at Tartu University (TU) and at the University of Gothenburg (GB). At TU, DNA was extracted with a lysis procedure in 10× Reaction Buffer B (0.8 M Tris-HCl, 0.2 M $(NH_4)_2SO_4$, 0.2% *w*/*v* Tween-20; Solis Biodyne, Tartu, Estonia), including proteinase K (0.5 mg/mL; Thermo Fisher Scientific, Waltham, Massachusetts) and incubated overnight at 56 C. The High Pure PCR Template Preparation Kit (Roche Diagnostics, Mannheim, Germany) was used for older specimens following the protocol of the manufacturer. Polymerase chain reaction (PCR) amplification of nuc rDNA internal transcribed spacer ITS1-5.8S-ITS (ITS barcode) was performed with primers ITSOF/LB-W or ITSOF/ITS4B (Tedersoo et al. 2008) using 5× HOT FIREPol Blend Master Mix Ready to Load (with 10 mM $MgCl_2$; Solis BioDyne) with 0.5 μM of each primer and 1–3 μL of DNA solution. Further PCR amplification and purification protocols follow those described in Saar and Voitk (2015). Sequences were generated by Macrogen Europe (Amsterdam, Netherlands) using primers ITS5 and ITS4 (White et al. 1990). Sequences were inspected and assembled using Sequencher 5.4 (Gene Codes, Ann Arbor, Michigan). DNA sequences, including collection data (TABLE 1), were submitted to GenBank and the PlutoF cloud database (Abarenkov et al. 2010), accessible through public Web output UNITE (http://unite.ut.ee; Kõljalg et al. 2013).

At GB, DNA extractions, PCR, and sequencing of the ITS and nuc rDNA 28S (28S) regions were performed as described in Larsson et al. (2018). For the most variable region of the second largest subunit of RNA polymerase II (*rpb2*), PCR was carried out with primers fRPB2-5F and bRPB2-7.1R as described in Lui et al. (1999) and Matheny (2005) and sequencing with

Table 1. Species names, specimen vouchers, origins, and GenBank accession nos.

Species name	Specimen voucher	Origin	GenBank accession nos. ITS	28S	*rpb2*
Cuphophyllus cinerellus	**GB-0156961**	**Sweden**	**MK573935**	**MN430913**	**MN556847**
Cuphophyllus cinerellus	OF-223054	Norway	MK547070		
Cuphophyllus cinerellus	OF-242127	Norway	MK547071		
Cuphophyllus cinerellus	OF-288058	Norway	MK547069		
Cuphophyllus cinerellus	OF-291322	Norway	MK547072		
Cuphophyllus cinerellus	OF-246174	Norway	MK547073		
Cuphophyllus cinerellus	OF-249863	Norway	MK547074		
Cuphophyllus cinerellus	GB-0156994	Norway	MK573933	MN430914	MN556848
Cuphophyllus cinerellus	GB-0156993	Sweden	MK573934		
Cuphophyllus cinerellus	C-F-47028	Russia	MK836089		
Cuphophyllus esteriae	**DAOM867478**	**Canada NL**	**MK547063**	**MN430911**	**MN556855**
Cuphophyllus esteriae	DAOM867479	Canada NL	MK547064		
Cuphophyllus esteriae	DAOM867480	Canada NL	MK547065		
Cuphophyllus esteriae	DAOM867481	Canada NL	MK547080		
Cuphophyllus esteriae	DAOM867482	Canada NL	MK547078	MN430912	
Cuphophyllus esteriae	DAOM867483	Canada NL	MK547079		
Cuphophyllus esteriae	DAOM867484	Canada NL	MK547081		
Cuphophyllus esteriae	DAOM867485	Canada NL	MK547082		
Cuphophyllus esteriae	C-F-121549	Greenland	MK547075		
Cuphophyllus flavipes	**TU117645**	**Germany**	**MK547077**		
Cuphophyllus flavipes	C-F-107960	Denmark	MK547066		
Cuphophyllus flavipes	C-F-20429	Denmark	MK547068		
Cuphophyllus flavipes	TU117644	Germany	MK547076		
Cuphophyllus flavipes	GB-0156991	Sweden	MK573930		
Cuphophyllus flavipes	TURA199692	Italy	MN453872	MN430919	MN556851
Cuphophyllus flavipes	TURA199694	Italy	MN453873	MN430920	MN556852
Cuphophyllus cf. *flavipes*	GB-0152637	Sweden	MK573932	MN430909	MN556849
Cuphophyllus cf. *flavipes*	GB-0152595	Sweden	MK573931	MN430910	MN556850
Cuphophyllus hygrocyboides	**G-00052005**	**France**	**MK573938**		
Cuphophyllus hygrocyboides	DAVFP Berch0283	Canada BC	MN394148		
Cuphophyllus hygrocyboides	GB-0156992	Sweden	MK573937	MN430917	MN534321
Cuphophyllus hygrocyboides	C-F-121571	Greenland	MK573936	MN430918	
Cuphophyllus lacmus	C-F-17644	Denmark	MK547067		
Cuphophyllus lamarum	**DAOM867486**	**Canada NL**	**MK547062**	**MN430915**	**MN556853**
Cuphophyllus lamarum	DAOM867487	Canada NL	MK547061	MN430916	MN556854
Cuphophyllus lamarum	DAOM867488	Canada NL	MK547083		
Cuphophyllus subviolaceus	**NYS-F-3099**	**USA**	**MN326457**		

Note. BC = British Columbia; NL = Newfoundland and Labrador. Type specimens indicated by bold font. For further information, see SUPPLEMENTARY TABLE 1.

primers fRPB2-5F, fRPB2-6F, and bRPB2-7R (Liu et al. 1999; Matheny 2005). Sequences were edited and assembled using Sequencher 5.1.

For the multigene data set, sequences of ITS, 28S, and *rpb2* of *Cuphophyllus* and *Ampulloclitocybe clavipes* (Pers.) Redhead, Lutzoni, Moncalvo & Vilgalys (outgroup) were taken from Lodge et al. (2013). These were aligned together with the newly generated ITS, 28S, and *rpb2* sequences of species of this study. Alignments of the three-gene regions were performed separately using the L-INS-i strategy implemented in MAFFT 7.017 (Katoh and Standley 2013) before concatenation of the data. The final alignment was adjusted using AliView 1.17.1 (Larsson 2014). Most of the characters from the ITS1 and ITS2 regions were excluded from the analysis due to ambiguous alignment.

For inference of phylogenetic relationships of the multilocus data set, heuristic searches for the most parsimonious trees were performed using PAUP* under the maximum parsimony (MP) criterion (Swofford 2003). All transformations were considered unordered and equally weighted. Variable regions with ambiguous alignment were excluded, and gaps were treated as missing data. Heuristic searches with 1000 random-addition sequence replicates and tree bisection-reconnection (TBR) branch swapping were performed. Relative robustness of clades was assessed by the bootstrap (BS) method using 1000 heuristic search replicates with 100 random taxon addition sequence replicates and TBR branch swapping. BS values >70% were considered to be evidence of strong support.

A Bayesian inference (BI) analysis was also carried out in MrBayes 3.2.6 (Ronquist and Heulsenbeck 2012) with a best-fit model of nucleotide evolution supplied separately for each gene region by MrModeltest 2.2 (Nylander 2004). The protein-coding *rpb2* gene was not subjected to partitioning of the third base in each codon. Eight default-setting Metropolis-coupled Markov chain Monte Carlo (MCMCMC) chains were run for 5 million generations, with trees sampled every 2500 generations and an initial burn-in of 500 samples (25%). The MCMC analysis converged well in advance of the burn-in threshold, and chain mixing was found to be satisfactory as assessed by using Tracer 1.5 (Drummond et al. 2012). After discarding the trees prior to the burn-in threshold, a 50% majority-rule consensus phylogram and posterior probabilities were computed from the remaining trees. Posterior probabilities >0.95 were considered to be evidence of strong support. The multilocus data alignment and MP tree files were submitted to TreeBASE (submission ID 25254).

The ITS data set was compiled based on results from the multilocus analysis and data presented in Lodge et al. (2013). In addition to the newly generated sequences, data of *C. canescens* and *C. basidiosus* were included. ITS sequence data of eight collections of the *Cuphophyllus pratensis* (Schaeff.) Bon complex were taken from GenBank and used for rooting of trees. The ITS of the target species in this study were compared with those in GenBank (Clark et al. 2016) and the UNITE database (Kõljalg et al. 2013) to seek additional available sequence data of our target and closely related taxa. Fourteen additional sequences of *C. colemannianus* (A. Bloxam) Bon, *C. flavipes, C. lacmus, C. subviolaceus*, and the holotype of *H. pesudopallidus* Hesler and A.H. Sm. were found and added to the data set. Alignment and phylogenetic analyses were performed as described above except for the BS analysis, which was performed using 1000 heuristic search replicates with 10 random taxon addition sequence replicates and TBR branch swapping, saving 100 trees in each replicate. The ITS alignment and MP tree files have been submitted to TreeBASE (submission ID 25255).

RESULTS

ITS sequence data of 37 specimens of *Cuphophyllus cinerellus, C. flavipes, C. hygrocyboides, C. lacmus*, and *C. subviolaceus* (Peck) Bon from Europe and North America were newly generated for this study, including the holotype of *C. hygrocyboides* (Kühner 69-319, G) and *C. subviolaceus* (NYSF-3099). The sequences have been deposited in GenBank (MK547061–MK547083, MK573930–MK573938, MK836089, MN326457, MN453872–MN453873; see TABLE 1) and also in the UNITE database (UDB numbers; see SUPPLEMENTARY TABLE 1). In addition, 12 28S and 10 *rpb2* sequences were generated for one or two representative specimens of the species examined in this study to place them in a multigene analysis. These sequences have been deposited in GenBank (MN430909–MN430920, MN534321, MN556847–MN556855; see TABLE 1).

The concatenated multilocus data set consisted of 28 taxa and 3569 sites. After exclusion of ambiguously aligned positions, mainly from ITS1 and ITS2 regions, the second intron region of the *rpb2*, and the beginning and the end of the separate gene regions, 2606 sites remained for analysis. Of these, 2045 were constant, 132 were variable but parsimony uninformative, and 429 were parsimony informative. The MP analysis yielded six equally most parsimonious trees (length = 1031 steps, consistency index [CI] = 0.6811, and retention index [RI] = 0.8113). One of these trees is presented in FIG. 1. MP bootstrap analysis recovered *C. esteriae* (74%), *C. lamarum* (100%), and *C. cinerellus* (87%) in a strongly supported clade (100%) (FIG. 1). Sequences of the *C. flavipes* clade emerged as the sister clade to the *C. esteriae–C. lamarum–C. cinerellus* clade with strong support (100%). There was low support across the backbone of this tree, but the *C. fornicatus* clade (100%) emerged as an early-divergent lineage in *Cuphophyllus* but without strong support. The other included taxa formed a weekly supported clade with no supported internal structure, except for the *C. esteriae–C. lamarum–C. cinerellus* and *C. flavipes* clades. The nucleotide evolution model K80+I+G was used for the 5.8S gene, GTR+I+G was used for the 28S gene, and K80+I+G was used for *rpb2* in the BI analysis, which recovered the same topology and terminal clades as the MP bootstrap analysis.

The aligned ITS-only data set consisted of 61 sequences and 873 sites. After exclusion of ambiguously aligned regions, mainly from the beginning and the end of the data set, 838 positions remained for analysis. Of these, 422 were constant, 27 were variable but parsimony uninformative, and 389 were parsimony informative. The MP analysis yielded 432 equally most parsimonious trees (length = 900 steps, CI = 0.7244, and RI = 0.9574). One of these trees is presented in FIG. 2. Bootstrap analysis recovered 10 weakly to strongly supported clades within the ingroup corresponding to *C. flavipes* (99%), *C. pseudopallidus* (64%), *C.* cf. *flavipes* (72%), *C. cinerellus* (89%), *C. esteriae* (92%), *C. lamarum* (100%), *C. colemannianus* (100%), *C. lacmus* (93%), *C. subviolaceus* (83%), and *C. hygrocyboides* (100%). The calculated strict consensus tree recovered the same clades as the BS tree (not shown). The nucleotide evolution model GTR+G was used for the ITS1 spacer, JC was used for the 5.8S gene, and HKY+I+G was used for the ITS2 spacer in the BI analysis, which recovered the same major clades as in the BS analysis.

Sequence data of *Cuphophyllus hygrocyboides* emerged in a moderately supported clade together with *C. colemannianus, C. lacmus*, and *C. subviolaceus*. Sequences of a British Columbian collection of *C. hygrocyboides* occurred within a clade with the type of *C. hygrocyboides* from the French Alps and one Greenlandic and one Fennoscandian specimen, confirming an intercontinental distribution range for this species. Sequence data of *C. lacmus* and

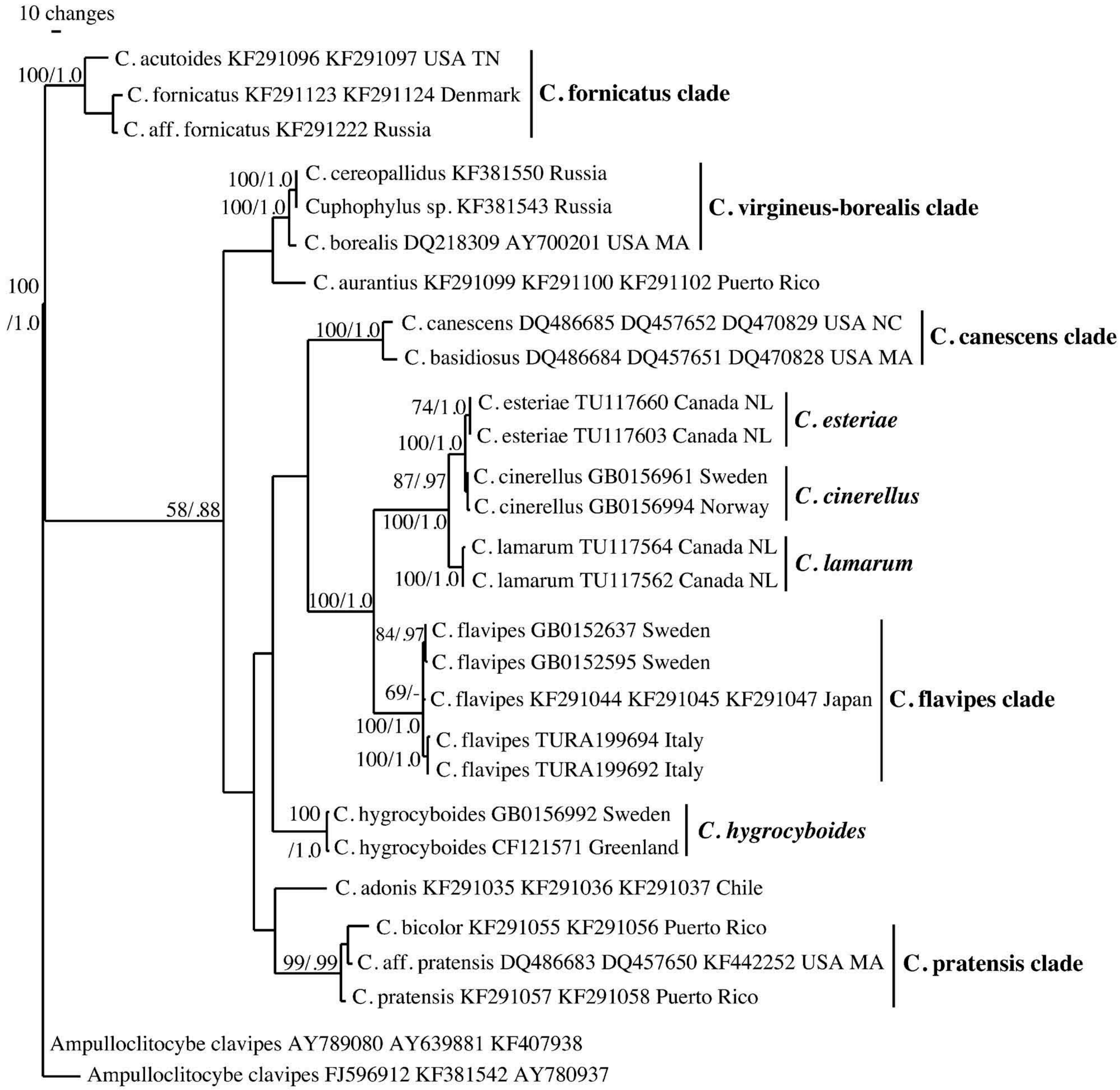

Figure 1. A phylogram from the multigene MP analysis of combined 5.8S, 28S, and *rpb2* sequence data of *Cuphophyllus*, showing the phylogenetic position of the arctic-alpine species treated in this study. Bootstrap values and Bayesian posterior probabilities are indicated on branches. Clades discussed in the text are indicated with bars and species epithets.

C. subviolaceus split in two supported clades, where the sequence of the holotype *of C. subviolaceus*, together with material cited as *Hygrophorus subviolaceus* in Hesler and Smith (1963; Matheny and Wolfenbarger, unpublished), fell within one of the clades, thereby fixing the clade name; the other is putatively named *C. lacmus* and represented by two collections from Denmark and British Columbia.

Cuphophyllus cinerellus formed a well-supported species-level lineage of several Fennoscandian collections and one collection from the Kamchatka Peninsula in the Russian Far East. Sister to it was the moderately supported new species, *C. esteriae*, with one collection from Greenland and eight from Newfoundland and Labrador. Sister to these two was the new species, *C. lamarum*, so far only known from *Sphagnum* bogs in the boreal region of Newfoundland and Labrador. Collections identified as the European species *C. flavipes* form two independent lineages (*C. flavipes* on one hand and *C.* cf. *flavipes* on the other), suggesting occurrence of cryptic speciation in this group. In addition, the ITS of the holotype of

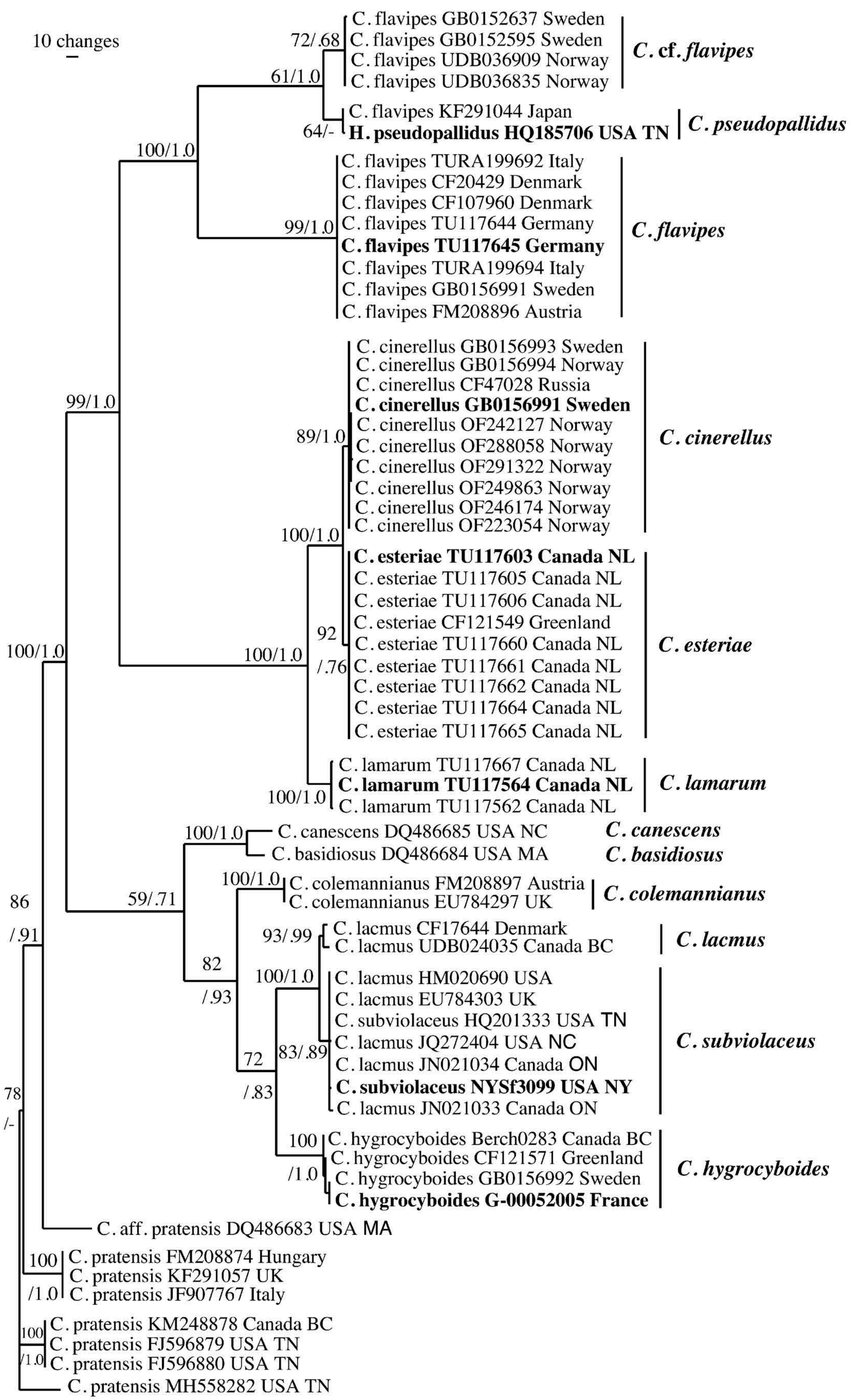

Figure 2. Phylogram showing the phylogenetic relationships among arctic-alpine species of *Cuphophyllus* treated in this study based on ITS sequence data. Bootstrap values and Bayesian posterior probabilities are indicated on branches. Clades discussed in the text are indicated with bars and species epithets. Sequences originating from type specimens are marked in bold.

H. pseudopallidus from North America (HQ185706; Matheny and Wolfenbarger, unpublished) was nearly identical to the ITS (KF291044; Lodge et al. 2013) of a Japanese specimen originally identified as *C. flavipes*; these two sequences form a monophyletic group but with low support. However, the low support value in the analyses may be due to partly missing data and a few ambiguous sites. Based on the result, we transfer *H. pesudopallidus*, now known from both eastern North America and Japan, to *Cuphophyllus*.

Spore size ranges for *C. cinerellus, C. esteriae*, and *C. lamarum* showed such overlap that the values were judged to be unhelpful for identification (see TAXONOMY). Single-observer spore measurements showed that the range in spore size of *C. esteriae* was greater than for *C. cinerellus*, the latter falling entirely within the range of the former (SUPPLEMENTARY FIG. 1). The average spore size measurements tended to cluster for each species but in very close proximity with the average size of *C. cinerellus*, slightly greater than that of *C. esteriae*. Statistical analysis with the Student's *t*-test showed that the observed differences in spore length were highly significant ($P = 0.0001$), and the difference between the occurrence of spores longer than 8 μm between the two species (19% for *C. esteriae* and 45% for *C. cinerellus*) was also significant (Fisher's test with Yates' correction, $P = 0.0004$). The difference in basidial size between the two species was much more obvious: most basidia of *C. cinerellus* were longer than 40 μm, whereas we did not find any basidia of *C. esteriae* longer than 40 μm; this difference was highly significant (Fisher's test with Yates' correction, $P = 0.0045$). A summary of morphological characters, habitat, and ecology used to identify and discriminate between the newly described species and similar species in *Cuphophyllus* is presented in SUPPLEMENTARY TABLE 2.

TAXONOMY

Cuphophyllus cinerellus (Kühner) Bon, Doc Mycol 14-(56):10. 1985. FIGS. 3A, 4A
MycoBank MBT389216

Epitypification: SWEDEN. TORNE LAPPMARK: Jokkmokk, Padjelanta, Arranoajvve, 11 Aug 2016, *E. Larsson 30-16* (**epitype** here designated GB-0156961). GenBank: ITS = MK573935, epitype (UNITE SH1329439.08FU).

≡ *Camarophyllus cinerellus* Kühner, Bull Soc Mycol Fr 93(1):144. 1977.

≡ *Hygrocybe cinerella* (Kühner) Arnolds, Persoonia 13:386. 1987.

Pileus (5–)10–30(–35) mm wide, plano-convex with somewhat involute margin, applanate in age with depressed center, rarely slightly umbonate, often with crenate margin; surface when young translucently striate half way to the center, dry, first smooth, later often becoming finely scaly especially at the center; fuscous to gray-brown, pale grayish brown with age, hygrophanous, becoming pale grayish buff upon drying, often with a lilac tint. Lamellae short to deeply decurrent, arcuate to triangular, furcate, pale gray to grayish brown, sometimes with a slight lilac tint. Stipe 10–45 × 2–6 mm, more or less equal and/or tapering toward the base, dry, slightly fibrillose; pale gray to grayish buff, often with a yellow tone in the lower third, sometimes with a whitish removable covering. Context concolorous, without a distinct odor or taste.

Basidiospores (6.5–)7.5–8(–9.5) × 5–6(–6.5) μm, average 8.0 × 5.4 μm, smooth, broadly elliptic to elliptic, occasionally approaching subglobose or pip-shaped, thin-walled, hyaline, white in deposit, $Q = 1.20–2.00$, average 1.50 (101 spores, 3 basidiocarps, 3 collections). Basidia 32–54 × 6.5–13.5 μm, 4-spored, 2-spored observed in some collections, clavate, hyaline. Basidioles numerous 25–40 × 3–5 μm, cylindrical to narrowly clavate. Cystidia absent. Lamellar trama subregular to slightly interwoven, made up cylindrical cells 40–75 μm long × 7–20 μm wide. Pileipellis a dense cutis of narrowly cylindric hyphae 3–7 μm wide. Clamp connections in all tissues.

Ecology and distribution: In arctic-alpine zones, usually on nutrient poor soils, in dry lichen alpine heath with *Empetrum nigrum, Vaccinium*, and *Betula nana*, also in moist *Sphagnum* and *Polytrichum* bogs among *Carex* and *Vaccinium*. Presumably biotrophic with moss and/or some other paludine plant associate, occurring Jun–Sep, primarily known from northern Fennoscandia, also from the Kamchatka Peninsula.

Specimens examined: NORWAY. Troms, 19 Aug 2017, *E. Larsson EL170-17* (GB-0156994). SWEDEN. LYCKSELE LAPPMARK: Hemavan, Laisaliden, 10 Aug 2015, *E. Larsson EL118-15* (GB-0156993); TORNE LAPPMARK: Abisko, Slåttatjåkkå, 14 Aug 1972, *R. Kühner 72-127* (**holotype** G00126049); see also SUPPLEMENTARY TABLE 1.

Commentary: The Eurasian distribution, longer and wider basidia, and variation in ITS sequences distinguish it from the very similar *C. esteriae* described below. The small size of the basidiomes, the often finely scaly pileus, and arctic-alpine habitat distinguish it from the more brownish and lighter-colored *C. lamarum* and species in the *C. lacmus* and *C. flavipes* complexes. This species has more pleomorphic spores than the other species studied, although size range is closer than for others.

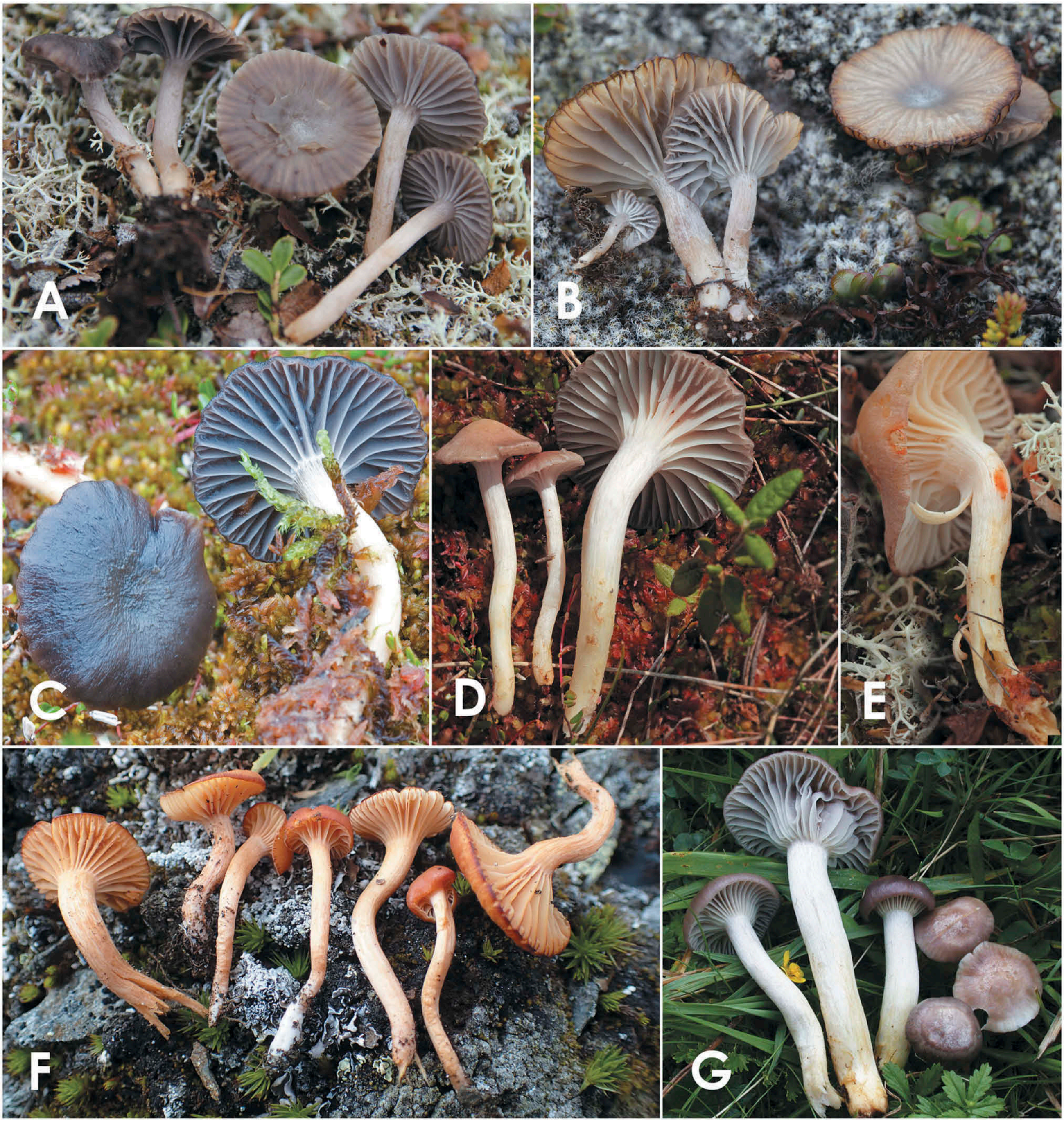

Figure 3. Described species of *Cuphophyllus*. A. *Cuphophyllus cinerellus* (epitype, EL30-16). B. *Cuphophyllus esteriae* (TCAV-04). C. *Cuphophyllus esteriae* (BH-097). D. *Cuphophyllus lamarum* (holotype, 10.07.13.av01). E. *Cuphophyllus lamarum*, single collections showing orange staining (10.07.28.av01). F. *Cuphophyllus hygrocyboides* (Berch0283). G. *Cuphophyllus flavipes*, photographed 2002, in same site as epitype (photo, Peter Karasch). Bars = 25 mm.

Cuphophyllus comosus (Bas & Arnolds) Lodge, Boertm. & E. Larss., comb. nov.

MycoBank MB833466

≡ *Hygrocybe comosa* Bas & Arnolds, Persoonia 19:251. 2007 (basionym).

Commentary: *Hygrocybe comosa* resembles *C. cinerellus* and *C. esteriae* and is clearly by morphology a species that belongs in *Cuphophyllus* by virtue of the thick subdecurrrent lamellae; hyaline, inamyloid, ovoid to ellipsoid basidiospores; basidium-to-basidiospore length ratios of 5.2–6.2; and irregular arrangement of subcylindrical hyphae in the lamellar trama (Bas and Arnolds 2007). Although this taxon was not specifically included in this study of arctic-alpine

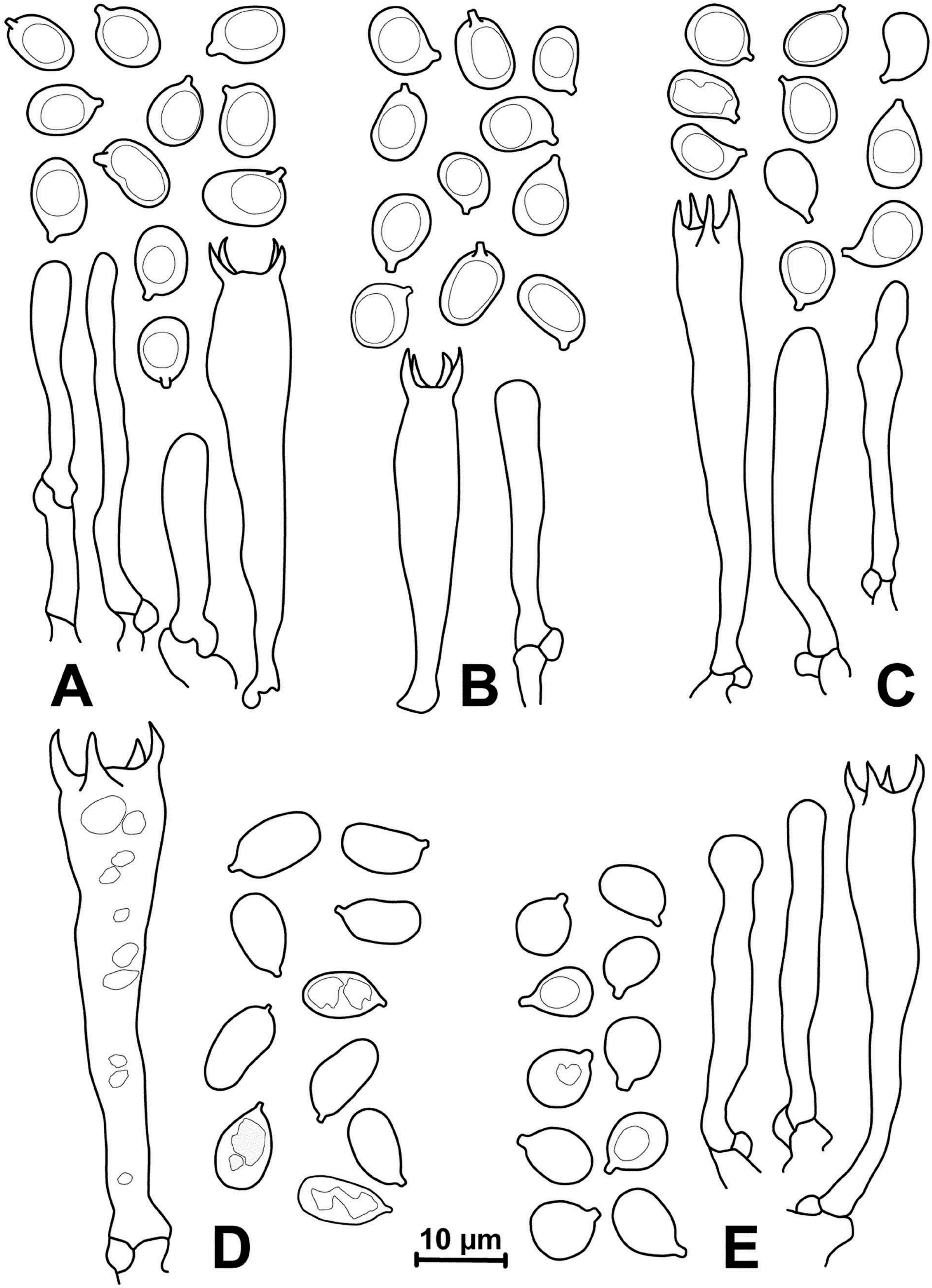

Figure 4. Microscopic drawings of the species described. A. *Cuphophyllus cinerellus* (epitype, EL30-16). B. *Cuphophyllus esteriae* (TCAV-09). C. *Cuphophyllus lamarum* (holotype, 10.07.13.av01). D. *Cuphophyllus hygrocyboides* (Berch0283). E. *Cuphophyllus flavipes* (epitype, G/122-00). Bar = 10 µm; same scale for all drawings.

species, we have included it for comparison based on morphology in the key. We therefore here make the combination in *Cuphophyllus*.

Cuphophyllus esteriae Voitk, I. Saar & E. Larss., sp. nov. FIGS. 3B–C, 4B

MycoBank MB833018

Typification: CANADA. NEWFOUNDLAND AND LABRADOR: Labrador, l'Anse Amour, in ericaceous heath with moss and *Sphagnum* (51.461222°N, 56.858738°W; 21 m above sea level [asl]), 15 Aug 2008, *Maria Voitk FNL Coll LS-075 (lat6)* (**holotype** DAOM867478, **isotype** TU117603). GenBank: ITS = MK547063 (UNITE SH1329438.08FU).

Etymology: *esteriae* (L.) honors Esteri Ohenoja, who has made a lasting contribution to the interest in and knowledge of arctic-alpine mycota in Newfoundland and Labrador. She was the first to collect and show us this species, thought at the time to be *C. cinerellus.Diagnosis*: A small *Cuphophyllus* from arctic-alpine barrens, very similar to *C. cinerellus* and less so to *C. lacmus* or *C. subviolaceus*. Differing from *C. cinerellus* by the Nearctic distribution and smaller basidia, and from *C. lacmus and C. subviolaceus* by the smaller basidiome size and habitat. The sequence divergence between *C. esteriae* and *C. cinnerellus* is 4 substitutions and two 3 bp and one 1 bp insertion/deletion events in the ITS1 region and 3 substitutions and one 3 bp and two 1 bp insertion/deletion events in the ITS2 region.

Pileus 4–32 mm wide, domed, often developing a shallow central depression, edges curved down becoming plane, occasionally even shallowly funnel-shaped, margin somewhat crenulate with age; surface lubricious when wet, at times becoming scurfy with age, opaque to translucently striate to the disc, hygrophanous; gray to dark grayish brown with violet tones, drying pale violaceous gray. Lamellae arced to decurrent, distant, not forked, usually only one tier of lamellulae, increasing cross-veining in age; light to dark brownish violet gray, edges slightly lighter than the faces. Stipe 6–28 × 1.5–4.5 mm, cylindric, usually straight; surface lightly fibrillose, glabrescent with age; light brownish gray but may be somewhat yellowish near the base. Context whitish, odor unremarkable.

Basidiospores (6–)6.5–7.5(–10.5) × (4–)5–6(–6.5) µm, average 7.6 × 5.3 µm, broadly elliptical, elliptical and occasionally subglobose or pip-shaped, hyaline, white in deposit, Q = 1.00–1.80, average 1.40 (190 spores, 9 basidiomes, 9 collections). Basidia 22–40 × 6–8 µm, 4-spored, 4-spored observed <10%, clavate, hyaline. Basidioles numerous. Cystidia absent. Lamellar trama subregular to slightly interwoven, made up cylindrical cells 40–75 × 5–15 µm. Pileipellis a dense cutis with hyphae 4–8 µm wide, but in places upright or trichodermial in appearance. Clamp connections in all tissues.

Ecology and distribution: In heath or arctic-alpine pioneer soil, often with *Sphagnum* or other moss, in small groups of one to four separate basidiomes. Presumably biotrophic with mosses and/or some other paludine plant associates, Jun–Sep, so far known from eastern North America (Greenland, Newfoundland and Labrador).

Other specimens examined: CANADA. NEWFOUNDLAND AND LABRADOR: Labrador, Battle Island (52.273698°N, 55.585155°W; 32 m asl), in moist pioneer soil with mosses including *Sphagnum*, 18 Aug 2008, *Andrus Voitk FNL Coll BH-096* (DAOM867479; TU117605; GenBank ITS = MK547064; UNITE = UDB034650); Labrador, Great Caribou Island (52.269026°N, 55.591865°W; 38 m asl), in barren heath among *Sphagnum*, 21 Aug 2008, *M. Burzynski FNL Coll BH-097* (DAOM867480, TU117606; GenBank ITS = MK547065; UNITE = UDB034651); Labrador, Great Caribou Island (52.269026°N, 55.591865°W; 38 m asl), in barren heath on pioneer soil, 20 Aug 2018, *A. Marceau FNL Coll CEAV-02* (DAOM867481, TU117662; GenBank ITS = MK547080; UNITE = UDB038329); ibid., in boggy area among *Sphagnum* on barren heath, 20 Aug 2018, *M. Voitk FNL Coll CEMV-03* (DAOM867482, TU117660; GenBank ITS = MK547078; UNITE = UDB038327); ibid., in moss on barren heath, 21 Aug 2018, *A. Voitk FNL Coll TCAV-01* (DAOM867483, TU117661); ibid., 43 m asl, in pioneer soil among *Rhacomitrium lanuginosum* on barren heath, 21 Aug 2018, *A. Voitk FNL Coll TCAV-04* (DAOM867484, TU117664; GenBank ITS = MK547081; UNITE = UDB038330); ibid., in bog among *Sphagnum*, 21 Aug 2018, *A. Voitk FNL Coll TCAV-09* (DAOM867485, TU117665; GenBank ITS = MK547082; UNITE = UDB038331).

Commentary: The shorter and narrower basidia, ITS sequence differences, and the North American distribution distinguish it from the very similar *C. cinerellus. Cuphophyllus esteriae* differs from *C. lamarum* by the arctic-alpine habitat, smaller basidiomes, darker pileus, more violet coloration, absent or only occasional pale yellow tint on the stipe base, somewhat longer spores, and ITS sequence data. Although found at times in *Sphagnum*, the habitat is entirely different from that of *C. lamarum* (described below). Like *C. cinerellus, C. esteriae* is one of the early macrofungi to colonize pioneer soil, appearing after lichens and bryophytes (including *Sphagnum* in wetter areas), together with some *Vaccinium* and other ericaceous plants.

Cuphophyllus flavipes (Britzelm.) Bon, Doc Mycol 14(56):11. 1985. FIGS. 3G, 4E

MycoBank MBT389217 (lectotype), MBT389219 (epitype)

Lectotypification: Hymenomyceten aus Südbayern teil 8: plate 448, no. 69. 1891 (**lectotype** here designated). *Epitypification*: GERMANY. Augsburg bei Ammersee, Goassl-Weide, Hartschimmel-Hof, 735 m asl, 28 Oct 2000, *Peter Karasch* (**epitype** here designated, TU117645). GenBank/UNITE: MK547077/UDB035647. UNITE SH1159999.08FU.

≡ *Hygrophorus flavipes* Britzelm., Hymenomyc Südbayern 8:10. 1891.

≡ *Camarophyllus flavipes* (Britzelm.) Clémençon, Beih Z Mykol 4:55. 1982.

≡ *Hygrocybe flavipes* (Britzelm.) Bon, Doc Mycol 14(53):6. 1985.

Pileus 18–39 mm wide, convex, smooth, lubricous (with adhered plant material), pale silver gray to brownish violet, translucently striate, hygrophanous. Lamellae deeply decurrent, arcuate, intervenose, pale gray. Stipe 60–100 × 35–55 mm, cylindrical, somewhat twisted, longitudinally fibrillose, pale gray, marbled and with yellow base. Context pale gray fibrous. Odor and taste, none.

Basidiospores smooth, subglobose to broadly elliptic, a few pip-shaped, (5–)6–7.5(–8) × (4.5–)5–6(–7) μm, average 6.8 × 5.8 μm (a single spore was 8 μm long and probably derived from a 2-spored basidium), Q = 1.02–1.47, average Q = 1.20 (10 spores, 1 basidiocarp, 1 collection). Basidia 40–58 × 6.5–8 μm, 4-spored, only very few 2-spored observed. Basidioles numerous, club-shaped, a few capitate, 19–36 × 3–4 μm. Pileipellis a thin (20–40 μm) ixocutis from a small and young basidiomes, a zebra pattern (incrusted pigment) on cells in subpellis not seen. Lamellar trama interwoven with cylindrical and branched noninflated cells, 3–7.5 μm wide. Lamellar edge fertile. Clamps abundant in all tissues.

Ecology and distribution: In acidic unimproved grasslands and lawns, widespread but rare, from lowland to montane, up to 2300 m asl, epitype in grassland among *Picea* and *Fagus* 735 m asl, German Alps, and confirmed from Austria, Denmark, Italy, and Sweden. *Specimens examined*: DENMARK. JYLLAND: Tågelund, 4 Sep 1993, *J. Vesterholt JV93-829* (C-F-20429); JYLLAND: Mosegård Strand, 30 Aug 1985, *J. Vesterholt JV85-747* (C-F-107960). GERMANY. OBERBAYERN: Ausburg bei Ammersee, 25 Sep 1999, *P. Karasch S/250999-1* (TU1117644). ITALY. ARBA: Pordenone, 27 Oct 2013, *E. Campo* (TURA199692); LONGARONE: Belluno, 10 Oct 2013, *E. Campo* (TURA199694). SWEDEN. SMÅLAND: Torsås, 29 Sep 2014, *R-G Carlsson RGC14-104* (GB-0156991)

Commentary: This is a widespread but rare European species, so far not known from North America. The yellow stipe base, fibrillose stipe, and lack of an odor differentiate it from the similar *C. lacmus*. Differs from the North American *C. lamarum* by the darker pileus, lamellae, stipe and context, habitat, and ITS sequence data. Differs from *C. cinerellus* by the size, smooth lubricous pileus surface, yellow stem base, and ITS sequence data.

Cuphophyllus hygrocyboides (Kühner) Bon, Doc Mycol 14(56):12. 1985. FIGS. 3F, 4D

≡ *Camarophyllus hygrocyboides* Kühner, Bull Soc Mycol Fr 93(1):144. 1977.

≡ *Hygrocybe hygrocyboides* (Kühner) Arnolds, Persoonia 13:386. 1987.

Pileus 10–30 mm wide, pulvinate when young, flattened to somewhat depressed with age, dry on older fruiting bodies, slightly sticky when young; surface hygrophanous, edge variably translucently striate, bright to dark orange-brown. Lamellae decurrent, subdistant, rather thick, intervenose, pale orange-brown. Stipe 20–40 × 2–4 mm, subcylindrical or slightly tapering downward, straight or slightly sinuous, dry and longitudinally fibrillose at least in age, pale orange-brown (paler than pileus). Context concolorous, odor unremarkable.

Basidiospores (6–)7–9(–10) × 4.5–5.5 μm, smooth, ellipsoid to ovoid, some with slightly concave side in side view, white in deposit (15 spores, 2 basidiomata 1 collection). Basidia 50–72 × 5.5–7 μm, 4-spored or occasionally 2-spored. Basidioles numerous, cylindrical to narrowly clavate, 30–58 × 2.5–4 μm. Pileipellis a cutis, hyphae 1.5–2.5 μm wide. Stipitipellis a cutis with hyphae 2–3 μm broad. Lamellar trama tightly interwoven and difficult to split in squash preparations. Clamps abundant in all tissues.

Ecology and distribution: On ground in alpine zone with moss and *Salix stolonifera* Coville, single or in groups of separate basidiomes. Likely circumboreal, known from the Alps, the Pyrenees, northern Fennoscandia, Greenland (Borgen and Boertmann 2008; Boertmann 2010), and British Columbia.

Specimens examined: CANADA. BRITISH COLUMBIA: North of Smithers, near Motase Peak in the Sicintine Range of the Skeena Mountains (56°8′46.9″N, 127°12′33.0″W; 1805 m asl), 26 Jul 2016, *S. Berch 0283* (DAVFP). FRANCE. SAVOIE: Parc National de la Vanoise, Région de Pralognan, 14 Sep 1969, *R. Kühner 69-319* (**holotype** G00052005). SWEDEN. TORNE LAPPMARK: Jukkasjärvi, Latnjavagge, 22 Aug 2013, *E. Larsson EL118-15* (GB-0156992).

Commentary: To our knowledge, *C. hygrocyboides* is the only such species of *Cuphophyllus* in the underexplored Canadian arctic-alpine habitat. In Fennoscandia, the multispecific *C. pratenis* has also been identified in high arctic regions. The bright orange-brown color and smaller size distinguish *C. hygrocyboides* from the darker brown *C. colemannianus*, which also has a northern distribution but is not found in exposed barrens. This is the first sequenced-confirmed report of *C. hygrocboides* from North America.

Cuphophyllus lamarum Voitk, Boertm. & I. Saar, sp. nov. FIGS. 3D–E, 4C

MycoBank MB833019

Typification: CANADA. NEWFOUNDLAND AND LABRADOR: West of the TransCanada Highway near the southern tip of Grand Lake (48.668595°N, 58.206114°W; 199 m asl), small group in raised *Sphagnum* bog, 13 Jul 2010, *A. Voitk 10.07.13.av01* (**holotype** DAOM867486, **isotypes** TU117564, C-F-121518). GenBank: ITS = MK547062 (UNITE UDB034246).

Etymology: lamarum (L.) of the bogs, in reference to the preferred habitat.

Diagnosis: A sphagnicolous North American *Cuphophyllus* with a light brown pileus and varying amounts of violaceous hues, yellow stipe, larger than *C. esteriae*. Sequence divergence between *C. esteriae* and *C. lamarum* is 23 substitutions and one 45 bp, one 9 bp, one 5 bp, one 3 bp, and two 1 bp insertion/deletion events in the ITS1 region and 20 substitutions, one 11 bp, and seven 1 bp insertion/deletion events in the ITS2 region.

Pileus 20–55 mm wide, round, domed, becoming almost plane with age, edge somewhat in-rolled well into maturity; surface dry to minimally lubricious, opaque, but indistinctly translucently striate at edge, hygrophanous; light to moderately tan, with light grayish violet hues, more yellowish over disc and gray-violet over periphery, drying cream-colored. Lamellae decurrent, distant, not forked, with two tiers of lamellulae and increasing cross-veining in age, whitish gray. Stipe 25–70 × 4–10 mm, uniform, even, often slightly curved; surface dry, light straw-colored, with increasing yellowish tones toward base. Flesh cream-colored; staining orange in injured areas in one of three specimens; smell unremarkable.Basidiospores (5.5–)6.5–7(–9) × (4.5–)5–6.5(–7) µm, smooth, subglobose, broadly elliptic, to pip-shaped, occasionally cylindrical, hyaline, white in deposit, Q = (1.10–)1.20–1.40(–1.80), average 1.30 (70 spores, 4 basidiocarps, 3 collections). Basidia 38–78 × 6–9 µm, 4-spored, clavate. Basidioles simple and numerous. Cystidia absent but tramal cells protruding at lamella edges may resemble cystidia (observed in one basidiome). Lamellar trama interwoven, cells 2.5–8 µm wide. Pileipellis a cutis with a few gelatinized hyphae on top, hyphae 1.5–5.5 µm wide. Clamp connections in all tissues.

Ecology and distribution: In small groups on raised *Sphagnum* bogs in western Newfoundland, presumably biotrophic with *Sphagnum* and/or some other paludine photobiont, Jul, at least a month before other species of the genus in this region.

Specimens examined: CANADA. NEWFOUNDLAND AND LABRADOR: Route 401 to Howley (49.212693°N, 57.072087°W; 98 m asl), in raised *Sphagnum* bog, 28 Jul 2010, *A. Voitk 10.07.28.av01* (DAOM867487, TU117562); ibid., in raised bog within 500 m of the former, 20 Jul 2018, *T. Booth 18.07.20.av01* (DAOM867488, TU117667).

Commentary: Other species of *Cuphophyllus* (e.g., *C. esteriae, C. cinerellus*) may be found with or near *Sphagnum*, in addition to other bryophytes, but to our knowledge *C. lamarum* is the only species of *Cuphophyllus* seemingly limited to raised *Sphagnum* bogs. However, because our experience is limited to three collections, this conclusion may be premature. Similarly, we have documented an apparent orange staining reaction to injury in one of three collections (FIG. 3E). Without more experience, the significance of this finding is unclear. *Cuphophyllus lamarum* differs from its close relatives, *C. cinerellus and C. esteriae*, by the unique habitat, boreal distribution, larger basidiome size, tannish pileus color, more yellow at the base of the stipe, statistically shorter spores, and ITS sequence data. The species is similar in size and shape to species in the *C. flavipes* and *C. lacmus-subviolaceus* complexes. The yellow stipe separates it from the latter complex, and the unique habitat in raised *Sphagnum* bogs from both. The North American distribution separates it from Eurasian species. The European *C. flavipes* also has a yellow stipe base but differs from *C. lamarum* by a lubricous pileus surface and darker pileus, lamellae, and stipe. In addition to the habitat and yellow stipe base, *C. lamarum* differs from the North American *C. subviolaceus* common in the same region by a more tan to brown (rather than gray) pileus and earlier phenology.

Cuphophyllus pseudopallidus (Hesler & A.H. Sm.) Lodge, Boertm. & E. Larss., comb. nov.

MycoBank MB833020

≡ *Hygrophorus pseudopallidus* Hesler & A.H. Sm., N Am Species *Hygrophorus*:94. 1963 (basionym).

Commentary: Phylogenetic analyses including the ITS sequence data of the holotype of *H. pseudopallidus* show that it belongs in *Cuphophyllus* and is closely related to *C. flavipes*. This is also incongruence with morphology. We therefore make the combination to *Cuphophyllus*.

KEY TO GRAY AND BROWN NORTHERN SPECIES OF *CUPHOPHYLLUS* FROM NORTH AMERICA AND EURASIA

1. Basidiomes predominantly warm brown, bay, tan, cream, or orangish brown 2
1′. Basidiomes predominantly gray or violaceous . 4
2. Stipe yellow at least near the base . . *C. lamarum*
2′. Stipe not yellow 3
3. Pileus <30 mm wide, brownish orange, orange; arctic-alpine .... *C. hygrocyboides*
3′. Pileus often >30 mm wide, brown; primarily in temperate climate zone but also known from arctic-alpine habitats .... *C. colemannianus*
4. Stipe base yellow .... *C. flavipes* complex (including *C. pseudopallidus*)
4′. Stipe base colored like rest of stem, not yellow 5
5. Pileus often >30 mm wide, smooth, moist to greasy .... *C. lacmus-subviolaceus* complex
5′. Pileus often <30 mm wide, dry or viscid, smooth or scaly toward the center 6
6. Pileus conico-convex when young, retaining a papilla with erect squamules .... *C. comosus*
6′. Pileus convex to plano-convex when young, scales minute on disc 7
7. In Eurasia, most basidia >40 µm long .... *C. cinerellus*
7′. In North America, all basidia ≤40 µm long .... *C. esteriae*

DISCUSSION

Cuphophyllus comprises a few cosmopolitan species interspersed among several parochial species, similar to several other fungal genera, e.g., *Morchella* Dill. ex Pers. (Richard et al. 2015) and *Cantharellus* Adans. ex Fr. (Buyck et al. 2014). The arctic-alpine species we review show examples of each: *C. hygrocyboides* forms a single clade with only minor intraspecific variation, containing specimens from western North America, Greenland, Fennoscandia, and central Europe. By contrast, the clade containing *C. cinerellus* seems to be limited to Eurasia. Macroscopically similar collections from eastern North America (Greenland, Newfoundland, and continental Labrador) form a homogenous, distinct, and well-supported sister clade of *C. cinerellus*, the new species *C. esteriae*.

We did not expect to find two cryptic species identified by the same name and initially debated among ourselves about the validity of or need for considering *C. cinerellus* and *C. esteriae* as separate species, in view of their macroscopic similarity and phylogenetic proximity. More critical reviews produced convincing evidence to support their separation: (i) they occupy different biogeographic regions separated by the Bering Strait on one side and the North Atlantic Ocean on the other, both significant barriers to genetic mixing; (ii) their molecular divergence has high statistical support, evidence that this is due to factors other than chance, whereas (iii) within their clades they (particularly *C. esteriae*) are homogeneous; (iv) the minimum distance to the closest UNITE "species hypothesis" (SH) is a 2.5% difference in basepair sequences, a value generally accepted as a valid indicator of species difference (https://unite.ut.ee); (v) the statistically (if not practically) significant difference in spore size; and (vi) the significant and readily perceptible difference in the size of their basidia.

When introducing new taxa, the onus is on the author to ensure that the species has not been described before. We have separated the new North American species *C. esteriae* from the similar European *C. cinerellus*. *Cuphophyllus comosus*, a rare small species as yet not sequenced, found once among mosses in peat bogs of the French Juras (Bas and Arnolds 2007), shares cinereous coloring with *C. esteriae* and *C. cinerellus* but differs from these species by the European distribution, smaller basidiome size, more slender habitus, greater stipe/pileus ratio, distinctly papillate pileus, large conical squamules on the papilla, and strongly interwoven lamellar trama.

The vast majority of species of the current known *Cuphophyllus* species in North America were described in a monograph on *Hygrophorus* sensu lato by Hesler and Smith (1963). Several of their species (e.g., *H. rainierensis, H. nordmanensis*, H. *burgdorfensis*) were described from higher elevations than our highest, *C. esteriae*, but none came from similar barren arctic-alpine habitats with pioneer soil. In addition, all the northern and/or higher-altitude species documented by Hesler and Smith (1963) were recorded from forests or grasslands, habitats where *C. esteriae* does not occur. Moreover, the three mentioned species also deviate by a viscid pileus. *Cuphophyllus esteriae* is distinguished from *C. recurvatus* (Peck) Lebeuf, described from upstate New York and also reported from Florida, by the arctic-alpine habitat, grayish in contrast to brown pileus, absence of gelatinized hyphae in the pileipellis, and narrower spores than those measured in Peck's type of *C. recurvatus* by S. A. Cantrell (unpublished data; 7–11 × 5.5–8 µm). In addition to characters listed in the individual descriptions, both *C. esteriae* and *C. cinerellus* differ from the other species treated in this work, as well as other members of the *C. lacmus-subviolaceus* and *C. flavipes* complexes, by the arctic-alpine distribution, smaller basidiome size, and ITS sequences. The lack of previous North American descriptions of species of *Cuphophyllus* from truly arctic-alpine habitats also ensures that *C. hygrocyboides* was not described on this continent before its 1977 description in Europe.

We have separated the new species *C. lamarum* from species in the *C. flavipes* complex (*C. flavipes* and an undescribed species in Europe and *C. pseudopallidus* from North America and Japan). Apart from the occasional pale yellow stipe of the smaller arctic-alpine species discussed above, there are no other species of *Cuphophyllus* with a yellow stipe described from Europe, although the epithet *cinereus* has been misapplied to such species on occasion (e.g., Helser and Smith 1963). Apart from *C. pseudopallidus*, Hesler and Smith (1963) described no other species of the current *Cuphophyllus* with a yellow stipe, and we are not aware of other such species descriptions from North America. Also, the lack of other species of *Cuphophyllus* from raised *Sphagnum* bogs confirms that *C. lamarum* has not been described before under another name.

Most North American species are not well known. The few descriptions available, other than the protologs, often conflict with the latter or seem otherwise misapplied. A full discussion of them requires a modern review including molecular studies of the *C. lacmus-subviolaceus* complex and other appropriate groups, something outside the scope of this study. Regarding *C. pseudopallidus* and *C. subviolaceus*, we thus key out these to a species complex. Where comparisons are made or these species used in the keys, they are interpreted from their respective protologs.

Our key to the gray, brown, purplish brown, and orange-brown *Cuphophyllus* species in the Northern Hemisphere excludes *C. radiatus*, as it is a taxonomic synonym of *C. flavipes* (Boertmann 2010). We also exclude treatment of *Hygrocybe cinerea* (Pers.) P.D. Orton & Watling, *C. berkeleyi* (P.D. Orton & Watling) Bon, and *Hygrophorus pallidus* Peck because they are not well-circumscribed species (Hesler and Smith 1963; Boertmann 2010).

We resolve the relationship of *C. subviolaceus* as a sister species to the putative *C. lacmus* by generating ITS sequence data from the type specimen of *C. subviolaceus* and thereby fix the name. The two species have been regarded as conspecific by many authors (e.g., Boertmann 2010), but we show that there are two species and that *C. subviolaceus* is phylogenetically distinct from the putative *C. lacmus*. Based on FIG. 2, we can see that both *C. subviolaceus* and the putative *C. lacmus* have an intercontinental distribution range. Because of the morphological similarities and confusion in naming, a more extensive sampling is needed to better characterize the species in this complex; however, this was not a target for this study.

Before our report, there were no public DNA deposits of either *C. cinerellus* or *C. hygrocyboides* available. Although our study contributes to the knowledge of these arctic-alpine species, it constitutes a preliminary report only. Because these regions are undersurveyed in North America, knowledge of the distribution of the species on this continent remains poor. We hope that this report may provide both background and stimulus for other workers to pursue the mycota of our vast northern regions. Tangentially, we also note that sequences taken from GenBank to construct our tree and our unpublished data revealed that likely other epithets also harbor cryptic species. Although pursuing these species is beyond the scope of the present study, our preliminary enquiries suggest that this is a fruitful area for future study.

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LITERATURE CITED

Abarenkov K, Tedersoo L, Nilsson HR, Vellak K, Saar I, Veldre V, Parmasto E, Prous M, Aan A, Ots M, Kurina O, Ostonen I, Jõgeva J, Halapuu S, Põldmaa K, Toots M, Truu J, Larsson K-H, Kõljalg U. 2010. PlutoF – a web based workbench for ecological and taxonomical research, with an online implementation for fungal ITS sequences. Evolutionary Bioinformatics 6:189–196.

Bas C, Arnolds EJM. 2007. *Hygrocybe comosa*, a remarkable new agaric from the French Jura. Persoonia 19:251–254.

Boertmann D. 2010. The genus Hygrocybe. 2nd revised edition. Fungi of Northern Europe Vol. 1. Copenhagen, Denmark: Svampetryk. 200 p.

Bon M. 1985 (1984). Le genre *Cuphophyllus* (Donk) sp. nov. Documents Mycologiques 14:9–12.

Borgen T, Arnolds E. 2004. Taxonomy, ecology and distribution of of *Hygrocybe* (Fr.) P. Kumm. and *Camarophyllopsis* Henrik (Fungi, Basidiomycota,Hygrocybeae) in Greenland. Meddelelser om Grønland Bioscience 54. Copenhagen, Denmark: Danish Polar Center. 64 p.

Borgen T, Boertmann D. 2008 New records of *Hygrocybe hygrocyboides* (Kühner) Arnolds (Fungi, Basidiomycota, Hygrocybeae). Sommerfeltia 31:17–27.

Buyck B, Kauff F, Eyssartier G, Couloux A, Hofstetter V. 2014. A multilocus phylogeny for worldwide *Cantharellus* (Cantharellales, Agaricomycetidae). Fungal Diversity 64:101–121.

Clark K, Karsch-Mizrachi I, Lipman DJ, Ostell J, Sayers E. 2016. Nucleic Acids Research 44(database issue): D67–D72.

Drummond AJ, Suchard MA, Xie D, Rambaut A. 2012. Bayesian Phylogenetics with BEAUti and the BEAST 1.7. Molecular Biology and Evolution 29:1969–1973.

Hesler LR, Smith AH. 1963. North American species of *Hygrophorus*. Knoxville, Tennessee: University of Tennessee Press. 416 p.

Halbwachs H, Easton G, Bol R, Hobbie E, Garnett M, Persoh D, Dixon L, Ostle N, Karasch P, Griffith G. 2018. Isotopic evidence of biotrophy and unusual nitrogen nutrition in soil-dwelling Hygrophoraceae: Hygrophoraceae ^{13}C/^{15}N natural abundance. Environmental Microbiology 20:3573–3588.

Katoh K, Standley DM. 2013. MAFFT: multiple sequence alignment software version 7: improvements in performance and usability. Molecular Biology and Evolution 30:772–780.

Kõljalg U, Nilsson RH, Abarenkov K, Tedersoo L, Taylor AFS, Bahram M, Bates SB, Bruns TD, Bengtsson-Palme J, Callaghan TM, Douglas B, Drenkhan T, Eberhardt U, Dueñas M, Griffith TGW, Hartmann M, Kirk PM, Kohout P, Larsson E, Lindahl BD, Lücking R, Martín RMP, Matheny PB, Nguen NH, Niskanen T, Oja J, Peay KG, Peintner U, Peterson M, Põldmaa K, Saag L, Saar I, Schüssler A, Scott JA, Senés C, Smith ME, Suija A, Taylor DL, Telleria T, Weiss M, Larsson K-H. 2013. Towards a unified paradigm for sequence-based identification of fungi. Molecular Ecology 22:5271–5277.

Kühner R. 1977. Agaricales de la zone alpine. Genre *Camarophyllus* (Fries) Kummer. Bulletin trimetrielle Societé Mycologique de France 93:121–144.

Larsson A. 2014. AliView: a fast and lightweight alignment viewer and editor for large data sets. Bioinformatics 22:3276–3278.

Larsson E, Vauras J, Cripps CL. 2018. *Inocybe praetervisa* group—a clade of four closely related species with partly different geographical distribution ranges in Europe. Mycoscience 59:277–287.

Liu YL, Whelen S, Hall BD. 1999. Phylogenetic relationships among ascomycetes: evidence from an RNA polymerase II subunit. Molecular Biology and Evolution 16:1799–1808.

Lodge DJ, Padamsee M, Matheny PB, Aime MC, Cantrell SA, Boertmann D, Kovalenko A, Vizzini A, Dentinger BTM, Kirk PM, Ainsworth AM, Moncalvo J-M, Vilgalys R, Larsson E, Lücking R, Griffith GW, Smith ME, Norvell LL, Desjardin DE, Redhead SA, Ovrebo CL, Lickey EB, Ercole E, Hughes KW, Courtecuisse R, Young A, Binder M, Minnis AM, Lindner DL, Ortiz-Santana B, Haight J, Læssøe T, Baroni TJ, Geml J, Hattori T. 2013. Molecular phylogeny, morphology, pigment chemistry and ecology in Hygrophoraceae (Agaricales). Fungal Diversity 64:1–99.

Matheny PB. 2005. Improving phylogenetic inference of mushrooms with RPB1 and RPB2 nucleotide sequence (*Inocybe*, Agaricales). Molecular Phylogenetics and Evolution 35:1–20.

Nylander JAA. 2004. MrModeltest v2. Program distributed by the author. Evolutionary Biology Centre, Uppsala University, Uppsala, Sweden.

Richard F, Sauvé M, Bellanger J-M, Clowez P, Hansen K, O'Donnell K, Urban A, Courtecuisse R, Moreau P-A. 2015. True morels (*Morchella*, Pezizales) of Europe and North America: evolutionary relationships inferred from multilocus data and a unified taxonomy. Mycologia 107:359–382.

Ronquist F, Heulsenbeck JP. 2012. MrBayes 3.2, efficient Bayesian phylogenetic inference and model choice across a large model space. Systematic Biology 61:539–542.

Saar I, Voitk A. 2015. Type studies of two *Tricholomopsis* species described by Peck. Mycological Progress 14:46.

Seitzman BH, Ouimette A, Mixon RL, Hobbie AE, Hibbett DS. 2011. Conservation of biotrophy in Hygrophoraceae inferred from combined stable isotope and phylogenetic analysis. Mycologia 103:280–290.

Swofford DL. 2003. PAUP*. Phylogenetic Analysis Using Parsimony (*and other methods). Version 4. Sunderland, Massachusetts: Sinauer Associates.

Tedersoo L, Jairus T, Horton BM, Abarenkov K, Suvi T, Saar I, Kõljalg U. 2008. Strong host preference of ectomycorrhizal fungi in a Tasmanian wet sclerophyll forest as revealed by DNA barcoding and taxon-specific primers. New Phytologist 180:479–490.

Tello SA, Silva-Flores P, Agerer R, Halbwachs H, Beck A, Persoh D. 2014. *Hygrocybe virginea* is a systemic endophyte of *Plantago lanceolata*. Mycological Progress 13:471–475.

White TJ, Bruns T, Lee L, Taylor JW. 1990. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: Innis MA, Gelfand DH, Sininski JJ, White TJ, eds. PCR protocols: a guide to methods and applications. New York: Academic Press. p. 315–322.

SUPPLEMENTAL TABLE 2. TABULAR KEY TO DESCRIBED AND SELECTED SIMILAR *CUPHOPHYLLUS* SPECIES

Species **Character**		**cinerellus**	**esteriae**	**lamarum**	**flavipes/ pseudopallidus**	**lacmus/ subviolaceus**	**hygrocyboides**	**colemannianus**
Cap	**diameter > 30 mm[1]**	rare	rare	often	often	often	never	common
	color	cinereous violet-gray	cinereous violet-gray	light grayish brown	cinereous violet-gray	cinereous violet-gray	bay to light brown	bay brown
Lamella edge (micro)		smooth	smooth	protruding hyphae	smooth	smooth	smooth	smooth
Stipe color		whitish light gray	base occasionally light yellow	yellowish, incr. toward base	yellow, incr. toward base	white to light grayish	brownish	white
Flesh staining[2]		none	none	orange[2]	none	none	none	none
Spore size (µm)	**range**	6.5–9.5 × 4.0–6.5	6.0–10.5 × 4.0–6.5	5.5–9.0 × 4.0–6.5	5.5–8.0 × 5.0–6.5[3]	6.0–8.5 × 4.0–6.5[4]	6.0–8.5 × 4–5[4]	6.0–10.0 × 4.0–7.0[4]
	average	8.0 × 5.4	7.5 × 5.3	7.0 × 5.2	6.8 × 5.8	not calculated	7.6 × 4.4[5]	not calculated
Basidium size (µm)		32–54 × 6.5–13.5	22–40 × 6.0–8.0	38–78 × 6.0–9.0	40–58 × 6.5–8.0	39–58 × 6.0–7.0[4]	50–72 × 5.5–7.0	49–60 × 7.0–8.0[4]
Habitat	**general**	arctic-alpine	arctic-alpine	boreal	boreal to alpine	boreal to alpine	arctic-alpine	boreal to arctic-alpine

	specific	tundra, bog, pioneer soil	tundra, bog, pioneer soil	bog	woodland, grassland	woodland, grassland	tundra, bog, pioneer soil	woodland, grassland
Photobiont associate[6]		bryophytes	bryophytes	*Sphagnum*	herbaceous, coniferous, ericaceous	herbaceous, coniferous, ericaceous	herbaceous bryophytes	herbaceous, ericaceous?
Fruiting time Newfoundland		n/a	Jul-Sep	Jul	n/a	Aug-Oct	n/a	Aug-Oct
Distribution		Eurasia	North America[7]	so far, only known from NL	Eurasia	Eurasia, North America	Fennoscandia, central Europe North America	Eurasia, North America

[1] majority of large (fully developed) caps

[2] experience limited to three collections; staining seen with only one

[3] epitype measurements

[4] quoted from Boertmann 2010 (including extremes)

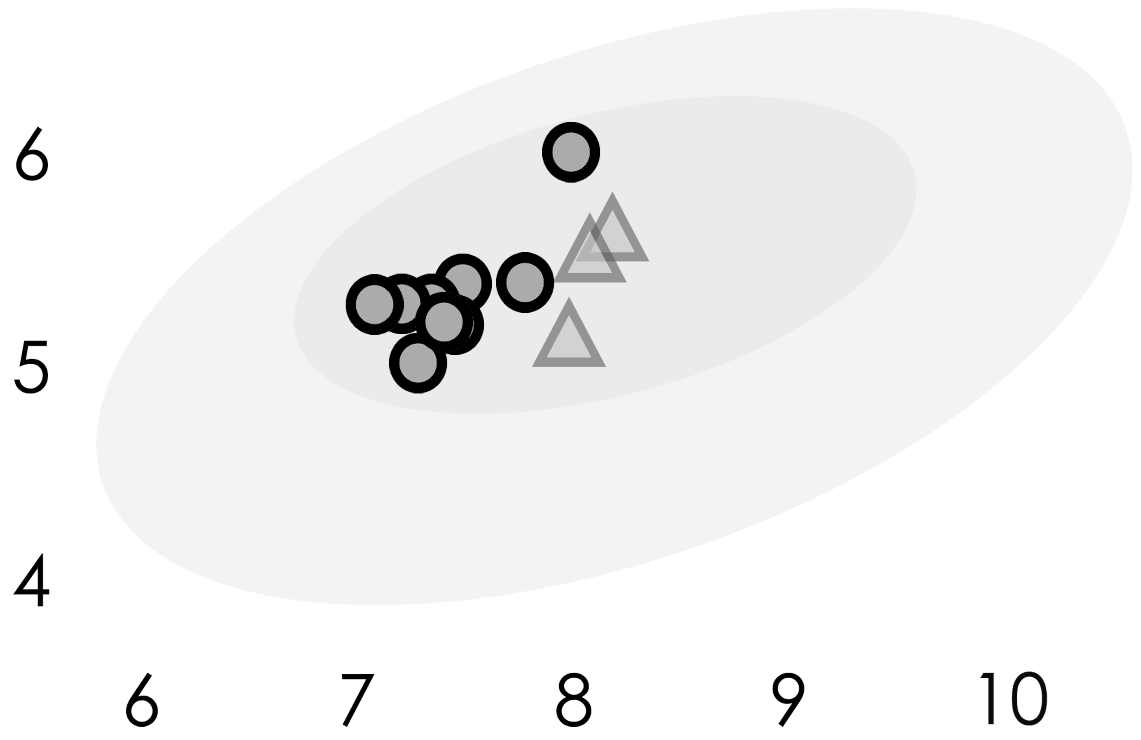
[5] Canadian collection values only

[6] assumed from adjacent plants

[7] so far only confirmed from eastern Newfoundlande

SUPPLEMENTAL TABLE 1. Information of specimens sequenced for this study

GB nr (UNITE nr) Collecting nr	UNITE nr	Collecting nr	DATE	Main Herbarium	Copy herbarium	NAME	COUNTRY/Prov	LOCATION	lat °N	long °E	elev (m asl)	COLLECTOR	HABITAT	SUBSTRATE	POTENTIAL PHOTOBIONTS	COMMENTS	PHOTO
MK547063	**UDB034648**	**LS-075 (lat6)**	**15-Aug-08**	**DAOM867478**	**TU117603**	***Cuphophyllus esteriae***	**CANADA, NL**	**L'Anse Amour, Labrador**	**51.4612**	**-56.8587**	**21**	**Maria Voitk**	**bog**	***Sphagnum***	**moss, *Sphagnum***	**HOLOTYPE**	**Y**
MK547064	UDB034650	BH-096 (BH1)	18-Aug-08	DAOM867479	TU117605	*Cuphophyllus esteriae*	CANADA, NL	Battle Island, Labrador	52.2760	-55.5844	32	Andrus Voitk	heath, peaty pond edge	peat			Y
MK547065	UDB034651	BH-097 (CE21)	21-Aug-08	DAOM867480	TU117606	*Cuphophyllus esteriae*	CANADA, NL	Great Caribou Island, Labrador	52.2733	-55.5911	38	Michael Burzynski	heath, barrens	*Sphagnum*	*Sphagnum*		Y
MK547080	UDB038329	CEAV-02	20-Aug-18	DAOM867481	TU117662	*Cuphophyllus esteriae*	CANADA, NL	Great Caribou Island, Labrador	52.2733	-55.5911	38	Anne Marceau	heath, barrens	pioneer soil			Y
MK547078	UDB038327	CEMV-03	20-Aug-18	DAOM867482	TU117660	*Cuphophyllus esteriae*	CANADA, NL	Great Caribou Island, Labrador	52.2733	-55.5911	38	Maria Voitk	bog, heath, barrens	*Sphagnum*	*Sphagnum*		Y
MK547079	UDB038328	TCAV-01	21-Aug-18	DAOM867483	TU117661	*Cuphophyllus esteriae*	CANADA, NL	Great Caribou Island, Labrador	52.2672	-55.6298	43	Andrus Voitk	heath, barrens	moss	moss		Y
MK547081	UDB038330	TCAV-04	21-Aug-18	DAOM867484	TU117664	*Cuphophyllus esteriae*	CANADA, NL	Great Caribou Island, Labrador	52.2672	-55.6298	43	Andrus Voitk	heath, barrens	pioneer soil	*Rhacomitrium lanuginosum*		Y
MK547082	UDB038331	TCAV-09	21-Aug-18	DAOM867485	TU117665 C-F-121572	*Cuphophyllus esteriae*	CANADA, NL	Great Caribou Island, Labrador	52.2672	-55.6298	43	Andrus Voitk	bog	*Sphagnum*	*Sphagnum*	yellowish base stem	Y
MK547075	UDB034982	TB95-071	13-Aug-95	C-F-121549		*Cuphophyllus esteriae*	Greenland	Paamiut, 2 km E of town				Torbjørn Borgen	Low arctic				
MK547062	**UDB034246**	**10.07.13.av01**	**2010-07-13**	**DAOM867486**	**TU117564** **C-F-121518**	***Cuphophyllus lamarum***	**CANADA, NL**	**Transcanada Highway, Stephenville high bog**	**48.6686**	**-58.2061**	**199**	**Andrus Voitk**	**bog**	***Sphagnum***	***Sphagnum***	**HOLOTYPE / SYNTYPE**	**Y**
MK547061	UDB034244	10.07.28.av01	2010-07-28	DAOM867487	TU117562	*Cuphophyllus lamarum*	CANADA, NL	Howley bogs	49.2349	-57.0721	94	Maria Voitk	bog	*Sphagnum*	*Sphagnum*	orange staining	Y
MK547083	UDB038332	18.07.20.av01	2018-07-20	DAOM867488	TU117667	*Cuphophyllus lamarum*	CANADA, NL	Howley bogs	49.2349	-57.0721	94	Tom Booth	bog	*Sphagnum*	*Sphagnum*		Y
MK547076	UDB035646	S/250999-1		TU117644		*Cuphophyllus flavipes*	GERMANY, OBERBAYERN	Augsburg bei Ammersee, near Edmühle				Peter Karasch					
MN453872		Campo131027		TURA199692		*Cuphophyllus flavipes*	Italy	Arba, Pordenone				E. Campo	grassland	Among grass and herbs			
MN453873		Campo131010		TURA199694		*Cuphophyllus flavipes*	Italy	Longarone, Belluno				E. Campo	grassland	Among grass and herbs			
MK573930		RGC14-104	14-09-28	GB-0156991		*Cuphophyllus flavipes*	Sweden	Småland, Torsås	56.2453	15.5939		R-G Carlsson	grassland	Among grass and herbs			
MK573932		RGC06-125	06-09-04	GB-0152637		*Cuphophyllus flavipes*	Sweden	Västergötland, Skövde	58.2449	13.5026		R-G Carlsson	grazed meadow	Among grass and herbs			
MK573931		RGC97-097	97-08-08	GB-0152595		*Cuphophyllus flavipes*	Sweden	Västergötland, Tidaholm	58.1338	13.4818		R-G Carlsson	grassland	Among grass and herbs			
MK547077	**UDB035647**	**G/122-00**		**TU117645**		***Cuphophyllus flavipes***	**GERMANY, OBERBAYERN**	**Augsburg bei Ammersee, Goassl-Weide, Hartschimmel-Hof**			**735**	**Peter Karasch**				**EPITYPE**	**Y**
MK547068	UDB03973	JV93-829	93-09-04	C-F-20429		*Cuphophyllus flavipes*	Denmark	Western Jylland, Tågelund W of Egtved				Jan Vesterholt	grassland				
MK547066	UDB03971	JV85-747	85-08-30	C-F-107960		*Cuphophyllus flavipes*	Denmark	Jylland, Moesgård Strand				Jan Vesterholt					Y
MK547070	UDB034975	GG2679	00-07-10	O223054		*Cuphophyllus cinerellus*	Norway	Oppland, Gran, Midttjernet			480	Geir Gaarder	Intermediate mire, by a little pond				
MK547071	UDB034976		02-05-15	O242127		*Cuphophyllus cinerellus*	Norway	Møre og Romsdal, Sunndal, Øksendal, Gaudalen			750	John Bjarne Jordal	Alpine moist heath		*Dryas*		
MK547069	UDB034974		99-07-18	O288058		*Cuphophyllus cinerellus*	Norway	Møre og Romsdal, Sunndal, Skrødalene			560	John Bjarne Jordal	Poor mire		*Carex nigra, Eriophorum angustifolium, Carex echinata*		
MK547072	UDB034978		09-08-13	O291322		*Cuphophyllus cinerellus*	Norway	Sør-Trøndelag , Oppdal, Grytdalen			700	John Bjarne Jordal	Ombrotrophic mire		*Sphagnum fuscum, Rubus chamaemorus*		
MK547073	UDB034979	GG6774	13-07-24	O246174		*Cuphophyllus cinerellus*	Norway	Nordland, Hemnes, Stormyra				Geir Gaarder	Mire				
MK547074	UDB034980		13-08-10	O249863		*Cuphophyllus cinerellus*	Norway	Oppland, Lom, Dumdalen				John Bjarne Jordal			*Empetrum*, *Dryas*		
MK573933		EL170-17	17-08-19	GB-0156994		*Cuphophyllus cinerellus*	Norway	Troms, Storfjord, Between Bossoyarri and Sallooaivi	69.025	20.475	480	Ellen Larsson	Alpine heath		*Empetrum nigrum, Vaccinium uliginosum, Betula nana* and *Arctostaphylos*		Y
MK573934		EL118-15	15-08-19	GB-0156993		*Cuphophyllus cinerellus*	Sweden	Lycksele lappmark. Tärna, Laisaliden	65.485	15.0605	670	Ellen Larsson	Alpine heath	*Sphagnum* sp.	*Empetrum nigrum, Carex nigra, Betula nana and Salix spp*		
MK573935		**EL30-16**	**16-08-11**	**GB-0156961**		*Cuphophyllus cinerellus*	**Sweden**	**Lule lappmark, Jokkmokk, Padjelanta, Arranoaijvve**	**67.2222**	**16.4815**	**800**	**Ellen Larsson**	**Alpine heath**		***Empetrum nigrum; Betula nana* and *Salix* spp.**	**EPITYPE**	**Y**
MK836089		TL-4692	97-08-04	C-F-47028		*Cuphophyllus cinerellus*	Russia	Kamchatka, Esso, Koryzrevsky range,Tupikin Klyuh	55.9667	158.783	675	Thomas Læssøe	turf-like soil				Y
MN394148		Berch0283	16-06-26	DAVFP		*Cuphophyllus hygrocyboides*	Canada, BC	Ner Smithers, Montase Peak in Sicintine Range	56.1464	-127.209	1805	Shannon Berch	Alpine heath	moss	*Salix stolonifera*		
MK573937		EL177-13	13-08-22	GB-0156992		*Cuphophyllus hygrocyboides*	Sweden	Torne lappmark, Jukkasjärvi, Abisko	68.3589	18.4836	950	Ellen Larsson	Alpine heath on calacreous soil		*Salix herbacea, S. reticulata, Dryas*, grasses and herbs		
MK573936		TB02-152	02-08-31	C-F-121571		*Cuphophyllus hygrocyboides*	Greenland	Narsarsuaq, Kiattut Qaqqaat	61.21N	45.37W	150	Torbjørn Borgen	Dry grassland slope	*Alchemilla alpina*	Grasses, *Alchemilla alpina*		Y
MK573938		**K69-319**	**69-09-14**	**G-00052005**		*Cuphophyllus hygrocyboides*	**France**	**Savoie, Parc nat. De la Vanoise. Région de Pralognan**	**45.2542**	**6.2417**		**R. Kühner**				**HOLOTYPE**	
MN322657				**NYSf3099**		**Cuphophyllus subviolaceus**	**USA, NY**					**Peck**				**HOLOTYPE**	
MK547067				C-F-17644		*Cuphophyllus lacmus*	Denmark										



Supplemental Figure 1. Comparison of spore measurements of sequence-identified specimens of *C. cinerellus* (range in smaller inner oval and average measurements represented by triangles) and *C. esteriae* (range in larger outer oval and average measurements represented by circles). Length on x-axis and width on y-axis are measured in µm. Note that the complete overlap of one species by the other make measurements unhelpful for identification, but that average measurements from individual specimens tend to cluster somewhat apart. Difference in spore size was statistically highly significant ($P = 0.0001$).