

Studies in the genus *Glutinoglossum*

Vincent P. Hustad¹

*Department of Plant Biology, University of Illinois at
Urbana-Champaign, 505 S. Goodwin Avenue, Urbana,
Illinois 61801*

Andrew N. Miller

*Illinois Natural History Survey, University of Illinois at
Urbana-Champaign, 1816 S. Oak Street, Champaign,
Illinois 61820*

Abstract: *Glutinoglossum* is a recently described genus in Geoglossomycetes, characterized by viscid black fruiting bodies. *Glutinoglossum* was described based on molecular evidence showing that the fungus formerly known as *Geoglossum glutinosum* formed a well-supported clade separate from *Geoglossum*. This study provides a comprehensive examination of *Glutinoglossum* based on maximum-likelihood and Bayesian analyses of a dataset that includes nuc rDNA internal transcribed spacer and partial 28S regions and the nuc genes for RNA polymerase II largest subunit (*RPB1*) and minichromosome maintenance complex component 7 (*MCM7*). Four new species are described, an emended description of *Glutinoglossum glutinosum* and a key to the genus *Glutinoglossum* is provided.

Key words: Ascomycota, earth tongues, Geoglossomycetes, *Geoglossum*, new species, systematics

INTRODUCTION

Geoglossomycetes, commonly referred to as earth tongues, is a class in the Ascomycota presently comprising one family (Geoglossaceae) and eight genera: *Geoglossum* Pers., *Glutinoglossum* Hustad, A.N. Mill., Dentinger & P.F. Cannon, *Hemileucoglossum* Arauzo, *Leucoglossum* S. Imai, *Nothomitra* Maas Geest., *Sabuloglossum* Hustad, A.N. Mill., Dentinger & P.F. Cannon, *Sarcoleotia* S. Ito & S. Imai and *Trichoglossum* Boud. (Hustad et al. 2013, Arauzo and Iglesias 2014). *Glutinoglossum* is a recently described genus of earth tongue fungi in the class Geoglossomycetes (Ascomycota). *Glutinoglossum* presently includes two species: *G. glutinosum* (Pers.) Hustad, A.N. Mill., Dentinger & P.F. Cannon and *G. heptaseptatum* Hustad, A.N. Mill., Dentinger & P.F. Cannon (Hustad et al. 2013). Fungi of the genus *Glutinoglossum* are characterized by the production of viscid, black, clavate ascocarps with a distinct gelatinous layer of elongated, hyaline,

sparsely septate paraphyses covering most of the surface of the ascocarp, and slowly maturing ascospores that are initially hyaline and lack septation, but later develop septa and become brown. *Glutinoglossum glutinosum*, commonly known as the glutinous earth tongue, is among the first species of Geoglossomycetes reported in mycological literature and has been subjected to a number of nomenclatural changes since the fungus was described by C.H. Persoon in 1796 as *Geoglossum glutinosum* Pers.

Taxonomic confusion has surrounded *Geoglossum glutinosum* for nearly as long as the species has been studied by mycologists. After his description of *G. glutinosum* in 1794, Persoon described *G. viscosum* Pers. in 1797 and *G. glutinosum* var. *lubricum* Pers. in 1822. These species descriptions are ambiguous, as expected in the years pre-dating contemporary microscopy. *G. viscosum* was described as a shorter form of *G. glutinosum*, and *G. glutinosum* var. *lubricum* was distinguished from *G. glutinosum* in being entirely covered with a viscid gelatinous layer. Both species later were examined and reduced to synonyms of *G. glutinosum* by Durand (1908).

Hazslinszky (1881) described *Cibalocoryne* as a subgenus of *Geoglossum* and included the species *Cibalocoryne viscosa* Hazsl. in his protolog. Because *Cibalocoryne* was not a validly published genus, this species was transferred by Saccardo (1884) to *Geoglossum viscosulum* (Hazsl.) Sacc. Imai (1942) continued to treat *Cibalocoryne* as a separate genus within the Geoglossaceae and later transferred *G. glutinosum* to *Cibalocoryne glutinosa* (Pers.) Imai. Hazslinszky's species later was synonymized under *Geoglossum glutinosum* by Nannfeldt (1942), who also stated that the "genus *Cibalocoryne* lacks thus every raison d'être". This sentiment was reinforced by Mains (1954) and Maas Geesteranus (1965) who found *Cibalocoryne* to be a *nomen ambiguum* and subordinate to *Geoglossum*.

Durand (1908) described the genus *Gloeoglossum* E.J. Durand to contain species of *Geoglossum* with "viscid-gelatinous consistency when fresh" and with paraphyses that are "not confined to the hymenium but continue with unchanged form down the stem to its base". Durand transferred all of the known viscid species of *Geoglossum* to this new genus, including *G. difforme* (Fr.) E.J. Durand (the type species of the genus), *G. affine* E.J. Durand and *G. glutinosum* (Pers.) E.J. Durand. Nannfeldt (1942) reduced *Gloeoglossum* to synonymy and transferred the three species back to *Geoglossum*, a transfer that also was supported by Mains (1954).

Species of *Glutinoglossum* are among the rarer earth tongues in North America, with specimens labeled *Geoglossum glutinosum* accounting for fewer than 5% of all Geoglossomycetes specimens in the 39-member herbaria of MycoPortal (<http://mycoportal.org/portal/index.php>) and 34/887 (3.8%) at MICH, which holds the largest collection of Geoglossomycetes in North America. While Geoglossomycetes are infrequently collected in New Zealand, 41/258 (16%) in PDD were identified as *Geoglossum glutinosum* or *Glutinoglossum*, suggesting a higher diversity of glutinous earth tongues might be present in New Zealand.

Further collecting and analyses of *Glutinoglossum* collections since Hustad et al. (2013) has resulted in the discovery of four previously undescribed cryptic species of *Glutinoglossum* from North America and Australasia. This paper describes these new species along with an emended description of *Glutinoglossum glutinosum* and provides an overview of the genus *Glutinoglossum*.

MATERIALS AND METHODS

Morphological analyses.—Samples were obtained from freshly collected material or herbarium specimens. Mature discharged ascospores were obtained by tapping fresh or dried ascomata in a drop of water on a slide (Mains 1954). Dehydrated ascomata were hand-sectioned and a small amount of hymenium was squash-mounted in 5% KOH for observation of micro-morphological characters. Images of pertinent micro-morphological characters were captured with a QImaging QColor 3 digital camera mounted on an Olympus BX51 compound microscope with differential interference microscopy. Images were processed with Adobe Photoshop 7.0 (Adobe Systems Inc., Mountain View, California). A minimum of 30 measurements were taken when possible for each micro-morphological character using NIH Image 1.63 (National Institutes of Health, Bethesda, Maryland). Mean and standard deviation of ascus and ascospore size were recorded. Taxonomic novelties and associated data were registered in MycoBank (Crous et al. 2004).

Molecular procedures.—Total genomic DNA was extracted from approximately 0.5 cm² hymenium tissue from a single dried ascoma using a QIAGEN DNeasy Plant Mini Kit (QIAGEN Inc., Valencia, California). Gene fragments were PCR-amplified following the methods outlined in Matheny et al. (2002), Promputtha and Miller (2010) and Raja et al. (2011), and purified using a Promega Wizard SV Gel and PCR Clean-Up System (Promega Corp., Fitchburg, Wisconsin). Sequences were generated on an ABI Applied Biosystems 3730XL high-throughput DNA capillary sequencer at the UIUC Keck Center for Comparative and Functional Genomics. Two regions of nuc rDNA were amplified and sequenced: the ca. 570 bp internal transcribed spacer (ITS) region, consisting of the ITS1, 5.8S and ITS2 regions, was amplified and sequenced using a combination of the primers ITS5, ITS1 and ITS4 (White et al.

1990); a ca. 630 bp fragment of the 5' end of the 28S rRNA coding sequence (28S) region containing the D1-D2 domains was amplified and sequenced with primers JS1 (Landvik 1996) and LR3 (Vilgalys and Hester 1990).

Two single-copy protein-coding genes also were used in addition to the ITS and 28S rDNA regions to further elucidate evolutionary relationships. The minichromosome maintenance complex component 7 gene (*MCM7*) is a single-copy gene that codes for a DNA replication licensing factor required for eukaryotic genome replication initiation and cell proliferation (Moir et al. 1982, Kearsey and Labib 1998). *MCM7* has been shown to produce highly accurate phylogenies in fungi (Aguileta et al. 2008, Schmitt et al. 2009) across a wide range of ascomycete taxa including Geoglossomycetes (Raja et al. 2011, Hustad et al. 2013). The primers 709F and 1348R (Schmitt et al. 2009) were used for PCR amplification and sequencing of the 642 bp segment of *MCM7*.

The second single-copy protein-coding gene used in these analyses was the RNA polymerase II subunit 1 gene (*RPB1*). Schoch et al. (2009) found *RPB1* to have the highest per-site informativeness (Townsend 2007) across six genes in the Ascomycota. *RPB1* analysis has been shown recently to provide reliable phylogenies within Geoglossomycetes (Hustad et al. 2013). The primers RPB1af (Stiller et al. 1997) and RPB1cr (Matheny et al. 2002) were used for amplification and sequencing of an 849 bp region of the *RPB1* gene.

Phylogenetic analyses.—Individual gene alignments were created by eye in Sequencher 5.0.1 (Gene Codes Corp., Ann Arbor, Michigan) and optimized if necessary with Muscle 3.8.31 (Edgar 2004) in Seaview 4.4.2 (Gouy et al. 2010). Ambiguous regions were removed from individual gene datasets with Gblocks 0.91b (Castresana 2000) under the following parameters: for ITS minimum number of sequences for both conserved and flanking regions = 13, maximum number of contiguous, nonconserved regions = 4, minimum length of a block = 2, and allowed gap positions in 50% of sequences; for 28S minimum number of sequences for both conserved and flanking regions = 11, maximum number of contiguous, nonconserved regions = 2, minimum length of a block = 2 and allowed gap positions in 50% of sequences.

The GTR+I+G was determined as the best-fit model of evolution for all four genes based on the Akaike information criterion (AIC) (Posada and Buckley 2004) utilized by jModelTest 2.1.6 (Guindon and Gascuel 2003, Darriba et al. 2012) on XSEDE platform of the CIPRES Science Gateway Teragrid (Miller et al. 2010). Maximum-likelihood (ML) analyses were performed on all four individual gene alignments with PhyML 3.0 (Guindon et al. 2010) on the ATGC server (<http://www.atgc-montpellier.fr/phyml/>) under the GTR+I+G model with six rate classes and invariable sites optimized. The best of subtree pruning and regrafting (SPR) and nearest neighbor interchange (NNI) was implemented during the heuristic search. Nonparametric bootstrap support (Felsenstein 1985) (BS) was determined with 1000 replicates. Clades with BS values $\geq 70\%$ were considered significant (Hillis and Bull 1993).

Bayesian inference employing a Markov chain Monte Carlo (MCMC) algorithm was performed with MrBayes 3.2.2

(Ronquist et al. 2012) on XSEDE. Four independent chains of MCMC were run 10 million generations with trees sampled every 1000 generations. Clades with Bayesian posterior probability (BPP) values of $\geq 95\%$ were considered significant (Alfaro et al. 2003). Tracer 1.5 (Rambaut and Drummond 2009) was used to estimate effective sample size (ESS) (i.e. burn-in) using the standard deviation of split frequencies produced by Bayesian analysis.

Individual datasets of ITS, 28S, *MCM7* and *RPB1* were examined for potential conflict before concatenation into a single dataset for total evidence analysis (Kluge 1989, Eernisse and Kluge 1993). Individual gene phylogenies were considered conflicting if clades with significant ML BS and BPP were conflicting in the individual tree topologies (Wiens 1998, Alfaro et al. 2003, Lutzoni et al. 2004). Because there were no significant conflicting clades among the individual gene trees, all genes were concatenated for final analyses in this order: ITS, 28S, *MCM7*, *RPB1*. Phylogenetic analyses then were performed on the concatenated dataset using the same methods used in the individual datasets. Alignments and trees are deposited in TreeBASE (<http://treebase.org>) under submission ID 16987.

RESULTS

Forty-five sequences, consisting of 12 ITS, 12 28S, 11 *MCM7* and 11 *RPB1*, were generated in this study (TABLE I). These were analyzed together with 7 ITS, 7 28S, 6 *MCM7* and 6 *RPB1* sequences from Hustad et al. (2011, 2013) and sequences of *Glutinoglossum* from Wang et al. (2005, 2011), Brock et al. (2009) along with one environmental ITS sequence suggested to be closely related to *Glutinoglossum* by a GenBank BLAST query (DQ421173). Twenty-four specimens, representing a total of six *Glutinoglossum* species, three outgroup species and one environmental sequence, were included in the analyses. Of the 24 specimens included in the final dataset, missing data were ITS for one taxon, 28S for four taxa, *MCM7* for seven taxa and *RPB1* for seven taxa (TABLE I). In the combined dataset, sequences for all four markers were available for 61% of the specimens and sequences of at least two genes were available for 83% of specimens.

The final combined data matrix had an aligned length of 3833 bp, which was reduced to 2681 bp after the removal of 1152 bp of ambiguously aligned or gap regions by Gblocks. Of the 2681 characters used in the final phylogenetic analyses, 460 were constant and 2221 were variable. A burn-in of 10% was estimated with Tracer 1.5 to be sufficient to remove the pre-stationary posterior probability distribution, resulting in an ESS value of 3537.858. The standard deviation of split frequencies was -9455.358 at the end of the four-gene Bayesian analysis. The most likely tree

produced by PhyML of the final combined dataset is illustrated (FIG. 1).

The combined data analyses suggest a total of six distinct species within the genus *Glutinoglossum*, four of which have not been previously described. Below we present a description of the genus *Glutinoglossum* and descriptions for all six species.

TAXONOMY

Glutinoglossum Hustad, A.N. Mill., Dentinger & P.F. Cannon

Mycobank MB801343

Type species: Glutinoglossum glutinosum (Pers.) Hustad, A.N. Mill., Dentinger & P.F. Cannon

Ascocarps single to scattered to gregarious, occasionally subcaespitose, 1–5 cm tall, often entirely viscid-gelatinous, usually entirely black, stipitate, with black fertile hymenium usually restricted to the upper third to upper half of the ascocarp. Asci clavate to cylindrical, narrow and long with ascospores variously clustering in the apical portions of the ascus or throughout, J+ apical pore. Ascospores cylindrical to clavate, often slightly curved, slow-maturing, initially hyaline and aseptate, becoming variously septate and tan to brown with maturity. Paraphyses prominent, mostly straight, hyaline below becoming pale brown toward the apex, apical cells variously enlarged and sometimes embedded in a distinct hyaline to light brown gelatinous layer.

Glutinoglossum americanum Hustad & A.N. Mill., sp. nov. FIG. 2

Mycobank MB811410

Typification: USA. TENNESSEE: Sevier County, Great Smoky Mountains National Park, 5 miles east of Gatlinburg, Greenbrier, trail to Whaley Cemetery, 35°42'27.2"N, 83°22'54.1"W, 549 m, 15 Aug 2009, ANM 2231 (**holotype** ILLS 67352).

Etymology: Native to the Americas.

Ascocarps single, scattered or subcaespitose, cylindrical-clavate, 1.5–4.5 cm tall, viscid, becoming gelatinous when wet; hymenium black, one-third to one-half the length of the ascocarp, cylindrical-clavate to ellipsoidal, 2–6 mm wide at widest point; stipe cylindrical-terete, distinctly viscid; slightly paler than the hymenium. Asci clavate, (170–)182–220(–235) × (12–)12.5–14.5(–15.5) μm ($201.2 \pm 18.9 \times 13.6 \pm 0.9 \mu\text{m}$) in rehydrated material, eight-spored, with mature ascospores often clustering toward the apex, leaving the bottom 25–45% of the ascus base hyaline (FIG. 2A). Ascospores fusiform-clavate, (56.5–)61.5–69.5(–75) × (3.5–)4–4.5(–5) μm ($65.6 \pm 3.8 \times 4 \pm 0.4 \mu\text{m}$), initially aseptate and hyaline, developing septations in

TABLE I. Species, collection/strain numbers, fungarium accession numbers and GenBank sequence accession numbers of the specimens used in this study

Species	Collection/strain	Fungarium	ITS	LSU	MCM7	RPB1
<i>Geoglossum difforme</i>	ANM 2169	ILLS 61036	JQ256418	JN673044	JN672990	KC222162
<i>Glutinoglossum americanum</i>	ANM 2231	ILLS 67352	KC222128	KC222141	KC222153	KC222170
<i>Glutinoglossum americanum</i>	ANM 2247	ILLS 64444	KP690086	KP690098	KP690110	KP690121
<i>Glutinoglossum australasicum</i>	VPH NZ13-57	PDD 103619	KP690087	KP690099	KP690111	KP690122
<i>Glutinoglossum australasicum</i>	VPH NZ13-61	PDD 103623	KP690088	KP690100	KP690112	KP690123
<i>Glutinoglossum australasicum</i>	s.n.	PDD 73996	HQ222869	N/A	N/A	N/A
<i>Glutinoglossum exiguum</i>	VPH NZ13-10	PDD 103574	KP690089	KP690101	KP690113	KP690124
<i>Glutinoglossum exiguum</i>	VPH NZ13-49	PDD 103611	KP690090	KP690102	N/A	N/A
<i>Glutinoglossum glutinosum</i>	J. Gaisler s.n.	ILLS 67353	KC222129	KC222142	N/A	KC222171
<i>Glutinoglossum glutinosum</i>	ANM 2498	ILLS 72217	KP690091	KP690103	KP690114	KP690125
<i>Glutinoglossum glutinosum</i>	ANM 2476	ILLS 64446	KP690092	KP690104	KP690115	KP690126
<i>Glutinoglossum glutinosum</i>	ANM 2456	ILLS 64445	KP690093	KP690105	KP690116	KP690127
<i>Glutinoglossum glutinosum</i>	ANM 2529	ILLS 64448	KP690094	KP690106	KP690117	KP690128
<i>Glutinoglossum glutinosum</i>	s.n.	HMAS 72096	HQ222870	N/A	N/A	N/A
<i>Glutinoglossum glutinosum</i>	1100649	N/A	N/A	AY789310	N/A	N/A
<i>Glutinoglossum glutinosum</i>	s.n.	K(M): 135145	EU784443	N/A	N/A	N/A
<i>Glutinoglossum heptaseptatum</i>	J. Gaisler s.n.	ILLS 63754	KC222130	KC222143	KC222155	KC222172
<i>Glutinoglossum heptaseptatum</i>	J. Gaisler s.n.	K(M): 165359	KC222131	KC222144	KC222156	N/A
<i>Glutinoglossum methvenii</i>	VPH NZ13-33	PDD 103597	KP690095	KP690107	KP690118	KP690129
<i>Glutinoglossum methvenii</i>	VPH NZ13-67	PDD 103629	KP690096	KP690108	KP690119	KP690130
<i>Glutinoglossum methvenii</i>	VPH NZ13-42	PDD 103604	KP690097	KP690109	KP690120	KP690131
<i>Sabuloglossum arenarium</i>	CFR 181007	ILLS 61043	JQ256426	JQ256440	JQ256452	KC222175
<i>Trichoglossum hirsutum</i>	J. Gaisler s.n.	ILLS 61045	JQ256428	JQ256442	JQ256454	KC222178
Env. Sequence – MN (USA)	53-35	N/A	DQ421173	N/A	N/A	N/A

a one-, three-, seven-pattern, predominantly seven-septate and brown at maturity (FIG. 2B), although mature discharged spores with fewer septations have been observed. Paraphyses hyaline below, becoming dark brown at apex, tips pyriform to capitate, apical cells inflated (3.5–7.5 µm diam in rehydrated material), often embedded in a thin layer of brown, amorphous matter (FIG. 2C).

Distribution and habitat: NORTH AMERICA: USA (Mains 1954, this paper). On soil usually among moss (often *Thuidium*) in mixed deciduous forest.

Additional specimens examined: USA. MICHIGAN: Alger County, Rock River. Aug 1929, *AHS* s.n. (MICH s.n.); Luce County, Newberry. On moss, 28 Aug 1923, *EBM 33–256* (MICH s.n.); Mackinac County, Hardwoods North of Hessel. 15 Aug 1949, *H Imshaug 3469* (MICH s.n.); NORTH CAROLINA: Haywood County, Great Smoky Mountains National Park, Cataloochee, Caldwell Fork Trail, 35°37'N, 83°6'W, 762 m. On soil in mixed deciduous forest, 14 Aug 2009, *ANM 2177* (ILLS 64360); Swain County, Great Smoky Mountains National Park, Smokemont. On soil in mixed deciduous forest, 35°33'N, 83°18'W, 640 m. 16 Aug 2009, *ANM 2247* (ILLS 64444).

Comments: This species may be confused with *Geoglossum affine* Durand, a species with similar distribution, viscid ascocarps and ascospore sizes. However the paraphyses of *G. affine* are closely septate at the apex and the individual apicular cells

are variously enlarged unlike in *G. americanum*. Moreover, we have examined voucher material of *G. affine* and find it should remain in the genus *Geoglossum* based on morphological and molecular data (not shown). While we have only seen collections of *G. americanum* from three states in eastern North America, this species might be widely distributed throughout the continent.

Glutinoglossum australasicum Hustad & A.N. Mill. sp. nov. Fig. 3

Mycobank MB811411

Typification: NEW ZEALAND. SOUTH CANTERBURY: Peel Forest, Fern Walk, 29 May 2013, *VPH NZ13–61* (**holotype** PDD 103623; **isotype** ILLS 72403).

Etymology: Native to Australasia.

Ascocarps single to scattered, cylindrical-clavate, viscid (although sometimes not distinctly), 2.5–5 cm tall; hymenium black, 2–4 mm wide at broadest point; stipe 1–2 mm wide at apex, slightly paler than hymenium in older specimens. Asci broad, clavate, (205–)208–256(–276) × (17–)17.5–19.5(–20.5) µm (231.7 ± 22.4 × 18.4 ± 0.9 µm) in rehydrated material, eight-spored, the ascospores found throughout the ascus, resulting in a small hyaline base (FIG. 3A). Ascospores initially hyaline although

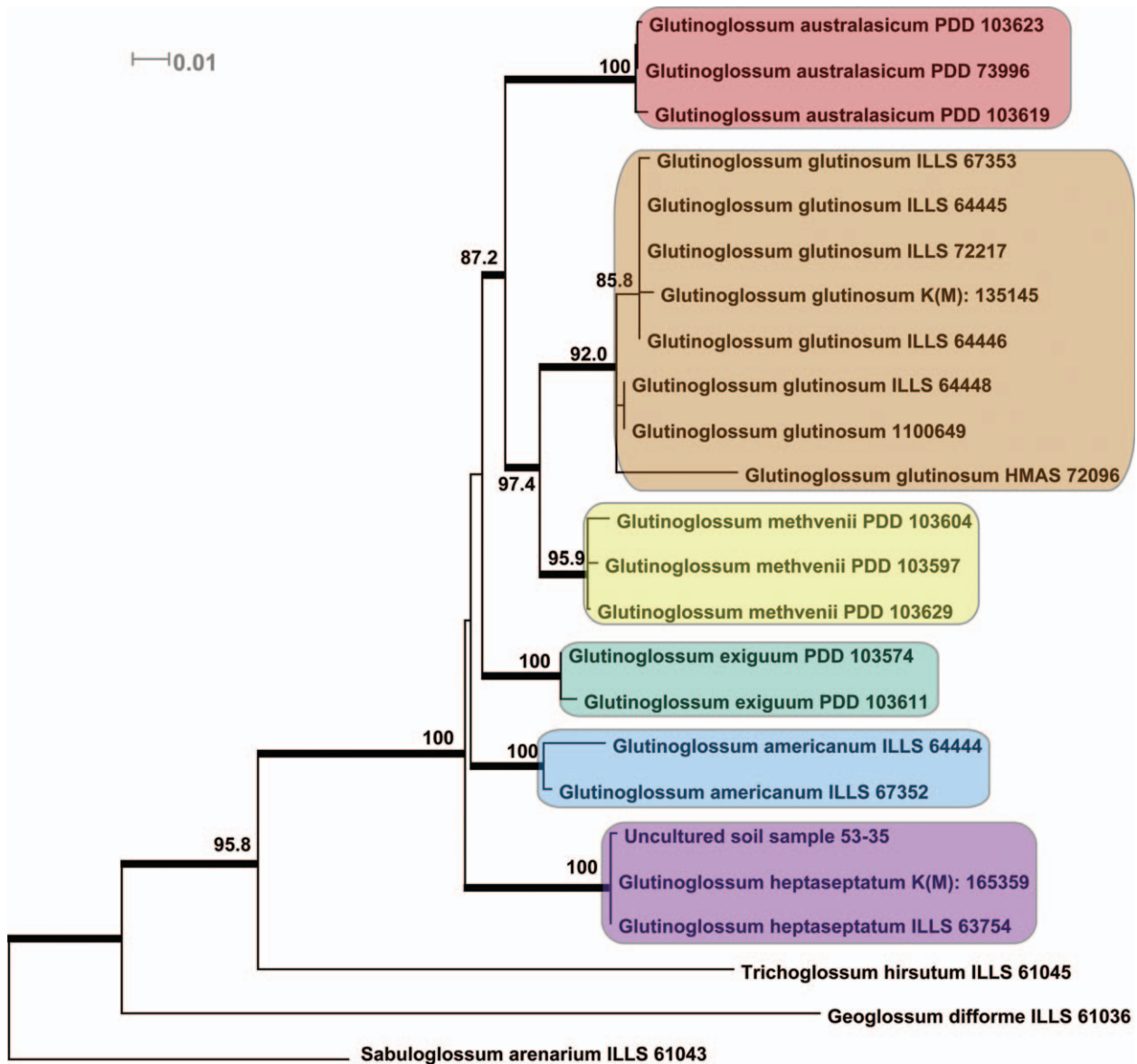


FIG. 1. PhyML maximum likelihood phylogeny of the genus *Glutinoglossum* based on a four-gene dataset (2681 bp), $(-\ln)L$ score = 9606.77868. Individual species are indicated with colored boxes. Numbers at nodes indicate significant BS values ($\geq 70\%$) based on 1000 replicates; thickened branches indicate significant BPP ($\geq 95\%$). *Sabuloglossum arenarium*, *Geoglossum difforme* and *Trichoglossum hirsutum* are included as outgroups. Numbers associated with taxon names are fungarium accession numbers or strain numbers from specimens obtained from GenBank.

becoming brown with maturity, elongate-clavate, slightly curved, predominantly seven-septate, although occasionally three-septate mature spores are present, $(77-)$ $80-92(-99) \times (3.5-)$ $4-5(-6) \mu\text{m}$ ($86.1 \pm 5.7 \times 4.6 \pm 0.5 \mu\text{m}$) (FIG. 3B). Paraphyses filiform, sparsely septate above, apices clavate-cylindrical, slightly agglutinated at the tips by a brown gelatinous matter, enlarged at apex ($3.5-6.5 \mu\text{m}$ diam in rehydrated material) (FIG. 3C).

Distribution and habitat: AUSTRALASIA: Australia (Spooner 1987), New Zealand (this paper). Scattered to gregarious in rich humus and forest soil.

Additional specimens examined: AUSTRALIA. MACQUARIE ISLAND: On wet moss/liverwort peat, beach ridge plateau. 25 Jan 1995. *GAL 6065* (WTU-F 24237); *GAL 6069* (WTU-F 24241); East Half Moon Bay. Above tussock grasses in wet hub field, among *Achrophyllum dentatum*, *Clasmotocolea paucistipula*, *Lophocolea bidentata*. 14 Feb 1995. *GAL 6352* (WTU-F s.n.); In a wet hub field, growing among *Lophocolea*, *Megaceros* and *Plagiochila*. 5 Feb 1995. *GAL 6183* (WTU-F 19902); NEW ZEALAND. BULLER: Paparoa National Park, Cave Creek Track, 26 May 2013, *VPH NZ13-48* (PDD 103610); Inland Pack Track, 26 May 2013, *VPH NZ13-44* (PDD 103606). GISBORNE: Te Urewara National park, Lake Whakamaroena, Waikareiti Track, 9 May 2001,

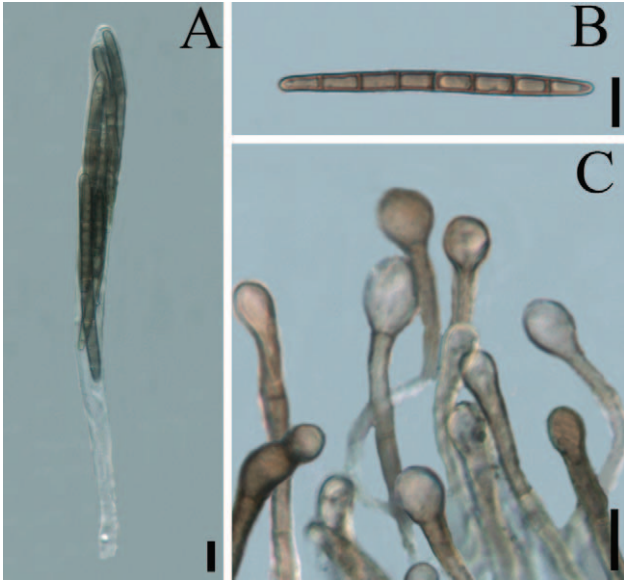


FIG. 2. Micrographs of *Glutinoglossum americanum*. A. Ascus from dried material, B. Discharged ascospore. C. Paraphyses from dried material. Micrographs from holotype material (ILLS 67352). Bars: A–C = 10 μ m.

E. Johannesen s.n., (PDD 73996); Trail to Lake Wakareiti, 12 May 2013, *VPH NZ13-15* (PDD 103579); MARLBOROUGH: Mount Richmond Forest Park, Nydia Track, 31 May 2013, *VPH NZ13-69* (PDD 103631); SOUTH CANTERBURY: Peel Forest, Fern Walk, 29 May 2013, *VPH NZ13-57* (PDD 103619); WESTLAND: Haast-Paringa Cattle Track, Northern car park, 27 May 2013, *VPH NZ13-51* (PDD 103613); *VPH NZ13-52* (PDD 103614).

Comments: Spooner (1987) describes a collection (Beaton 134) from Victoria, Australia, with uniformly

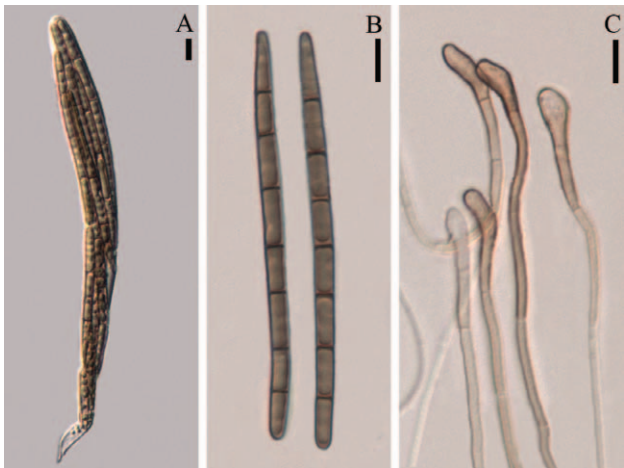


FIG. 3. Micrographs of *Glutinoglossum australasicum*. A. Discharged ascospores. B. Ascus from dried material. C. Paraphyses from dried material. Micrographs from holotype material (PDD 103623). Bars: A–C = 10 μ m.

seven-septate spores that matches the description of *Glutinoglossum australasicum*. This species seems to be the most abundant *Glutinoglossum* in Australasia, accounting for more than 85% of specimens that have been collected from Australia and New Zealand.

***Glutinoglossum exiguum* Hustad & A.N. Mill. sp. nov.**

FIG. 4

Mycobank MB811412

Typification: NEW ZEALAND. TAUPO: Whiranaki Forest Park, Whiranaki Waterfall Trail, on damp mossy embankment, 11 May 2013, *VPH NZ13-10* (**holotype** PDD 103574; **isotype** ILLS 72404).

Etymology: Refers to the small, but well-proportioned ascocarps and ascospores.

Ascocarps scattered, clavate-capitate, 2.5–3.5 cm, black, viscid or not when fresh, with prominent paraphyses extending from the hymenium, visible with a hand lens when fresh; hymenium black, extending one-fourth to one-third the total length of the fruiting body, 2–3 mm wide; stipe black, 1–1.5 mm wide at apex. Asci clavate-cylindrical, (165–)185–245(–261) \times (13.5–)15–16(–17) μ m ($215.7 \pm 25.9 \times 15.3 \pm 1 \mu$ m) in rehydrated material, ascospores occupying 75–85% of the ascus, resulting in a hyaline base (FIG. 4A). Ascospores initially hyaline, becoming brown in maturity, elongate-clavate, (74–)83–92(–95) \times (3–)4–5(–5.5) μ m ($88.5 \pm 4.2 \times 4.5 \pm 0.4 \mu$ m), seven-septate when mature (FIG. 4B). Paraphyses sparsely septate, hyaline below, brown above, distinctly pyriform-globose and inflated at the apex (4.6–7.1 μ m diam in rehydrated material), embedded in a matrix of light brown gelatinous material (FIG. 4C).

Distribution and habitat: AUSTRALASIA: Australia (this paper), New Zealand (this paper). Single to scattered on moss in mixed deciduous forest.

Additional specimens examined: AUSTRALIA. MACQUARIE ISLAND. Handspike Corner Herb Field. In fibrous moist peat beneath *Stilbocarpa*. 20 Apr 1995, *GAL 7225* (WTU-F 23956). NEW ZEALAND. BULLER. Paparoa National Park, Cave Creek Track. 26 May 2013, *VPH NZ13-49* (PDD 103611).

Comments: This species is morphologically similar to *Glutinoglossum australasicum*, but careful morphological examination is capable of differentiating between the two species. *G. australasicum* has a generally larger ascocarp and is found in greater abundance. *G. exiguum* has distinctly pyriform-globose inflated paraphyses tips that are conspicuous in fresh material with a hand lens. Ascospore sizes of the two species largely overlap, but the ascospores in *G. australasicum* are slightly longer and wider.

Dennis (1961) cites a specimen (Berggren 6) from Waitaki, New Zealand, (described as a *nomen nudum* by Cooke as *Geoglossum glabrum* var. *minor*) under



FIG. 4. Micrographs of *Glutinoglossum exiguum*. A. Ascus from dried material. B. Discharged ascospore. C. Paraphyses from dried material. Micrographs from holotype material (PDD 103574). Bars: A–C = 10 µm.

Geoglossum glutinosum in which the ascospores are uniformly seven-septate. We have not been able to locate this material and no further description of the fungus is provided, but it is likely either *Glutinoglossum australasicum* or *Glutinoglossum exiguum*.

Glutinoglossum glutinosum (Pers.) Hustad, A.N. Mill., Dentinger & P.F. Cannon

- = *Geoglossum glutinosum* Pers., *Observ. Myc.* 1:11. 1796.
- = *Geoglossum glutinosum* var. *glutinosum* Pers., *Observ. Myc.* 1:11. 1796.
- = *Geoglossum viscosum* Pers., *Comment. Fung. Clav.* 39. 1797.
- = *Geoglossum glutinosum* B *lubricum* Pers., *Myc. Eur.* 1:197. 1822.
- = *Geoglossum (Cibalocoryne) viscosulum* Hazsl., *Értek. Term. Köréb.* 19:8. 1881.
- = *Geoglossum viscosulum* Sacc., *Syll. Fung.* VIII:43. 1889.
- = *Gloeoglossum glutinosum* (Pers.) Durand, *Ann. Myc.* VI:419. 1908.
- = *Cibalocoryne* [as 'Cibarocoryne'] *glutinosa* (Pers. ex Fr.) Imai, *Bot. Mag. (Tokyo)* 56:525. 1942.

Typification: Europe, Persoon, *Lugd. Bat.* no. 910.261–767 (L 0110938).

Ascocarps single, scattered or caespitose, viscid, becoming gelatinous when wet, clavate, 1.5–5.5 cm tall; hymenium black, one-third to one-half the length of the ascocarp, bilaterally compressed, clavate, cylindrical or ellipsoidal, 3–6 mm wide, sometimes with a vertical median groove; stipe dark brown to black, terete, glabrous, viscid, 2–3 mm wide at apex. Asci cylindrical to narrowly clavate, (175–)220–265(–290) × (10–)12–16(–18) µm (240 ± 18.2 × 13.6 ± 2.1 µm) in rehydrated material, eight-spored; the ascospores occupying the upper 65–75% of the ascus, resulting in a hyaline ascus base. Ascospores fusiform-clavate, straight to slightly curved, (51–)59–65(–77) × (3.5–)4–5(–5.5) µm (62.8 ± 3.6 × 4.4 ± 0.5 µm), often aseptate when young, usually three-septate when mature, although occasionally one-, five- and seven-septate mature ascospores are observed, initially hyaline, eventually becoming brown. Paraphyses sparsely septate, hyaline below, light to dark brown above, broadly obovoid to pyriform (4–11 µm diam in rehydrated material).

Distribution and habitat: AFRICA: Macaronesia (Spooner 1987), Morocco (El Kholfy et al. 2014). ASIA: Bhutan (Prasher and Sharma 1997), China (Zhuang 1998), India (Maas Geesteranus 1965, Prasher and Sharma 1997), Japan (Imai 1941), Philippines (Baker 1914). AUSTRALASIA: Australia (Massee 1897, Spooner 1987, www.gbif.org), New Zealand (Shirley 2004, www.gbif.org). EUROPE: Austria (Massee 1897), Belgium (Massee 1897), Bulgaria (Hinkova and Stoichev 1983), Czech Republic (Velenovsky 1934, Hustad et al. 2013), Denmark (Ohenoja 2000, www.gbif.org), Finland (Massee 1897, Ohenoja 1995, 2000, www.gbif.org), France (Massee 1897, Moingeon and Moingeon 2003), Germany (Massee 1897, Benkert 1976, www.gbif.org), Greece (Zervakis et al. 2002), Ireland (Mitchel 2006, www.gbif.org), The Netherlands (Maas Geesteranus 1964, Hustad et al. 2013), Norway (Ohenoja 2000, www.gbif.org), Portugal (Massee 1897), Slovakia (Kučera and Lizon 2012), Spain (Arauzo and Iglesias 2014), Sweden (Massee 1897, Nannfeldt 1942, www.gbif.org), Switzerland (Massee 1897), United Kingdom (Massee 1897, Dennis 1978, Hustad et al. 2013, www.gbif.org). NORTH AMERICA: Bermuda (Waterston et al. 1945), Canada (Durand 1908, Voitk 2013), Mexico (Ramirez-Lopez and Rios 2007), USA (Mains 1954, Hustad et al. 2013). On soil in wet places and unfertilized grassland. This fungus typically is found associated with hardwoods in North America, whereas it is commonly encountered in pastures and dune slacks in Europe. In India it has been reported from

soil in oak forests or among mosses on stony slopes at 2000 m (Maas Geesteranus 1965).

Additional specimens examined: CZECH REPUBLIC. CENTRAL BOHEMIA: Mladá Boleslav, Baba u Kosmonos, deciduous forest, south slope, 30 Oct 2010, *J. Gaisler s.n.* (ILLS 64443); LIBEREC: Hamr?tejn, deciduous forest, South slope, 25 Aug 2010, *J. Gaisler s.n.* (ILLS 64451); Jablonné v Podje?tédí, in grass and moss, 20 Oct 2010, *Z. Egertová s.n.* (ILLS 64453); Rasovka, mowed meadow, southeast slope, 4 Oct 2010, *J. Gaisler s.n.* (ILLS 67353); THE NETHERLANDS. NORTH HOLLAND: Bergen, on slaperdijk, N52°43', E4°39', 24 Nov 2008, *CFR-241108-D* (ILLS 64449); UNITED KINGDOM. ENGLAND: Lancashire, Clitheroe, Billington, Whalley Old Road, Moonside Cottage, on acid, mossy soil, N53°48', W2°25', 12 Oct 1996, *I. Ridge s.n.* (ILLS 64450); SCOTLAND: Skye, Glen Eynort, in short grass along roadway, N57°15', W6°20', 17 Oct 2011, *ANM 2498* (ILLS 72217); Sleat, Tokavaig, in short grass along roadway N57°7', W5°58', 16 Oct 2011, *ANM 2485* (ILLS 64447); Talisker Beach, in sheep-grazed grass, N57°16', W6°27', 21 Oct 2011, *ANM 2529* (ILLS 64448); WALES: Trefor, on short grass and moss, N52°59', W4°26', 9 Oct 2011, *ANM 2456* (ILLS 64445); Snowdonia National Park, in sheep-grazed grass and moss, N53°4', W4°4', 13 Oct 2011, *ANM 2476* (ILLS 64446); UNITED STATES. NEW HAMPSHIRE: Coos County, Randolph, 19 Aug 1938, *EBM 4207* (MICH).

Comments: We provide an emended description with a narrower range of ascospore sizes more closely approximating the measurements of Persoon's type specimen reported by Durand (1908). There does not seem to be definitive evidence that true *Glutinoglossum glutinosum* exists in Australasia. All specimens collected from Australia and New Zealand have been either *G. australasicum* or *G. exiguum*. The existence of true *G. glutinosum* in Australasia would not be without precedent as species of Geoglossomycetes described from the northern hemisphere (e.g. *Geoglossum cookeanum*, *Trichoglossum hirsutum*) have been collected from Australasia. A wide variety of ascospore sizes have been reported for this species, both from North America and Europe. Owing to the evidence of cryptic speciation in *Glutinoglossum*, it seems likely that many of the published measurements are the result of examination of mixed collections and non-discharged ascospore

Glutinoglossum heptaseptatum Hustad, A.N. Miller, Dentinger & P.F. Cannon

Typification: CZECH REPUBLIC. HRADEC KRÁLOVÉ: Betlem, moist pasture with moss. 20 Oct 2010. *J. Gaisler s.n.* (ILLS 63754).

Ascocarps single or scattered, viscid, 2–4.5 cm tall; hymenium black, viscid, one-fourth to one-third the length of the ascocarp, 3–6 mm wide; stipe dark brown, terete, viscid. Asci clavate, 170–205 × 18–22 µm (190.9

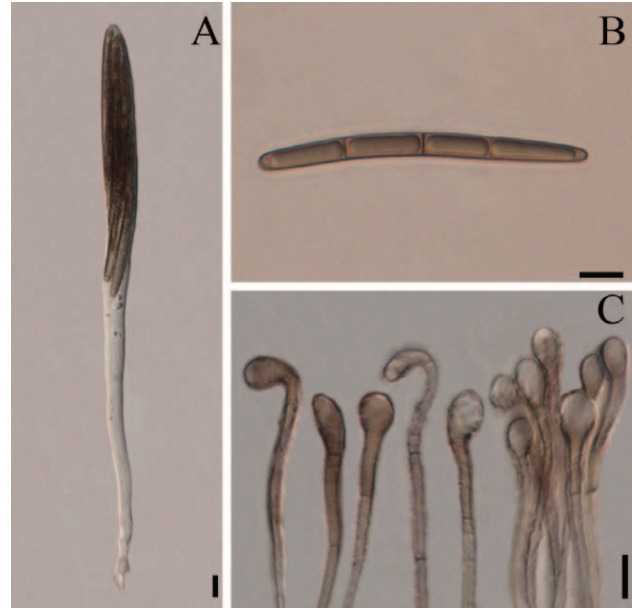


FIG. 5. Micrographs of *Glutinoglossum methvenii*. A. Ascus from dried material. B. Discharged ascospore. C. Paraphyses from dried material. Micrographs from holotype material (PDD 103629). Bars: A–C = 10 µm.

± 12.6 × 19.8 ± 1.1 µm) in rehydrated material, eight-spored, the ascospores occupying the upper 60–75% of ascus, resulting in a hyaline ascus base. Ascospores initially hyaline, brown at maturity, (59–)61–70(–74) × (3.5–)4–5.5(–6.5) µm (65.9 ± 4.8 × 4.8 ± 0.6 µm), predominantly seven-septate at maturity. Paraphyses sparsely septate, hyaline below, light brown above, embedded in a light brown gelatinous matrix, slightly inflated obovoid to pyriform at the apex (4–8 µm diam in rehydrated material).

Distribution and habitat: EUROPE: Czech Republic (Hustad et al. 2013). NORTH AMERICA: USA, Minnesota (Waldrop et al. 2006). On soil in wet places.

Additional specimens examined: CZECH REPUBLIC. HRADEC KRÁLOVÉ: Betlem, 12 km north of Liberec, Protected Landscape Area Jizerske hory (Jizera Mountains), Grassland Research Station Liberec, 50°50'N, 15°5'W. Moist pasture with *Festuca rubra*, *Agrostis capillaris*, *Cirsium palustre*, and moss, Oct 2009, *J. Gaisler s.n.* (K(M): 165359).

Comments: The ascocarps of this fungus presently have been definitively collected only from the type locality, however three identical ITS sequences (DQ421171, DQ421172, DQ421173) that nearly match the holotype sequence of *G. heptaseptatum* (576/577 bp matching) have been recovered from environmental soil samples from Cedar Creek Natural History Area in Minnesota, USA (Waldrop et al. 2006). This site is a restored grassland subdivided into one, two, four, eight and 16 grassland-savanna species plots planted

1994–1995. While no ascocarp collections have been made, this report is the first cited collection of this species other than the type locality and greatly expands the potential range of this species.

Glutinoglossum methvenii Hustad & A.N. Mill. sp. nov.

Mycobank MB811413

FIG. 5

Typification: NEW ZEALAND. MARLBOROUGH: Mount Richmond Forest Park, Nydia Track, on mossy embankment in mixed deciduous forest, 31 May 2013, *VPH NZ13-67* (**holotype** PDD 103629; **isotype** ILLS 72405).

Etymology: Named in honor of Andrew Methven.

Ascocarps scattered to gregarious on damp soil or fallen deciduous wood, 3–4.5 cm tall, viscid-gelatinous when wet, black; hymenium black, one-fourth to one-third the length of the ascocarp although not well differentiated from the stipe, bilaterally compressed, clavate to spatulate, 3–5 mm wide; stipe black, terete, viscid, 1–2 mm wide. Asci cylindrical-clavate, (218–)227–254(–273) × (15.5–)16–17(–18) μm (242.9 ± 11.2 × 16.7 ± 0.5 μm) in rehydrated material, with a long hyaline base occupying nearly 50% of the total length of the ascus when mature (FIG. 5A). Ascospores initially hyaline, brown when mature, elongate-clavate, (65–)69–76(–79) × (4–)5–6(–6.5) μm (72.7 ± 3.7 × 5.7 ± 0.4 μm), the majority three-septate (FIG. 5B), although occasional seven-septate mature spores were noticed. Paraphyses sparsely septate, occasionally curved to slightly hooked at the apex; slightly inflated at the apex (3.5–8 μm diam in rehydrated material), occasionally covered with brown gelatinous matter (FIG. 5C).

Distribution and habitat: AUSTRALIA (Spooner 1987), NEW ZEALAND (this paper). On moss and on dead deciduous log in mixed deciduous forest.

Additional specimens examined: NEW ZEALAND. BAY OF PLENTY: Whinray Scenic Reserve, Motu Falls Trail. Terrestrial on damp soil, 16 May 2013, *VPH NZ13-33* (PDD 103597); BULLER: Greymouth, Point Elizabeth Track. On fallen log, 24 May 2013, *VPH NZ 13-41* (PDD 103603); *VPH NZ13-42* (PDD 103604); Paparoa National Park, Inland Pack Track. On soil, 26 May 2013, *VPH NZ13-45* (PDD 103607).

Comments: This species is identical to a collection (Beaton 338) described by Spooner (1987). The short, stout ascospores and the occasionally curved paraphyses tips distinguish it from all other *Glutinoglossum* species.

KEY TO KNOWN SPECIES OF *GLUTINOGLOSSUM*

- 1 Mature discharged ascospores mostly three-septate 2

- 1 Mature discharged ascospores predominantly seven-septate 3
- 2 Ascospores predominantly three-septate at maturity, mostly 70–80 × 5–6 μm; paraphyses occasionally curved to hooked; Australasia *G. methvenii*
- 2 Ascospores mostly three-septate, though occasionally seven-septate at maturity, narrower than above, mostly 55–75 × 4–5 μm; paraphyses tips enlarged, clavate; northern hemisphere *G. glutinosum*
- 3 Northern hemisphere 4
- 3 Southern hemisphere 5
- 4 Asci broad (18–22 μm wide), hyaline base of ascus occupying up to 25% of total ascus length *G. heptaseptatum*
- 4 Asci narrower than above (12–15 μm wide), hyaline base of ascus occupying 25–45% of total ascus length *G. americanum*
- 5 Asci 205–270 × 17–20 μm; ascocarps slightly larger (up to 5 cm tall); paraphyses tips more clavate-cylindrical than below *G. australasicum*
- 5 Asci generally smaller (165–260 × 13.5–17 μm); ascocarps also slightly smaller (max. 3.5 cm tall); paraphyses tips capitate to broadly ellipsoidal *G. exiguum*

ACKNOWLEDGMENTS

This project was financially supported in part by grants from the University of Illinois Graduate College Dissertation Travel Grant, the Mycological Society of America NAMA Memorial Award, the Mycological Society of America AH Smith Award and the American Society of Plant Taxonomists Graduate Student Research Grant to VPH. The authors thank Jan Gaisler and Kees Roobeek for kindly providing specimens used in this study and Theresa Lebel, Wanda Daley and Peter Johnston for assistance with obtaining specimens from New Zealand. Daniel Raudabaugh and Stephanie Turza are acknowledged for laboratory assistance.

LITERATURE CITED

Aguileta G, Marthey S, Chiapello H, Lebrun MH, Rodolphe F et al. 2008. Assessing the performance of single-copy genes for recovering robust phylogenies. *Syst Biol* 57:613–627, doi:10.1080/10635150802306527

Alfaro ME, Zoller S, Lutzoni F. 2003. Bayes or bootstrap? A simulation study comparing the performance of Bayesian Markov chain Monte Carlo sampling and bootstrapping in assessing phylogenetic confidence. *Mol Biol Evol* 20:255–266, doi:10.1093/molbev/msg028

Arauzo S, Iglesias P. 2014. La familia Geoglossaceae ss. str. En la Peninsula Ibérica y la Macaronesia. *Errotari* 11:166–259.

Baker CF. 1914. The lower fungi of the Philippine Islands. *Leaflets Philippine Bot* 6:2065–2318.

Benkert D. 1976. Bemerkenswerte Ascomyceten der DDR II. Die Gattungen *Geoglossum* und *Trichoglossum* in der DDR. *Mykol Mitteilung* 20:47–92.

Brock PM, Döring H, Bidartondo MI. 2009. How to know the unknown fungi: the role of a herbarium. *New Phytol* 181:719–724, doi:10.1111/j.1469-8137.2008.02703

- Castresana J. 2000. Selection of conserved blocks from multiple alignments and their use in phylogenetic analysis. *Mol Biol Evol* 17:540–552, doi:10.1093/oxfordjournals.molbev.a026334
- Crous PW, Gams W, Stalpers JA, Robert V, Stegehuis G. 2004. MycoBank: an online initiative to launch mycology into the 21st century. *St Mycol* 50:19–22.
- Darriba D, Taboada GL, Doallo R, Posada D. 2012. jModelTest 2: more models, new heuristics and parallel computing. *Nat Methods* 9:772, doi:10.1038/nmeth.2109
- Dennis RWG. 1961. Some inoperculate discomycetes from New Zealand. *Kew Bull* 15:293–320, doi:10.2307/4109373
- . 1978. British ascomycetes. Vaduz, Liechtenstein: Cramer. 585 p.
- Durand EJ. 1908. The Geoglossaceae of North America. *Ann Mycol* 6:387–477.
- Edgar R. 2004. MUSCLE: multiple sequence alignment with high accuracy and high throughput. *Nucleic Acids Res* 32:1792–1797, doi:10.1093/nar/gkh340
- Eernisse DJ, Kluge AG. 1993. Taxonomic congruence versus total evidence, and amniote phylogeny inferred from fossils, molecules and morphology. *Mol Biol Evol* 10:1170–1195.
- El Kholfy S, Outcoumit A, Touhami AO, Belahbib N, Benkirane R, Douira A. 2014. Bibliographic inventory of Tangier's fungi: Catalog of the Ascomycetes fungal flora. *Int J Plant, Animal Environ Sci* 4:77–92.
- Felsenstein J. 1985. Confidence limits on phylogenies: an approach using the bootstrap. *Evolution* 39:783–791, doi:10.2307/2408678
- Gouy M, Guindon S, Gascuel O. 2010. SeaView 4: a multiplatform graphical user interface for sequence alignment and phylogenetic tree building. *Mol Biol Evol* 27:221–224, doi:10.1093/molbev/msp259
- Guindon S, Dufayard JF, Lefort V, Anisimova M, Hordijk W, Gascuel O. 2010. New algorithms and methods to estimate maximum-likelihood phylogenies: Assessing the performance of PhyML 3.0. *Syst Biol* 59:307–321, doi:10.1093/sysbio/syq010
- , Gascuel O. 2003. A simple, fast and accurate method to estimate large phylogenies by maximum-likelihood. *Syst Biol* 52:696–704, doi:10.1080/10635150390235520
- Hazslinszky F. 1881. Rendhagyó köggombák. *Érték termés kör* 11:1–24, Figs. 1–6.
- Hillis DM, Bull JJ. 1993. An empirical test of bootstrapping as a method for assessing confidence in phylogenetic analysis. *Syst Biol* 42:182–192, doi:10.2307/2992540
- Hinkova T, Stoichev G. 1983. New and rare macromycetes for Bulgaria. *Fitologiya* 23:70–72.
- Hustad VP, Miller AN, Dentinger BTM, Cannon PF. 2013. Generic circumscriptions in Geoglossomycetes. *Persoonia* 31:101–111, doi:10.3767/003158513X671235
- , ———, Moingeon J-M, Priou J-P. 2011. Inclusion of *Nothomitra* in Geoglossomycetes. *Mycosphere* 2:646–654, doi:10.5943/mycosphere/2/6/5
- Imai S. 1941. Geoglossaceae Japoniae. *J Fac Agric Hokkaido Univ* 45:155–264.
- . 1942. Contributiones ad studia monographica Geoglossacearum. *Bot Mag (Tokyo)* 56:523–527, doi:10.15281/jplantres1887.56.523
- Kearsey SE, Labib K. 1998. MCM proteins: evolution, properties and role in DNA replication. *Biochim Biophys Acta* 1398:113–136, doi:10.1016/S0167-4781(98)00033-5
- Kluge AG. 1989. A concern for evidence and a phylogenetic hypothesis of relationships among *Epicrates* (Boidae, Serpentes). *Syst Biol* 38:7–25, doi:10.1093/sysbio/38.1.7
- Kučera V and Lizoň P. 2012. Geoglossaceous fungi in Slovakia III. The genus *Geoglossum*. *Biologia* 67:654–658.
- Landvik S. 1996. *Neolecta*, a fruit-body producing genus of the basal ascomycetes, as shown by SSU and LSU rDNA sequences. *Mycol Res* 100:199–202, doi:10.1016/S0953-7562(96)80122-5
- Lutzoni F, Kauff F, Cox C, McLaughlin D, Celio G. et al. 2004. Assembling the Fungal Tree of Life: progress classification and evolution of subcellular traits. *Am J Bot* 91:1446–1480, doi:10.3732/ajb.91.10.1446
- Maas Geesteranus RA. 1964. De fungi van Nederland I. Geoglossaceae—aardtongen. *Wentensch Meded Koninkl Nederl Natuurhist Verenig* 52:1–24.
- . 1965. Geoglossaceae of India and adjacent countries. *Persoonia* 4:19–46.
- Mains EB. 1954. North American species of *Geoglossum* and *Trichoglossum*. *Mycologia* 46:586–631.
- Masse G. 1897. A monograph of the Geoglossaceae. *Ann Bot* 11:225–306.
- Matheny PB, Liu YJ, Ammirati JF, Hall BD. 2002. Using RPB1 sequences to improve phylogenetic inference among mushrooms (*Inocybe*: Agaricales). *Am J Bot* 89:688–698, doi:10.3732/ajb.89.4.688
- Miller MA, Pfeiffer W, Schwartz T. 2010. Creating the CIPRES science gateway for inference of large phylogenetic trees. Proceedings of the Gateway Computing Environments Workshop (GCE), 14 Nov 2010, New Orleans, Louisiana.
- Mitchel D. 2006. Survey of the grassland fungi of County Clare. Kilkenny: Heritage Council.
- Moingeon S, Moingeon JM. 2003. Les Geoglossaceae en Franche-Comte. *Bull Soc Hist Nat Doubs* 89:61–66.
- Moir D, Steward SE, Osmond BC, Botstein D. 1982. Cold-sensitive cell division-cycle mutants of yeast: isolation, properties and pseudoreversion studies. *Genetics* 100:547–563.
- Nannfeldt JA. 1942. The Geoglossaceae of Sweden. *Ark Bot* 30A:1–67.
- Ohenoja E. 1995. Occurrence of *Geoglossum*, *Trichoglossum* and *Microglossum* (Ascomycota, Leotiales) in Finland. *Doc Mycol* 25:285–294.
- . 2000. Geoglossaceae. In: Hansen L, Knudsen H, eds. *Nordic Macromycetes*. Vol. 1. Copenhagen: Nordsvamp. p 177–183.
- Persoon CH. 1794. Neuer Versuch einer systematischen Eintheilung der Schwämme. *Neues Mag Bot* 1:63–128.
- . 1796. *Observationes Mycologicae* 1. Leipzig, Germany: Gesnerus, Usterius & Wolfius.

- . 1797. *Commentatio de Fungis Clavaeformibus.*, Leipzig: Germany: Petrum, Phillipum & Wolf.
- . 1822. *Mycologia Europaea*, Erlangen: Germany: Palmius, vol 1.
- Posada D, Buckley TR. 2004. Model selection and model averaging in phylogenetics: advantages of Akaike information criterion and Bayesian approaches over likelihood ratio tests. *Syst Biol* 53:793–808, doi:10.1080/10635150490522304
- Prasher IB, Sharma R. 1997. *Geoglossum* Pers. Geoglossaceae, Leotiales in eastern Himalayas. In: Chahal SS, Prashar IB, Randhawa HS, Arya S, eds. *Achievements and prospects in mycology and plant pathology*. Dehradun, India: International Book Distributors. p 12–19.
- Promputtha I, Miller AN. 2010. Three new species of *Acanthostigma* (Tubeufiaceae, Dothideomycetes) from the Great Smoky Mountains National Park. *Mycologia* 102:574–587, doi:10.3852/09-051
- Raja HA, Schoch CL, Hustad VP, Shearer CA, Miller AN. 2011. Testing the phylogenetic utility of *MCM7* in the Ascomycota. *MycKeys* 1:46–56, doi:10.3897/mycokeys.1.1966
- Rambaut A, Drummond AJ. 2009. Tracer 1.5. Available from <http://tree.bio.ed.ac.uk/software/tracer>.
- Ramirez-Lopez I, Rios MV. 2007. El conocimiento taxonómico de Geoglossaceae sensu lato (Fungi: Ascomycetes) en México con énfasis en la zona centro y sur. *Rev Mex Micol* 25:41–49.
- Ronquist F, Teslenko M, van der Mark P, Ayers D, Darling A et al. 2012. MrBayes 3.2: Efficient Bayesian phylogenetic inference and model choice across a large model space. *Syst Biol* 61:539–542, doi:10.1093/sysbio/sys029
- Saccardo PA. 1884. *Conspectus eorum Discomycetum hucusque cognitorum*. *Bot Cent* 18:213–220, 247–257.
- Schmitt I, Crespo A, Divakar PK, Fankhouser JD, Herman-Sackett E et al. 2009. New primers for single-copy genes in fungal phylogenetics and systematics. *Persoonia* 23:35–40, doi:10.3767/003158509X470602
- Schoch CL, Sung G-H, López-Giráldez F, Townsend JP, Miadlikowska J et al. 2009. The Ascomycota tree of life: A phylum wide phylogeny clarifies the origin and evolution of fundamental reproductive and ecological traits. *Syst Biol* 58:224–239, doi:10.1093/sysbio/syp020
- Shirley C. 2004. Fungi of Murphy's Bush. *Auckland Bot Soc J* 59:101–103.
- Spooner BM. 1987. Helotiales of Australasia: Geoglossaceae, Orbiliaceae, Sclerotiniaceae, Hyaloscyphaceae. *Bib Mycol* 116:1–711.
- Stiller JW, Hall BD. 1997. The origin of red algae: implications for plastid evolution. *PNAS* 94: 4520–4525, doi:10.1073/pnas.94.9.4520
- Townsend JP. 2007. Profiling phylogenetic informativeness. *Syst Biol* 56:222–231, doi:10.1080/10635150701311362
- Velenovsky J. 1934. *Monographica Discomycetum Bohemiae.*, Novyt Bydzov: Janata.
- Vilgalys R, Hester M. 1990. Rapid identification and mapping of enzymatically amplified ribosomal DNA from several *Cryptococcus* species. *J Bacteriol* 172:4238–4246.
- Voitk A. 2013. Earth tongues of Newfoundland and Labrador. *Omphalina* 4:14–19.
- Waldrop MW, Zak DR, Blackwood CB, Curtis CD, Tilman D. 2006. Resource availability controls fungal diversity across a plant diversity gradient. *Ecol Lett* 9:1127–1135., doi:10.1111/j.1461-0248.2006.00965.x
- Wang Z, Binder M, Hibbett D. 2005. Life history and systematics of the aquatic discomycetes *Mitrella* (Helotiales, Ascomycota) based on cultural, morphological and molecular studies. *Am J Bot* 92:1565–1574, doi:10.3732/ajb.92.9.1565
- , Nilsson RH, Lopez-Giraldez F, Zhuang W, Dai Y, Johnston PR, Townsend JP. 2011. Tasting soil fungal diversity with earth tongues: Phylogenetic test of SATé alignments for environmental ITS data. *PLoS One* 6: e19039.
- Waterston JM, Sinden JW, Whetzel HH. 1945. Notes on the Geoglossaceae of Bermuda. *Mycologia* 37:32–36, doi:10.2307/3754847
- White TJ, Bruns T, Lee S, Taylor JW. 1990. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: Innis MA, Gelfand DH, Sninsky JJ, White TJ, eds. *PCR protocols: a guide to methods and applications*. New York: Academic Press. p 315–322.
- Wiens JJ. 1998. Combining datasets with different phylogenetic histories. *Syst Biol* 47:568–581, doi:10.1080/106351598260581
- Zervakis GI, Dimou DM, Polemis E, Karadelev M. 2002. Mycodiversity studies in selected ecosystems of Greece II. Macrofungi associated with conifers in the Taygetos Mountain (Peloponnese). *Mycotaxon* 82:177–205.
- Zhuang W. 1998. Geoglossaceae. *Flora Fungorum Sin* 8: 78–127.