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Four New Families of Arbuscular Mycorrhizal Fungi Within the Order Glomerales

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Abstract: Based on molecular phylogenetic analyses, but also considering morphological characters, four new families are separated from the family Glomeraceae within the Glomerales and the class Glomeromycetes. The revised family Glomeraceae comprises only four genera: the type genus *Glomus*, *Complexispora*, *Sclerocarpum* and *Simiglomus*. Septoglomeraceae fam. nov. comprises, besides *Septoglomus*, also *Funneliformis*, *Funneliglomus*, *Blaszkowskia* and *Viscospora*. Sclerocystaceae fam. nov. is represented by the type genus *Sclerocystis*, but also by *Halonatospora*, *Oehlia*, *Parvocarpum*, *Rhizoglomus* and *Silvaspora*. Kamienskiaceae fam. nov. encompasses *Kamienskia*, *Microkamienskia* and *Epigeocarpum*. Finally, Dominikiaceae fam. nov. includes the genera *Dominikia*, *Macrodominikia* gen. nov. *Microdominikia*, *Nanoglomus* and *Orientoglomus*. The genera *Oehlia* and *Halonatospora* form two other clades well separated from *Silvaspora*, *Sclerocystis* and *Rhizoglomus* and might represent two further families within the Glomerales. Such deeper-going separation is to our opinion fully supported by molecular phylogeny, but in view of the low numbers of taxa, this separation is not yet needed at this stage of research progress.

Keywords: classification; systematics; taxonomy; arbuscular mycorrhizal fungi

1. Introduction

The fungal phylum Glomeromycota (arbuscular mycorrhizal fungi, AMF) currently comprises three classes, six orders, sixteen families and > 50 genera [1–6]. In the class Glomeromycetes, four orders were described, which are called Diversisporales, Entrophosporales, Gigasporales and Glomerales [4,7–10]. Within the Glomerales and Entrophosporales there has been just one family for each other, the Glomeraceae and the Entrophosporaceae [9,11], while Diversisporales comprises the type family (Diversisporaceae) and three other families: Acaulosporaceae, Pacisporaceae and Sacculosporaceae [9,12–14]. The order Gigasporales comprises the five families Gigasporaceae, Scutellosporaceae, Racocetraceae, Dentiscutataceae and Intraornatosporaceae [2,3,15,16]. Finally, in the most ancient clades of AM fungi there are Archaeosporales, including the families Archaeosporaceae, Ambisporaceae, Geosiphonaceae and Polonosporaceae, and Paraglomerales, comprising Paraglomeraceae and Pervetustaceae [2,3].

Within the last few years, new genera were separated within the Glomerales, due to concomitant morphological and molecular analyses and especially phylogenetic progresses [5,6,17–21]. The Glomeraceae is the family with highest species richness in Glomeromycota (> 150), and the latest phylogenetic trees for Glomeraceae species revealed at least four or five major clades [4,6,17,19–21]. *Glomus macrocarpum*, *Septoglomus constrictum*, *Sclerocystis coremioides*, *Kamienskia bistrata* and *Dominikia minuta* might be the most representative species of these five major clades. The family Glomeraceae also includes two genera of outstanding importance, which are counted among the 50

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most cited fungal genera within the last decades, *Funneliformis* and *Rhizoglomus* due to the species *F. mosseae* and *R. irregulare*, and often used in basic research, applied sciences and also commercially applied, e.g. to enhance soil fertility and improve plant nutrition and root health [22]. Two complexes of genera were already reported in previous works, the *Dominikia* and the *Septoglomus* complexes [5,19). Based on our observations (phylogenetic and morphological analyses), our hypothesis is that at least five major clades/complexes exist in Glomeraceae, and thus the family could be divided into five different families, based on these five major clades/complexes. Thus, the objectives of this study were to perform a thorough phylogenetic analysis on the described Glomerales species and to re-organize all the families and genera justified within the order. At the end, a revision of the family Glomeraceae will be needed, and new families would have to be described, based on significative type species.

2. Materials and Methods

2.1. Phylogenetic Analyses

To reconstruct the phylogeny, two alignments (datasets) were generated with AM fungal sequences from the genera of Glomerales (Supplementary Material, Spreadsheet S1). The first dataset was used to reconstruct the phylogeny based on partial SSU, ITS region, and partial LSU nrDNA. The second was used to reconstruct the phylogeny based on partial SSU, 5.8S, and partial LSU nrDNA. *Entrophospora etunicata* (W.N. Becker & Gerd.) Błaszk., Niezgoda, B.T. Goto & Magurno was included as outgroup in all analyses. For some isolates just sequences from the partial LSU nrDNA were used, which are indicated in the phylogenetic trees generated. Sequences from *Septoglomus titan* were not used in the phylogenetic analyses because we decided do not include sequences in the alignment with less than 500bp (sequences from *S. titan* have 457bp). According to Silva et al. [5] it is not possible to be sure about the phylogenetic position of this species due to the short LSU nrDNA fragment and unavailability of its ITS sequences. The datasets were aligned in Mafft [23], using the default parameters. Prior to phylogenetic analyses, the model of nucleotide substitution was estimated using Topali 2.5 [24]. Bayesian (two runs over 5×10^6 generations, with a sample frequency of 500 and a burnin value of 25%) and maximum likelihood (1,000 bootstrap) analyses were performed, respectively, in MrBayes 3.1.2 [25] and PhyML [26], launched from Topali 2.5, using the GTR + G model.

2.2. Specimen Analyses

We analyzed morphologically all species belonging to the Glomerales, and in detail specimens representing > 120 taxa. Holotype, isotype, paratype and ex-type materials were examined along with representatives from institutional herbaria — OSC, FH, Z+ZT, URM (Recife, Brazil), Embrapa Agrobiologia (Seropédica, Brazil), DCS-UFLA (Lavras, Brazil), International Culture Collection of Vesicular-Arbuscular Mycorrhizal Fungi (INVAM), Swiss collection of Arbuscular Mycorrhizal Fungi (SAF, Agroscope ART Zurich) — and private AM fungal collections curated by Sieverding, Oehl, Trappe, Błaszkowski, and McGee, see also Oehl et al. [9]. The Hall & Abbott [27] photographic slide collection was also reviewed. Older specimens (mounted on microscopic slides prior to 1990) were mostly mounted in lactophenol, while others were fixed with polyvinyl alcohol-lactic acid-glycerol (PVLG) or in a mixture of PVLG + Melzer's reagent, which post-1990 are the principal fixing media [28]. Newly mounted spores and sporocarps from collections or from cultures were fixed using the latter two fixing media, or occasionally also in a mixture of 1:1 lactic acid to water, in Melzer's reagent, and in water. When available, spores freshly isolated from soils or bait cultures were also mounted and analyzed. All spore observations and all information on spore characteristics are based on spores extracted from soil, from trap cultures or from single or multiple spore-derived pure cultures. No information is provided from in-vitro-cultured materials. Spore wall terminology follows the nomenclature of Walker [29] and Stürmer & Morton [30]. Analyses of the spore walls, the germination structures and all other mycorrhizal structures were performed using compound

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microscopes at 100–1000×. For this paper, all original species descriptions and published species emendations were also considered and thoroughly studied.

3. Results

3.1. Molecular Phylogeny

By observing the generated phylogenetic trees, five different clades were separated in the Glomerales (Figures 1 and 2). The clade "A", composed by the genera *Halonatospora*, *Oehlia*, *Rhizoglomus*, *Sclerocystis* and *Silvaspora*, had strong support for Bayesian inference (BI) and ML analyses (1.00/88%) in the first tree obtained (Figure 1), however was not supported by ML analyses and presented a support of 0.88 for BI in the second tree (Figure 2). The clade "B" presented strong support for BI in the two trees obtained, however shown support for ML (94%) just in the first tree. This clade comprises the genera *Epigeocarpum*, *Kamienskia* and *Microkamienskia*. The clades "C", "D" and "E" showed strong support in all analyses of the two trees. Clade "C" is represented by *Blaszkowskia*, *Funneliformis*, *Funneliglomus*, *Septoglomus* and *Viscospora*, clade "D" by *Complexispora*, *Glomus* and *Sclerocarpum*, and clade "E" by *Dominikia*, *Macrodominikia*, *Microdominikia*, *Nanoglomus* and *Orientoglomus*, respectively. Our findings indicate that each clade represent a different family in Glomerales with in total 22 AMF genera, which can be separated morphologically by a combination of spore morphology, characteristics of the subtending hyphae and pore closure, and spore formation features in sporocarps, spore clusters or exclusively singly in soils (Supplementary Material, Table S1).

3.2. Taxonomy

Glomerales J.B. Morton and Benny, emend. Błaszk., B.T. Goto, and Magurno, Mycotaxon 37: 473. 1990.

MycoBank MB 90425

Description: Spores formed in soils or in roots, terminally on or intercalary in hyphae, either singly, in loose spore clusters, or in multiple-spored loose to compact sporocarps, when compact spores are randomly distributed or organized around a central plexus of hyphae. Compact sporocarps with or without peridium, and with or without intrasporocarpic hyphal gleba. Spores with one mono-to-multiple layered wall. Wall of the subtending hyphae (SH) conspicuously continuous and concolorous with the spore wall, or slightly lighter in color than the spore wall; SH funnel-shaped, cylindrical or constricted; straight, curved or flared; pore at spore base open, or closed by a bridging, straight or curved septum or by introverted wall thickening, or SH closed at some distance from the spore base by a septum or plug-like structure forming typical vesicular-arbuscular mycorrhiza, with mycorrhizal structures that stain blue to dark blue in trypan blue.

Type family: Glomeraceae Piroz. & Dalpé

Other families: Septoglomeraceae Oehl et al., Sclerocystaceae Oehl et al., Dominikiaceae G.A. Silva et al., Kamienskiaceae G.A. Silva et al.

Type genus: *Glomus* Tul. & C. Tul.

Other genera: *Sclerocarpum* B.T. Goto et al., *Complexispora* Błaszk. et al., *Simiglomus* Sieverd. et al., *Blaszkowskia* G.A. Silva & Oehl, *Septoglomus* Sieverd. et al., *Funneliformis* C. Walker & A. Schüssler, *Funneliglomus* Corazon-Guivin et al., *Viscospora* Sieverd. et al., *Rhizoglomus* Sieverd. et al., *Oehlia* Błaszk. et al., *Sclerocystis* Berk. & Broome, *Halonatospora* Błaszk. et al., *Kamienskia* et al. Błaszk. et al., *Microkamienskia* G.A. Silva et al., *Dominikia* Błaszk. et al., *Macrodominikia* Oehl et al., *Microdominikia* Oehl et al., *Nanoglomus* Corazon-

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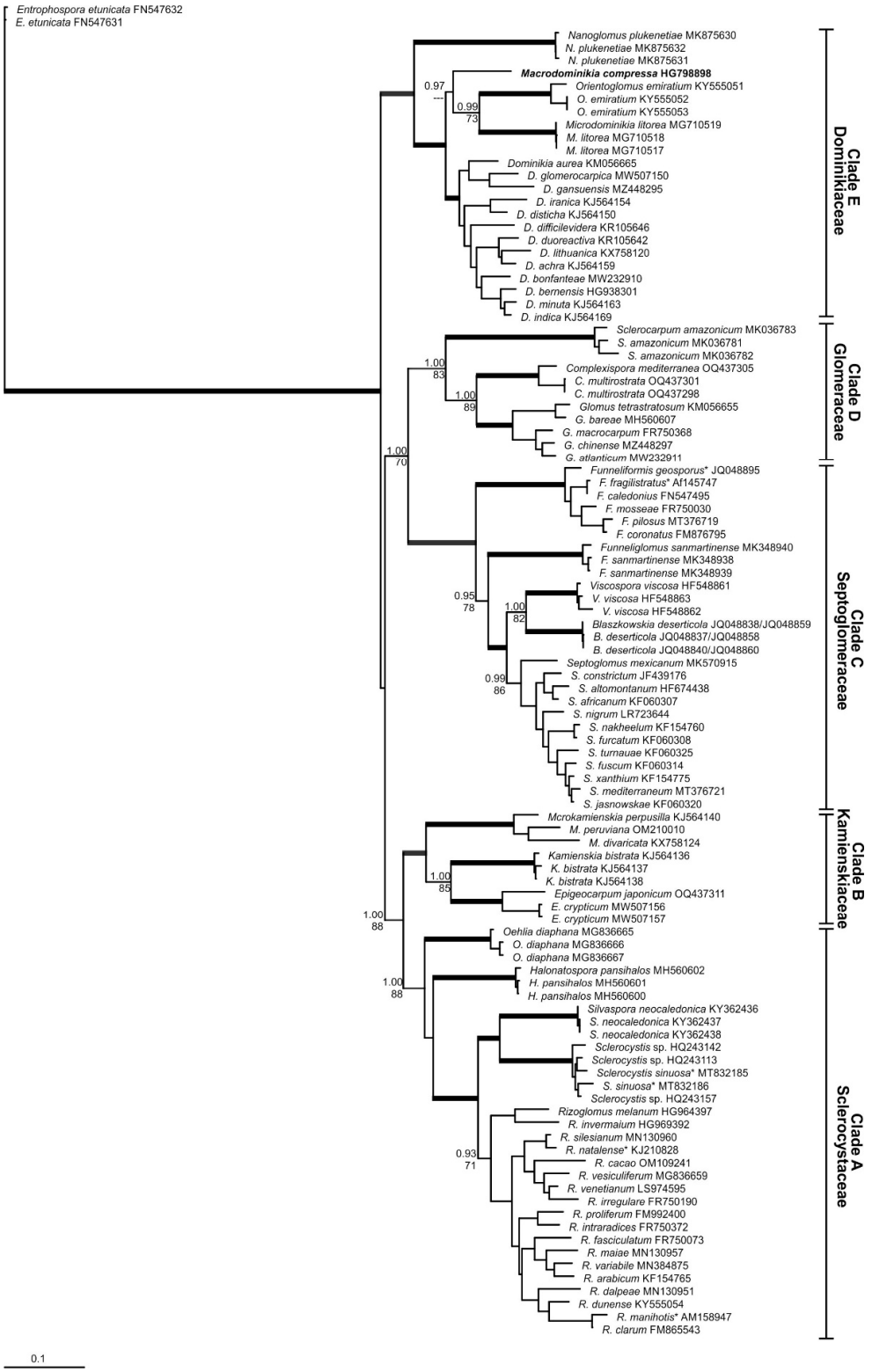

Figure 1. Phylogenetic tree obtained by analysis from partial SSU, ITS region, and partial LSU nrDNA sequences of Glomerales. Sequences are labeled with their database accession numbers. Support values from Bayesian inference (BI) and maximum likelihood (ML) are shown just for genera level or above. Only support values of at least 70% are shown. Thick branches represent clades with more than 90% of support in all analyses. The new genus is in bold. The tree was rooted by Entrophospora etunicata. Sequences with only the partial LSU nrDNA are indicated by *.

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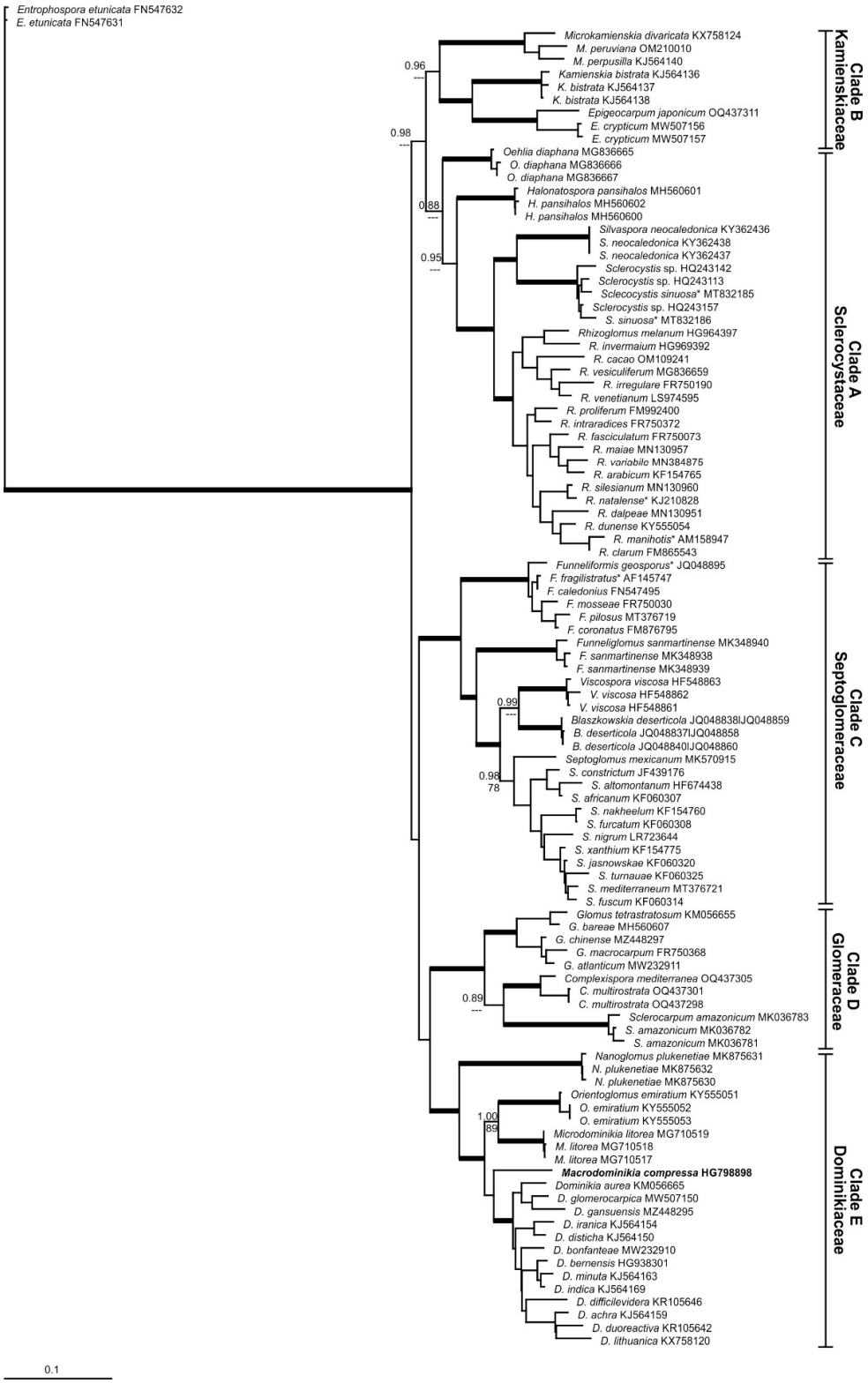

Figure 2. Phylogenetic tree obtained by analysis from partial SSU, 5.8S, and partial LSU nrDNA sequences of Glomerales. Sequences are labeled with their database accession numbers. Support values from Bayesian inference (BI) and maximum likelihood (ML) are shown just for genera level or above. Only support values of at least 70% are shown. Thick branches represent clades with more than 90% of support in all analyses. The new genus is in bold. The tree was rooted by Entrophospora etunicata. Sequences with only the partial LSU nrDNA are indicated by *.

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Guivin et al., *Orientoglomus* G.A. Silva et al., *Epigeocarpum* Błaszk., et al., *Silvaspora* Błaszk., et al., *Parvocarpum* Magurno.

Glomeraceae Piroz. & Dalpé emend. Oehl, G.A. Silva & Sieverd., Symbiosis 7: 19. 1989.

MycoBank 82026

Emended description: Spores formed in soil and sometimes in roots, terminally on or intercalary in hyphae, either singly, in loose spore clusters, or in multiple-spored loose to compact sporocarps, when compact, then spores are randomly distributed, but not organized around a central plexus of hyphae. Compact sporocarps with or without peridium, and with or without intrasporocarpic hyphal gleba. Spores with one (mono-)bi-to-multiple layered wall. SH wall conspicuously continuous and concolorous with the spore wall, or slightly lighter in color than the spore wall; SH cylindrical or slightly funnel-shaped or slightly constricted; straight, curved or flared; pore at spore base open, or closed at spore base by a straight or curved septum or by introverted wall thickening, or at some distance from the spore base by a bridging septum arising from the SH wall; forming typical vesicular-arbuscular mycorrhiza, with mycorrhizal structures that stain blue to dark blue in trypan blue.

Type genus: *Glomus* Tul. & C. Tul.

Other genera: *Sclerocarpum* B.T. Goto et al., *Simiglomus* Sieverd. et al., *Complexispora* Błaszk. et al.

Glomus Tul. & C. Tul., Giorn. Bot. Ital., Anno 1, 2(7–8): 63. 1845.

MycoBank 20244

Emended description: Spores formed singly within soil or sometimes roots, in disorganized, multiple-spored loose spore clusters or in compact sporocarps; compact, but not radially organized around a central hyphal plexus, without or with peridium, spores are either in sporocarp, or organized. Spores with a mono-to-multiple layered wall. SH wall conspicuously continuous and concolorous with the spore wall, or slightly lighter in color than the spore wall. Spore pore closure often by introverted wall thickening, sometimes supported by a short bridging septum, rarely open. Forming typical vesicular-arbuscular mycorrhiza, with mycorrhizal structures that stain blue to dark blue in trypan blue.

Type species: ***Glomus macrocarpum*** Tul & C. Tul., Giorn. Bot. Ital., Anno 1, 2(7–8): 63. 1845.

MycoBank MB 240247

≡ *Endogone macrocarpa* (Tul. & C. Tul.) Tul. & C. Tul., Fungi Hypog.: 182. 1851.

MycoBank MB 218537

≡ *Endogone guttulata* E. Fisch., Ber. Schweiz. Bot. Ges. 32: 13. 1923.

≡ *Endogone nuda* Petch., Ann. R. Bot. Gdns Peradeniya 9: 322. 1925.

≡ *Endogone pampaloniana* Bacc., Nuovo Giorn. Bot. Ital., n.s. 10: 90. 1903.

≡ *Paurocotylis fulva* var. *zelandica* Cooke, Grevillea 8: 59. 1879.

Simiglomus Sieverd., G.A. Silva & Oehl, Mycotaxon 116: 104. 2011.

MycoBank MB 518435

Description: Spores formed in soil or in roots, terminally on or intercalary in hyphae, in small to multiple-spored loose clusters, randomly distributed, but not organized around a central plexus of hyphae. Loose sporocarps generally without peridium, and without intrasporocarpic hyphal gleba. Spores with one mono-to-multiple layered wall. SH wall conspicuously continuous and concolorous with the spore wall, or slightly lighter in color than the spore wall; SH cylindrical or slightly to rarely slightly funnel-shaped or constricted; straight, curved or flared; pore open at spore base, but often in some distance by one to multiple bridging, straight to curved, septum, not supported by introverted wall thickening; forming typical vesicular-arbuscular mycorrhiza, with mycorrhizal structures that stain blue to dark blue in trypan blue.

Type species: ***Simiglomus hoi*** (S.M. Berch & Trappe) G.A. Silva, Oehl & Sieverd.

MycoBank MB 518461

Basionym: *Glomus hoi* S.M. Berch & Trappe, Mycologia 77: 654. 1985.

Sclerocarpum B.T. Goto, Błaszk., Niezgoda, Kozłowska & Jobim, Mycol. Progr. 18(3): 375. 2019.

MycoBank MB 828316

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Description: Producing spores in scleroid, epigeous and sub-hypogeous, light- to dark-colored unorganized sporocarps, very hard to break, with a peridium and a gleba comprising hyphae and glomoid glomerospores (= spores) with a single SH; spores hyaline; globose to subglobose; small, < 100 µm diam; frequently ovoid; Spore wall composed a few smooth layers, of which one layer laminate and much thicker than the outermost layer, forming the spore surface. SH funnel-shaped with a wall continuous with the spore wall; pore narrow, open or occluded by thickening of the SH wall, or (rarely) occluded by a straight or slightly invaginated septum continuous with some innermost laminae of spore wall layer 2; septum, positioned at the spore base.

Type species: ***Sclerocarpum amazonicum*** B.T. Goto, Błaszk., Niezgoda, Kozłowska & Jobim, Mycol. Progr. 18: 377. 2019.

MycoBank MB 828317

Specific morphological observation on *Sclerocarpum*

Especially *Sclerocarpum* shows well-defined, outstanding morphological characteristics of their hard sporocarps, their spores, which are exclusively formed within these sporocarps and of their funnel-shaped subtending hyphae including a pronounced wall-thickening at the spore base at the point of pore closure. Three *Glomus* species described in the past, clearly combine with all these characteristics. These are *G. convolutum*, *G. pellucidum* and *G. segmentatum* [31–33]. Consequently, these species are hereafter transferred to the genus *Sclerocarpum*.

Sclerocarpum convolutum (Gerd. & Trappe) Oehl, Sieverd. & G.A. Silva, comb. nov.

MycoBank MB 855472

Basionym: *Glomus convolutum* Gerd. & Trappe, Mycol. Mem. 5: 42. 1974. Mycological Memoirs 5: 42. 1974.

MycoBank MB 314590

Sclerocarpum segmentatum (Trappe, Spooner & Ivory) Oehl, Sieverd. & G.A. Silva, comb. nov.

MycoBank MB 855473

Basionym: *Glomus segmentatum* Trappe, Spooner & Ivory, Trans. Br. Mycol. Soc. 73: 362. 1979.

MycoBank MB 314610

Sclerocarpum pellucidum (McGee & Pattinson) Oehl, Sieverd. & G.A. Silva, comb. nov.

MycoBank MB 855474

Basionym: *Glomus pellucidum* McGee & Pattinson, Austral. Syst. Bot. 15: 120. 2002.

MycoBank MB 374910

Complexispora Błaszk., B.T. Goto, Niezgoda & Magurno, Mycol. Progr. 22(5, no. 34): 7. 2023.

MycoBank MB 847607

Description: Producing hypogeous glomoid spores singly and in clusters. Spores four to six layers, of which two consist of tightly adherent sublayers (laminae). SH cylindrical to funnel-shaped, concolorous with the spore wall layer, with a wall composed of layers continuous with all spore wall layers, except for the innermost layer. Pore closed by a septum continuous with the innermost spore wall layer, occasionally also by a septum connecting the inner surfaces of the main structural laminate spore wall layer. Forming mycorrhiza with vesicles and arbuscules staining dark in Trypan blue.

Type species: ***Complexispora multistratosa*** Błaszk., B.T. Goto, Niezgoda & Magurno, Mycol. Progr. 22(5, no. 34): 6. 2023.

MycoBank MB 847608

Septoglomeraceae Oehl, G.A. Silva, Sieverd. fam. nov.

Mycobank MB 855466

Description: Spores formed in soil and sometimes in roots, terminally on or intercalary in hyphae, either singly, in loose spore clusters or sporocarps. Compact sporocarps with or without peridium, without intrasporocarpic hyphal gleba. Spores with one mono-to-multiple layered wall. SH wall conspicuously continuous and concolorous with the spore wall, or slightly lighter in color than the spore wall; SH pronounced funnel-shaped, slightly funnel-shaped, cylindrical or constricted, rarely inflated; straight, curved or flared; forming typical vesicular-arbuscular mycorrhiza, with mycorrhizal structures that stain blue to dark blue in trypan blue.

Type genus: *Septoglomus* Sieverd. et al.

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Other genera: *Funneliformis* C. Walker & A. Schüssler, *Funneliglomus* Corazon-Guivin et al., *Viscospora* Sieverd. et al., *Blaszkowskia* G.A. Silva & Oehl

Type species: *Septoglomus constrictum* (Trappe) Sieverd.

Septoglomus Sieverd., G.A. Silva & Oehl, Mycotaxon 116: 105. 2011.

MycoBank MB 518436

Emended description: Spores formed in soil and sometimes in roots, terminally on or intercalary in hyphae, either singly, in loose spore clusters. Spores with one mono-to-multiple layered wall. SH wall conspicuously continuous and concolorous with the spore wall, or slightly lighter in color than the spore wall; SH constricted to cylindrical, rarely slightly funnel-shaped, or inflated in some distance to the spore base; straight, curved or flared; pore closed by a broad, pronounced septum or a plug at spore base or in some distance from spore; forming typical vesicular-arbuscular mycorrhiza, with mycorrhizal structures that stain blue to dark blue in trypan blue.

Type species: ***Septoglomus constrictum*** (Trappe) Sieverd., G.A. Silva & Oehl. Mycotaxon 116: 105. 2011.

MycoBank MB 518462

Basionym: *Glomus constrictum* Trappe, Mycotaxon 6: 361. 1977.

MycoBank MB 314589

≡ *Funneliformis constrictus* (Trappe) C. Walker & A. Schüssler, The Glomeromycota – a species list: 14. 2010.

MycoBank MB 542904

Blaszkowskia G.A. Silva & Oehl, Mycol. Progr. 22(11, no. 74): 5. 2023.

MycoBank MB 847414

Description: Spores light yellow to yellow brown to dark brown or dark red brown, generally 20–200 µm, with > 1 spore wall layer; SH funnel-shaped, rarely constricted, thick-walled; spore pores might be open at the spore base, but often closed by one to several thin septa within SH at some distance from the spore base. SH thicker-walled than mycelium hyphae and pigmented over long distances and within the whole spore clusters, including intraradical spore formations (> 100 µm).

Type species: ***Blaszkowskia deserticola*** (Trappe, Bloss & J.A. Menge) Oehl & G.A. Silva. Mycol. Progress 22 (11, no. 74): 5. 2023.

MycoBank MB 847415

Basionym: *Glomus deserticola* Trappe, Bloss & J.A. Menge, Mycotaxon 20: 123. 1984.

MycoBank MB 106847

≡ *Septoglomus deserticola* (Trappe, Bloss & J.A. Menge) G.A. Silva, Oehl & Sieverd. Mycotaxon 116: 106. 2011.

MycoBank MB 518463

Funneliglomus Corazon-Guivin, G.A. Silva & Oehl, Sydowia 71: 19. 2019.

MycoBank MB 829266

Description: Spores formed in soil or rarely in roots, terminally on or intercalary in hyphae, singly or in small clusters; the conspicuous SH is concolorous with spore wall color (or slightly lighter in color), SH is species-specific and funnel-shaped to slightly funnel-shaped, to rarely cylindrical or slightly constricted, or inflated in some distance to the spore base; straight, curved or flared. Pore regularly closed by a conspicuous septum that species-specifically may arise from the structural wall layer, from an additional adherent innermost, (semi-)flexible lamina, or from both but not by introverted wall thickening, which is lacking. Forming typical vesicular-arbuscular mycorrhiza, with mycorrhizal structures that stain blue to dark blue in trypan blue.

Type species: ***Funneliglomus sanmartinense*** Corazon-Guivin, G.A. Silva & Oehl. Sydowia 71: 21. 2019

MycoBank MB 829267 (*Funneliglomus sanmartinensis*), MycoBank MB 830216 (corrected to *Funneliglomus sanmartinense* by Mycobank after publication)

Funneliformis C. Walker & A. Schüssler, emend. Oehl, G.A. Silva & Sieverd., The Glomeromycota – a species list with new families and genera: 13. 2010.

MycoBank MB 542894

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Description: Spores formed within soil or rarely in roots, singly or sometimes in sporocarps with a few to several spores per sporocarp only; the conspicuous SH is concolorous with spore wall color (or slightly lighter in color), SH is species-specific and generally pronounced funnel-shaped to slightly funnel-shaped to rarely cylindrical. Wall differentiation and pigmentation may continue over long distances from the spore base (often > 50–250 µm), then mycelium may become hyaline. Pore regularly closed by a conspicuous septum that species-specifically arises from the structural wall layer, from an additional adherent innermost, (semi-)flexible lamina, or from both but not by introverted wall thickening, which is lacking. Forming typical vesicular-arbuscular mycorrhiza, with mycorrhizal structures that stain blue to dark blue in trypan blue.

Type species: ***Funneliformis mosseae*** (T.H. Nicolson & Gerd.) C. Walker & A. Schüssler, The Glomeromycota – a species list with new families and genera: 13. 2010.

MycoBank MB 542895

Basionym: *Endogone mosseae* T.H. Nicolson & Gerd., Mycologia 60: 314. 1968.

MycoBank MB 330367

≡ *Glomus mosseae* (T.H. Nicolson & Gerd.) Gerd. & Trappe, Mycol. Mem. 5: 40. 1974.

MycoBank MB 314604

Viscospora Sieverd., Oehl & G.A. Silva, Mycotaxon 116: 108: 2011.

MycoBank MB 518439

Emended description: Spores hyaline to white or subhyaline, terminally or intercalary on pronounced funnel-shaped to slightly funnel-shaped to cylindrical or inflated subtending hyphae, hyaline, singly, or in loose clusters with up to 100 spores per cluster. Outer layer is evanescent, viscose, gathering large amounts of debris during degradation. SWL2 is persistent, unite to laminate, and might stain whitish yellow to yellowish, when exposed to Melzer's reagent. SH straight or recurved, sometimes folded to rarely flared. Regularly, one to several straight to curved septa are formed in the SH close to the spore base or in a distance of up to 10–100 µm to the spore base. The spore wall layers continue in the SH, with similar to slightly smaller thickness towards the mycelium hyphae.

Type species: ***Viscospora viscosa*** (T.H Nicolson) Sieverd., Oehl & G.A. Silva. Mycotaxon 116: 108. 2011.

MycoBank MB 518471

Basionym *Glomus viscosum* T.H. Nicolson, Mycol. Res. 99: 1502. 1995.

MycoBank MB 413125

≡ *Septoglomus viscosum* (T.H. Nicolson) C. Walker, D. Redecker, D. Stiller & A. Schüßler: Mycorrhiza 23: 524. 2013

MycoBank MB 550089

Sclerocystaceae Oehl, G.A. Silva, & Sieverd. fam. nov.

MycoBank MB 855467

Description: Spores formed in loose sporocarps, in clusters, or singly in soil, and frequently in roots. When formed in compact sporocarps, spores can be randomly distributed on SH of different length, or well-organized around a central plexus of hyphae. SH wall continuous with the spore wall, and for a certain distance concolorous with the spore wall, or slightly lighter in color. SH cylindrical or seldom slightly funnel shaped at spore base. Pore at spore base open, or closed by a septum. Spore walls show rarely one, to generally two to three (and up to five) distinct layers, of which one or several of the outermost may separate when pressure is applied to spores. Forming vesicular-arbuscular mycorrhizae, whose fungal structures stain blue to dark blue in trypan blue.

Type genus: *Sclerocystis* Berk. & Broome

Other genera: *Oehlia* Błaszk. et al., *Rhizoglomus* Sieverd. et al., *Halonatospora* Błaszk. et al., *Silvaspora* Błaszk. et al., *Parvocarpum* Magurno

Sclerocystis Berk. & Broome, J. Linn. Soc., Bot. 14 (73 & 74): 137. 1873.

MycoBank MB20512

Emended description: Spores formed in compact sporocarps, in soils or roots, in cultures sometimes also singly or in loose clusters. When formed in compact sporocarps, then spores are

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regularly organized, arising radially around a central plexus of hyphae. SH wall continuous with the spore wall, and for a certain distance concolorous with the spore wall, or slightly lighter in color. SH funnel-shaped to cylindrical at spore base. Pore at spore base regularly closed by a fine septum, which is formed within the small pore channel within the spore base. Spore walls show more than one, and generally two or rarely > two distinct layers. Forming vesicular-arbuscular mycorrhizae, whose fungal structures stain blue to dark blue in trypan blue.

Type species: ***Sclerocystis coremioides*** Berk. & Broome, J. Linn. Soc., Bot. 14(no. 74): 137 (1873) [1875]

MycoBank MB 213141

≡ *Glomus coremioides* (Berk. & Broome) D. Redecker & J.B. Morton

MycoBank MB 464612

Rhizoglomus Sieverd., G.A. Silva & Oehl, Mycotaxon 129: 377. 2015.

MycoBank MB 803191

Description: Spores formed in loose sporocarps, in clusters, or singly, in soil and frequently also in roots. When formed in compact sporocarps, they are not organized around a central plexus of hyphae. SH wall continuous with the spore wall, and for a certain distance concolorous with the spore wall, or slightly lighter in color. SH cylindrical or seldom slightly funnel shaped at spore base. Pore at spore base regularly open, rarely closed by a septum. Spore walls show more than one, and generally two to three (and up to five) distinct layers, of which one or several of the outermost may separate when pressure is applied to spores. Forming vesicular-arbuscular mycorrhizae, whose fungal structures stain blue to dark blue in trypan blue.

Type species: ***Rhizoglomus intraradices*** (N.C. Schenck & G.S. Sm.) Sieverd., G.A. Silva & Oehl, Mycotaxon 129(2): 378 (2015) [2014]

MycoBank MB 803192

Basionym: *Glomus intraradices* N.C. Schenck & G.S. Sm., Mycologia 74: 78. 1982.

MycoBank MB 110704

≡ *Rhizophagus intraradices* (N.C. Schenck & G.S. Sm.) C. Walker & A. Schussler, The Glomeromycota: 19. 2010.

MycoBank MB 542910

Oehlia Błaszk., Kozłowska, Niezgoda, B.T. Goto & Dalpe, Nova Hedwigia 107: 507. 2018.

MycoBank MB 824689

Emended description: Spores formed in loose sporocarps, in clusters, or singly, in soil and frequently also in roots. When formed in compact sporocarps, they are not organized around a central plexus of hyphae. SH wall continuous with the spore wall, and for a certain distance concolorous with the spore wall, or slightly lighter in color. SH cylindrical or seldom slightly funnel shaped at spore base. Pore at spore base regularly closed by a septum. Spore wall shows more than one, and generally two to three (and up to five) distinct layers. Forming vesicular-arbuscular mycorrhizae, whose fungal structures stain blue to dark blue in trypan blue.

Type species: ***Oehlia diaphana*** (J.B.Morton & C.Walker) Błaszk., Kozłowska & Dalpe, Nova Hedwigia 107(3-4): 507 (2018)

MycoBank MB 824693

Basionym: *Glomus diaphanum* J.B.Morton & C.Walker. Mycotaxon 21: 433, 1984.

MycoBank MB 106161

Parvocarpum Magurno, MycoKeys 107: 283. 2024.

MycoBank MB 853558

Description: Producing glomoid spores mainly in small sporocarps, irregularly around a small central plexus of interwoven, intrasporocarpic hyphae, or singly in soils. SH generally short and cylindrical, often breaking at the spore base during sporocarp degradation. Pore closed by introverted wall thickening at the spore base, and additionally by a septum arising from the laminated wall layer and/or an additional flexible inner layer.

Type species: ***Parvocarpum badium*** (Oehl, Redecker & Sieverd.) Magurno, MycoKeys 107: 284. 2024

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MycoBank MB 853560

Basionym: *Glomus badium* Oehl, D. Redecker & Sieverd., J. Appl. Bot. Food Qual. 79: 39. 2005.

MycoBank MB 341387

≡ *Funneliformis badius* (Oehl, D. Redecker & Sieverd.) C. Walker & A. Schüssler, The Glomeromycota – a species list: 13. 2010.

MycoBank MB 541897

Silvaspora Błaszk., Niezgoda, B.T. Goto, Crossay & Magurno, Frontiers in Microbiology 12 (no. 655910): 14. 2021.

MycoBank 838881

Description: Forming pigmented spores with 2-3 wall layers, of which only layer 1, forming the spore surface, is impermanent and hyaline to brightly colored. SH colored similarly to the spore wall, cylindrical, slightly funnel-shaped, or constricted at the spore base, with a pore occluded due to thickening of that SH wall layer continuous with the laminate spore wall layer, rarely slightly open. Forming mycorrhiza with arbuscules, vesicles, and hyphae staining dark in trypan blue.

Type species: ***Silvaspora neocaledonica*** (D. Redecker, Crossay & Cilia) Błaszk., Niezgoda, B.T. Goto, Crossay & Magurno, Frontiers in Microbiology 12(no. 655910): 14 (2021)

MycoBank MB 838882

≡ *Rhizoglomus neocaledonicum* (D. Redecker, Crossay & Cilia) Oehl, Turrini & Giovann.

MycoBank MB 827095

Basionym: *Rhizophagus neocaledonicus* D. Redecker, Crossay & Cilia, Mycological Progress 17: 739. 2018.

MycoBank MB 820537

Halonatospora Błaszk., Niezgoda, B.T. Goto & Kozłowska, Botany 96(11): 743. 2018.

MycoBank MB 826963

Description: Spores formed in loose clusters, or singly, in soil and frequently in roots. When formed in sporocarps, they are not organized around a central plexus of hyphae. SH wall continuous with the spore wall, and for a certain distance concolorous with the spore wall, or slightly lighter in color. Outer spore wall layer strongly swells in PVLG, forming a halo with radiate columns. SH cylindrical or slightly flared, sometimes slightly constricted at spore base, straight or slightly curved. Pore open or closed by a septum at spore base. Spore wall shows more than one, and generally two to three (and up to five) distinct layers. Forming vesicular-arbuscular mycorrhizae, whose fungal structures stain blue to dark blue in trypan blue.

Type species: ***Halonatospora pansihalos*** (S.M. Berch & Koske) Błaszk., Niezgoda, B.T. Goto & Kozłowska, Can. J. Bot. 96(11): 743 (2018)

MycoBank MB 826964

Basionym: *Glomus pansihalos* S.M. Berch & Koske. Mycologia 78: 832, 1986.

MycoBank MB 358213

Kamienskiaceae G.A. Silva, Sieverd. & Oehl

MycoBank MB 855468

Description: Spores hypogeous and/or intraradical, produced in loose clusters to compact sporocarps, and never organized around a central plexus of hyphae. Spores hyaline, small, up to 50 µm diam. when globose. Spore wall with two permanent, smooth layers, of which one may stain in Melzer's reagent. The structural laminate layer is layer 1 or 2. SH cylindrical to funnel-shaped with an open pore. Forming vesicular-arbuscular mycorrhizae staining dark in trypan blue.

Type genus: *Kamienskia* Błaszk. et al.,

Other genus: *Microkamienskia* Corazon-Guivin, G.A. Silva & Oehl, *Epigeocarpum* Błaszk., B.T. Goto, Jobim, Niezgoda & Marguno

Kamienskia Błaszk., Chwat & Kovács, Nova Hedwigia 100(1-2): 230. (2014) [2015].

MycoBank MB 808260

Emended description: Spores hypogeous and/or intraradical, produced in loose clusters to compact, but fast degrading sporocarps, but never organized around a central plexus of hyphae. Spores hyaline, small, up to 50 µm diam. when globose. Spore wall with two permanent, smooth

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layers. The structural laminate layer might be layer 1 or 2. SH regularly funnel-shaped with an open pore. Forming vesicular-arbuscular mycorrhizae staining dark in trypan blue.

Type species: ***Kamienskia bistrata*** (Błaszk., D. Redecker, Koegel, Symanczik, Oehl & Kovács) Błaszk., Chwat & Kovács, Nova Hedwigia 100(1-2): 230. (2014) [2015].

MycoBank MB 808261

Basionym: *Glomus bistratum* Błaszk., D. Redecker, Koegel, Symanczik, Oehl & Kovács, Botany 87: 267. 2009.

MycoBank MB 512540

Microkamienskia Corazon-Guivin, G.A. Silva & Oehl, Nova Hedwigia 109: 359. 2019.

MycoBank MB 830814

Description: Spores hypogeous and/or intraradical, produced in loose clusters to compact, but fast degrading sporocarps, but never organized around a central plexus of hyphae. Spores hyaline, small, up to 50 μm diam. when globose. Spore wall with two permanent, smooth layers, of which one may stain in Melzer's reagent. The structural laminate layer might be layer 1 or 2. SH cylindrical to rarely slightly funnel-shaped with an open pore. Forming vesicular-arbuscular mycorrhizae staining dark in trypan blue.

Type species: ***Microkamienskia perpusilla*** (Błaszk. & Kovács) Corazon-Guivin, G.A. Silva & Oehl. Nova Hedwigia 109: 361. 2019.

MycoBank MB 830815

Basionym: *Glomus perpusillum* Błaszk. & Kovács. Mycologia 101: 249. 2009.

MycoBank MB 512346

≡ *Kamienskia perpusilla* (Błaszk. & Kovács) Błaszk., Chwat & Kovács. Nova Hedwigia 100: 231. 2015-

MycoBank MB 808264

Epigeocarpum Błaszk., B.T. Goto, Jobim, Niezgoda & Marguno, Frontiers in Microbiology 12 (no. 655910): 10. 2021.

MycoBank MB 838879

Description: Spores hyaline to light yellow, formed in compact, unorganized sporocarps. Spores have 2–3 wall layers, laminate layer usually transferring into a crown-like structure due to contracting in spores crushed in PVLG and PVLG + Melzer's reagent. The channel connecting the lumen of the SH with the interior of spores is closed by a septum usually positioned at half the thickness of the laminate layer; SH funnel-shaped, SH lumen gradually narrowing in maturing spores due to internal thickening of the laminate wall layer.

Type species: ***Epigeocarpum crypticum*** Jobim, Błaszk., Niezgoda, Magurno & B.T. Goto, Frontiers Microbiol. 12(no. 655910): 14. 2021.

MycoBank MB 838880

Dominikiaceae G.A. Silva, Sieverd. & Oehl

MycoBank MB 855469

Description: Spores hypogeous and/or intraradical, produced in loose clusters to compact sporocarps, but never organized around a central plexus of hyphae. Spores hyaline to creamy or yellow to yellow brown; spore wall with one to regularly two, or > 2 layers. SH wall continuous with the spore wall, and for a certain distance concolorous with the spore wall, or slightly lighter in color. Spore pore often closed at spore base by a thin bridging septum, sometimes supported by a certain degree of introverted wall thickening, rarely continuing in the SH for a short distance (2-10 μm). Forming vesicular-arbuscular mycorrhizae staining dark in trypan blue.

Type genus: *Dominikia* Błaszk. et al.,

Other genera: *Macrodominikia* Oehl et al., *Microdominikia* Oehl et al., *Nanoglomus* Corazon-Guivin et al., *Orientoglomus* G.A. Silva et al.

Dominikia Błaszk., Chwat & Kovács, Nova Hedwigia 100(1-2): 228. (2014) [2015].

MycoBank MB 808255

Emended description: Spores hypogeous and/or intraradical, produced in loose clusters to compact sporocarps but never organized around a central plexus of hyphae. Spores hyaline to creamy

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or yellow to yellow brown, small, up to 70 µm diam; spore wall with one to regularly two, or > 2 layers. SW wall continuous with the spore wall, and for a certain distance concolorous with the spore wall, or slightly lighter in color. Spore pore often closed at spore base by a thin bridging septum, sometimes supported by a certain degree of introverted wall thickening, rarely continuing in the SH for a short distance. Forming vesicular-arbuscular mycorrhizae staining dark in trypan blue.

Type species: ***Dominikia minuta*** (Błaszk., Tadych & Madej) Błaszk., Chwat & Kovács, Nova Hedwigia 100: 230. (2014) [2015].

MycoBank MB 808256

Basionym: *Glomus minutum* Błaszk., Tadych & Madej. Mycotaxon 76: 189. 2000.

Macrodominikia Oehl, Sieverd. & G.A. Silva, gen. nov.

MycoBank 855470

Description: Spores hypogeous and/or intraradical, produced singly or in loose clusters to loose sporocarps but never organized around a central plexus of hyphae. Spores creamy to yellow to yellow brown, generally 70-110 µm diam; spore wall with one to regularly two, or > 2 layers. SH wall continuous with the spore wall, and for a certain distance concolorous with the spore wall, or slightly lighter in color. Spore pore often closed at spore base by a thin bridging septum, supported by a large degree of introverted wall thickening, continuing in the SH for 10-50 µm distance, forming an irregular, tortuous pore channel. Forming vesicular-arbuscular mycorrhizae staining dark in trypan blue.

Etymology: Macro-, -dominikia, referencing to the spores, which are larger than known hitherto for other genera in the Dominikiaceae

Type species: ***Macrodominikia compressa*** (Sieverd., Oehl, Palenz., Sánchez-Castro & G.A. Silva) Oehl, Siev. & G.A. Silva. comb. nov.

MycoBank MB 855471

Basionym: *Glomus compressum* Błaszk., Tadych & Mad Sieverd., Oehl, Palenz., Sánchez-Castro & G.A. Silva. Nova Hedwigia 99: 433. 2014.

MycoBank MB 807530

≡ *Dominikia compressa* (Sieverd., Oehl, Palenz., Sánchez-Castro & G.A. Silva) Oehl, Palenz., Sánchez-Castro & G.A. Silva. Nova Hedwigia. 101: 71. (2014) [2015]

MycoBank MB 809861

Microdominikia Oehl, Corazon-Guivin & G.A. Silva, Mycological Progress 18(12): 1400. 2019.

MycoBank MB 831098

Description: Spores formed singly or in clusters in soils or rarely in roots, terminally or intercalary on SH, globose to subglobose, generally hyaline to subhyaline, 10–45 µm in diameter, rarely egg-shaped or irregular, 25–50 × 20–35 µm, with a permanent outer spore wall layer. SH straight or recurved, usually funnel-shaped, more rarely cylindrical, rarely slightly constricted at the spore base. Spore pores open at the spore base, but closed within SH at some distance.

Type species: ***Microdominikia litorea*** (Błaszk. & Kozłowska) Oehl, Corazon-Guivin & G.A. Silva, Mycological Progress 18 (12): 1400 (2019)

MycoBank MB 831099

Basionym: *Dominikia litorea* Błaszk. & Kozłowska, Phytotaxa 338(3): 246 (2018)

MycoBank MB 823832

Nanoglomus Corazon-Guivin, G.A. Silva & Oehl, Mycological Progress 18(12): 1398. 2019.

MycoBank MB 831096

Description: *Nanoglomus* species differentiate small spores, generally <40 µm in diameter, when globose, singly or in loose clusters, terminally or intercalary on cylindrical to slightly funnel-shaped, rarely inflating SH, which are concolorous with the spores and have a fine, straight, or rarely curved septum closing the pore at the spore base. So far, they can be differentiated from *Dominikia* spp. by the generally smaller and thinner walled spores, and by molecular phylogeny on the partial SSU, ITS region, and partial LSU rDNA.

Type species: ***Nanoglomus plukenetiae*** Corazon-Guivin, G.A. Silva & Oehl, Mycological Progress 18 (12): 1398 (2019)

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MycoBank MB 831097.

Orientoglomus G.A. Silva, Oehl & Corazon-Guivin, Mycological Progress 18(12): 1400. 2019.

MycoBank MB 831100

Description: Glomeraceae spores formed singly or in clusters in soil or rarely in roots, terminally on SH, globose to subglobose, 40–90 µm in diameter, rarely egg-shaped, 50–100 × 40–85 µm; SH wall with significant and regular thickening, resulting in a cylindrical pore channel. Outermost spore wall layer permanent, smooth, at least one spore wall layer laminate. Pore open or closed by a straight to curved to septum.

Type species: ***Orientoglomus emiratium*** (Błaszk., Kozłowska, Mullath, AlDhaheri & Al-Yahya'ei) G.A. Silva, Oehl & Corazon-Guivin, Mycological Progress 18 (12): 1403 (2019)

MycoBank MB 831101.

Basionym: *Dominikia emiratia* Błaszk., Kozłowska, Mullath, AlDhaheri & Al-Yahya'ei, Botany 95(7): 632 (2017)

MycoBank MB 819815

4. Discussion

In the present study, the family Glomeraceae of the order Glomerales was divided into five different families, based on five different phylogenetic major clades, well known and increasingly elaborated within the last fifteen years of research on the ribosomal gene. Also in this study, the clades "B", "C", "D" and "E" were very well supported by BI and ML analyses, using different datasets (except, clade "B", supported by ML just in the first tree) to generate the trees and indicate very stable taxa. The clade "A" was supported with good values just in the first tree (Figure 1). The clade "A" formed a more heterogeneous group, and we believe that in the future this group will be divided in different families. For now, we understand that the better solution is maintain this group as a single family in Glomerales, mainly because the support in the first tree was very strong. These same clades were found in the tree generated by Corazon-Guivin et al. [21] and Tedersoo et al. [6]. The families Glomeraceae, Dominikiaceae, Septoglomeraceae and Sclerocystaceae are also supported by Corazon-Guivin et al. (19,20).

The genera *Oehlia* and *Halonatospora* form two other clades well separated from *Silvaspora*, *Sclerocystis* and *Rhizoglomus* and might represent two further families within the Glomerales. Such deeper-going separation is to our opinion fully supported by molecular phylogeny, but in view of the low numbers of taxa, this separation is also not yet needed at this stage of research progress. In our tree, *Halotospora* was placed near *Oehlia*, *Silvaspora*, *Sclerocystis* and *Rhizoglomus*. However, according to the tree from Tedersoo et al. [6], *Halonatospora* is near *Kamienskia*, *Microkamienskia* and *Epigeocarpum*. The tree generated by Tedersoo et al. [6] has different orders from Glomeromycota. The position of some genera can be divergent in relation to our trees from Glomerales, which confirms also the lower degree of relationship between *Halonatospora*, *Oehlia* and the other genera clustering together within the Sclerocystaceae.

Morphologically, all genera listed within Glomerales, can clearly be identified based on the combination of their major sporocarp, spore, pore closure, subtending hyphae and spore formation characteristics (see also Silva et al. [5], including an identification key for all genera within the Glomerales). However, attribution to a distinct family is not unequivocally possible, since none of the major morphological characteristics are unique for one of the five families separated here. Some simple indications however, might at least give hints to the family attribution, which is the genus identification itself. Otherwise, our simplified supplementary material (Table S1), shall help to differentiate and attribute fast a specific morphotype to a specific genus and thus to the correct family, to which the morphotype and the genus belongs to. In this study, we accordingly transferred three *Glomus* species to the genus *Sclerocarpum*, based on their unique morphological characteristics, fitting exactly to the latter genus. For other Glomeraceae species, new combination would also be justified, either based on molecular phylogeny or morphology of the 'fruit bodies', including sporocarps, spores, and their subtending hyphae, but at this stage there is no urgent need to do so.

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The genus *Parvocarpum* [6], was not placed in our analyses because the only sequence available for the *P. badium* (the type genus) represent a SSU fragment (564bp) and 53bp of ITS1. We conclude that this genus was described mainly based on the high quantity of environmental sequences, which could be attributed to this short SSU fragment [6]. Solely for morphological reasons, there were already sufficient arguments to describe this genus, when the species *P. badium* was originally described, but at that time the phylogenetic resolution was not given [34], and the most related genus *Sclerocystis* had been synonymized with *Glomus* just a few years before, instead of clearly separated.

In our first tree (Figure 1), *Macroominikia compressa* is not grouped with other *Dominikia* spp. In the second tree (Figure 2), *M. compressa* forms a clade with other *Dominikia* spp., but with no support. According to Tedersoo et al. [6], this species represents a new genus, which is confirmed here and also supported by its distinct spore morphology [5,34]. By including environmental sequences to reliable spore and sequence identifications [36], most probably more genera and even families can be separated, since the phylogenetic resolution will further increase. *Rhizoglomus* spp. for instance might then be separated from Sclerocystaceae. This separation is already suggested by our phylogenetic trees.

Blaszkowskia was invalidated by Tedersoo et al. [6]. The tree generated by these authors present two different major clades for *Viscospora* sensu Terderso et al. [6], one clade is divided in two subclades, the first with one sequence from *B. deserticola* and one environmental sequence and the second with one sequence from *V. viscosa* and six environmental sequences. The second clade has just environmental sequences (28 sequences) and for us can be a new different genus represented by these sequences. Thus, to our opinion, *Blaszkowskia* continues to be a valid genus. For morphological reasons, the genera *Viscospora* and *Blaszkowskia*, can be easily separated and continued [5].

During the first stages of molecular phylogenetic inclusion into the taxonomy and classification, there were several major synonymizations of AMF genera, also in the Glomeraceae, even for genera with a distinct spore morphology, followed by a time of doubts and resurrections of genera such as *Sclerocystis*. In the meantime, molecular phylogeny has become the driving force to separate taxa, especially on the genus and species levels. So far, morphology still finds arguments to separate such taxa. With the expected inclusion of environmental sequences directly into taxonomy, morphological identification might lose its importance and significance, and taxa numbers might strongly increase in Glomeromycota, and especially in Glomerales. However, then researchers experienced in morphological identification will start to find the morphs behind such environmental-sequences-born, exclusively molecular-based taxa.

Supplementary Materials: The following supporting information can be downloaded at: www.mdpi.com/xxx/s1, Spreadsheet S1: GenBank accession numbers for the sequences used in this study; Table S1: Main morphological characters of the families and genera within the order Glomerales.

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Data Availability Statement: All data (sequences) presented in this article are in GenBank (https://www.ncbi.nlm.nih.gov)

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References

1. Baltruschat, H.; Santos, V.M.; Silva, D.K.A.; Schellenberg, I.; Deubel, A.; Sieverding, E.; Oehl, F. Unexpectedly high diversity of arbuscular mycorrhizal fungi in fertile Chernozem croplands in Central Europe. *Catena* **2019**, 182, 104135. https://doi.org/10.1016/j.catena.2019.104135
2. Wijayawardene, N.N.; Hyde, K.D.; Al-Ani, L.K.T.; Tedersoo, L.; Haelewaters, D.; Becerra, A.G.; Thines, M. et al. Outline of fungi and fungus-like taxa. *Mycosphere* **2020**, 11: 1060–1456. https://doi.org/10.5943/mycosphere/11/1/8
3. Wijayawardene, N.N.; Hyde, K.D.; Dai, D.Q.; Sánchez-García, M.; Goto, B.T.; Saxena, R.K.; Erdoğdu, M, et al. Outline of fungi and fungus-like taxa - 2021. *Mycosphere* **2022,** 13, 53–453. https://doi.org/10.5943/mycosphere/13/1/2
4. Błaszkowski, J.; Sánchez-García, M.; Niezgoda, P.; Zubek, S.; Fernández, F.; Vila, A.; Al-Yahya'ei, M.N.; Symanczik, S.; Milczarski, P.; Malinowski, R.; Cabello, M.; Goto, B.T.; Casieri, L.; Malicka, M.; Bierza, W.; Magurno, F. A new order, Entrophosporales, and three new *Entrophospora* species in Glomeromycota. *Front. Microbiol.* **2022,** 13, 962856. https://doi.org/10.3389/fmicb.2022.962856.
5. Silva, G.A.; Corazon-Guivin, M.A.; Assis, D.M.A.; Oehl, F. *Blaszkowskia*, a new genus in Glomeraceae. *Mycol. Prog.* 2023, 22, 74. https://doi.org/10.1007/s11557-023-01919-z
6. Tedersoo, L.; Magurno, F.; Alkahtani, S.; Mikryukov, V. Phylogenetic classification of arbuscular mycorrhizal fungi: new species and higher-ranking taxa in Glomeromycota and Mucoromycota (class Endogonomycetes). *MycoKeys* **2024**, 107, 249–271. https://doi.org/10.3897/mycokeys.107.125549
7. Morton, J.B.; Benny, G.L. Revised classification of arbuscular mycorrhizal fungi (Zygomycetes): a new order, Glomales, two new suborders, Glomineae and Gigasporineae, and two new families, Acaulosporaceae and Gigasporaceae, with an emendation of Glomaceae. *Mycotaxon* **1990**, 37, 471–491.
8. Schüßler, A.; Schwarzott, D.; Walker, C. A new fungal phylum, the Glomeromycota: phylogeny and evolution. *Mycol. Res.* **2001,** 105, 1413–1421. https://doi.org/10.1017/S0953756201005196
9. Oehl, F.; Silva, G.A.; Goto, B.T.; Sieverding, E. Glomeromycota: three new genera, and glomoid species reorganized. *Mycotaxon* **2011**, 116, 75–120. https://doi.org/10.5248/116.75
10. Silva, G.A.; Maia, L.C.; Oehl, F. Phylogenetic systematics of the Gigasporales. *Mycotaxon* **2013,** 122, 207–220. https://doi.org/10.5248/122.207
11. Błaszkowski, J.; Yamato, M.; Niezgoda, P.; Zubek, S.; Milczarski, P.; Malinowski, R.; Goto, B.T.; Uszok, S.; Casieri, L.; Magurno, F. A new genus, *Complexispora*, with two new species, *C. multistratosa* and *C. mediterranea*, and *Epigeocarpum japonicum* sp. nov. *Mycol. Prog.* **2023**, 22, 1–15. https://doi.org/10.1007/s11557-023-01882-9
12. Willis, A.; Błaszkowski, J.; Prabhu, T.; Chwat, G.; Góralska, A.; Sashidhar, B.; Harris, P.; D'Souza, J.; Vaingankar, J.; Adholeya, A. *Sacculospora felinovii*, a novel arbuscular mycorrhizal fungal species (Glomeromycota) from dunes on the west coast of India. *Mycol. Prog.* **2016**, 15, 791-798. https://doi.org/10.1007/s11557-016-1208-6
13. Błaszkowski, J.; Kozłowska, A.; Crossay, T.; Symanczik, S.; Al-Yahya'ei, M.N. A new family, Pervetustaceae with a new genus, *Pervetustus*, and *P. simplex* sp. nov. (Paraglomerales), and a new genus, *Innospora* with *I. majewskii* comb. nov. (Paraglomeraceae) in the Glomeromycotina. *Nova Hedwigia* **2017**, 105, 397-410. https://doi.org/10.1127/nova_hedwigia/2017/0419
14. Lin, T.C.; Silva, G.A.; Oehl, F. *Acaulospora tsugae*, a new species in the Glomeromycetes from Taiwan, and a key to species in Acaulosporaceae. *Nova Hedwigia* **2019,** 108, 475–488. https://doi.org/10.1127/nova_hedwigia/2018/0513
15. Oehl, F.; Souza, F.A.; Sieverding, E. Revision of *Scutellospora* and description of five new genera and three new families in the arbuscular mycorrhiza-forming Glomeromycetes. *Mycotaxon* **2008,** 106, 311–360.
16. Goto, B.T.; Silva, G.A.; Assis, D.M.A.; Silva, D.K.A.; Souza, R.G.; Ferreira, A.C.A.; Jobim, K.; Mello, C.M.A.; Vieira, H.E.E.; Maia, L.C.; Oehl, F. Intraornatosporaceae, (Gigasporales), a new family with two new genera and two new species. *Mycotaxon* **2012,** 119, 117–132. https://doi.org/10.5248/119.117
17. Błaszkowski, J.; Kozłowska, A.; Niezgoda, P.; Goto, B.T.; Dalpé, Y. A new genus, *Oehlia* with *Oehlia diaphana* comb. nov. and an emended description of *Rhizoglomus vesiculiferum* comb. nov. In the Glomeromycotina. Nova Hedwigia **2018**, 107, 501–518. https://doi.org/10.1127/nova_hedwigia/2018/0488
18. Jobim, K.; Błaszkowski, J.; Niezgoda, P.; Kozłowska, A.; Zubek, S.; Mleczko, P.; Chachuła, P.; Ishikawa, N.K.; Goto, B.T. New sporocarpic taxa in the phylum Glomeromycota: *Sclerocarpum amazonicum* gen. et sp. nov. in the family Glomeraceae (Glomerales) and *Diversispora sporocarpia* sp. nov. in the Diversisporaceae (Diversisporales). *Mycol. Prog.* **2019,** 18, 369-384. https://doi.org/10.1007/s11557-018-01462-2
19. Corazon-Guivin, M.A.; Cerna-Mendoza, A.; Guerrero-Abad, J.C.; Vallejos-Tapullima, A.; Carballar-Hernández, S.; Silva, G.A.; Oehl, F. *Nanoglomus plukenetiae*, a new fungus from Peru, and a key to small-spored Glomeraceae species, including three new genera in the "*Dominikia* complex/clades". *Mycol. Prog.* **2019,** 18, 1395–1409. https://doi.org/10.1007/s11557- 019- 01522-1

20. Corazon-Guivin, M.A.; Mendoza, A.C.; Guerrero-Abad, J.C.; Vallejos-Tapullima, A.; Carballar-Hernández, S.; Silva, G.A.; Oehl, F. *Funneliglomus*, gen. nov., and *Funneliglomus sanmartinensis*, a new arbuscular mycorrhizal fungus from the Amazonia region in Peru. *Sydowia* **2019**, 71, 17–24. https://doi.org/10.12905/0380. sydowia71-2019- 0017
21. Corazon-Guivin, M.A.; Mendoza, A.C.; Guerrero-Abad, J.C.; Vallejos-Tapullima, A.; Carballar-Hernández, S.; Silva, G.A.; Oehl, F. *Microkamienskia* gen. nov. and *Microkamienskia peruviana*, a new arbuscular mycorrhizal fungus from Western Amazonia. *Nova Hedwigia* **2019**, 109, 355–368. https://doi.org/10.1127/nova_ hedwigia/2019/0551
22. Bhunjun, C.S.; Chen, Y.J.; Phukhamsakda, C.; Boekhout, T.; Groenewald, J.Z.; Mckenzie, E.H.C.; Crous, P. W. et al. What are the 100 most cited fungal genera? *Studies in mycology* **2024**, 108, 1-412. https://doi.org/10.3114/sim.2024.108.01
23. Katoh, K.; Rozewicki, J.; Yamada, K.D. MAFFT online service: multiple sequence alignment, interactive sequence choice and visualization. *Brief Bioinformatics* **2019,** 20, 1160–1166. https://doi.org/10.1093/bib/bbx108
24. Milne, I.; Wright, F.; Rowe, G.; Marshal, D.F.; Husmeier, D.; McGuire, G. TOPALi: Software for automatic identification of recombinant sequences within DNA multiple alignments. *Bioinformatics* **2004**, 20, 1806–1807. https://doi.org/10.1093/bioinformatics/bth155
25. Ronquist, F.; Huelsenbeck, J.P. MrBayes 3: Bayesian phylogenetic inference under mixed models. *Bioinformatics* **2003,** 19: 1572–1574. https://doi.org/10.1093/bioinformatics/btg180
26. Guindon, S.; Gascuel, O. A simple, fast, and accurate algorithm to estimate large phylogenies by maximum likelihood. *System Biol.* **2003,** 52, 696–704. https://doi.org/10.1080/10635150390235520
27. Hall, I.R.; Abbott, L.K. *Photographic slide collection illustrating features of the Endogonaceae*. 3rd edition. Invery Agricultural Research Centre and Soil Science Department, University of Western Australia, Australia, plus 400 color transparencies, 1979**;** pp. 1–27**.**
28. Brundrett, M.; Melville, L.; Peterson, L. *Practical methods in mycorrhizal research*; Mycologue Publications, University of Guelph: Guelph, Canada, 1994**.**
29. Walker, C. Taxonomic concepts in the Endogonaceae: spore wall characteristics in species descriptions. *Mycotaxon* **1983**, 18, 443–455.
30. Stürmer, S.L.; Morton, J.B. Developmental patterns defining morphological characters in spores of four species in *Glomus*. *Mycologia* **1997,** 89, 72-81. https://doi.org/10.1080/00275514.1997.12026756
31. Gerdemann, J.W.; Trappe, J.M. The Endogonaceae in the Pacific North West. *Mycologia Memoirs* **1974**, 5, 1-76
32. Trappe, J.M. *Glomus segmentatus* sp. nov. *Trans. Br. Mycol. Soc.* **1979**, 73, 361-362.
33. McGee, P.A.; Trappe, J.M. The Australian zygomycetous mycorrhizal fungi. II. Further Australian sporocarpic Glomaceae. *Aust. Syst. Bot.* **2002,** 15, 115-124. https://doi.org/10.1071/SB00038
34. Oehl, F.; Redecker, D.; Sieverding, E. *Glomus badium*, a new sporocarpic mycorrhizal fungal species from European grasslands with higher soil pH. *J. Appl. Bot. Food. Qual.* **2005,** 79, 38-43.
35. Oehl, F.; Sanchez-Castro, I.; Palenzuela, J.; Silva, G.A.; Sieverding, E. *Glomus compressum*, a new arbuscular mycorrhizal fungus from different agro-ecosystems in central europe. *Nova Hedwigia* **2014**, 99, 429-439. https://doi.org/10.1127/0029-5035/2014/0200
36. Delavaux, C.S.; Ramos, R.J.; Stürmer, S.L.; Bever, J.D. An updated LSU database and pipeline for environmental DNA identification of arbuscular mycorrhizal fungi. *Mycorrhiza* **2024**, 34, 369–373. https://doi.org/10.1007/s00572-024-01159-3