STUDIES IN MYCOLOGY 50: 489–515. 2004.

Ribosomal gene phylogeny and species delimitation in *Geotrichum* and its teleomorphs

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Abstract: A taxonomic revision is presented of all filamentous *Hemiascomycetes* that reproduce with predominantly arthric conidiogenesis. On the basis of SSU rDNA data, two widely divergent groups (1 and 2) are known to exist. Both are distantly related to the *Hemiascomycetes*, and show remarkable diversity in ITS rDNA, leading to the supposition that phylogenetically ancient fungi are concerned. The teleomorph / anamorph genera occurring in Groups 1 vs. 2 are classified in (1) *Galactomyces* and *Dipodascus* with *Geotrichum* anamorphs, vs. (2) *Magnusiomyces* with *Saprochaete* anamorphs. Taxonomy at the species level is based on ITS rDNA sequences and nDNA/DNA reassociation data. In total, 32 taxa are recognized. The phenetic data set applied is nutritional physiology. A key to the species is provided.

Taxonomic novelties: *Saprochaete chiloënsis* (Ramírez & González) Kurtzman, Robnett, de Hoog & M.Th. Smith comb. nov., *S. clavata* (de Hoog *et al.*) de Hoog & M.Th. Smith comb. nov., *S. suaveolens* (Krzemecki) de Hoog & M.Th. Smith comb. nov., *S. fungicola* de Hoog & M.Th. Smith sp. nov., *S. ingens* (van der Walt & van Kerken) de Hoog & M.Th. Smith sp. nov., *S. japonica* de Hoog & M.Th. Smith sp. nov., *S. ludwigii* (Hansen) de Hoog & M.Th. Smith comb. nov., *S. psychrophila* de Hoog & M.Th. Smith sp. nov., *S. quercus* de Hoog & M.Th. Smith sp. nov., *S. capitata* (Diddens & Lodder) de Hoog & M.Th. Smith comb. nov., *S. sericea* (Stautz) de Hoog & M.Th. Smith comb. nov., *S. gigas* (J. Smit & L. Meyer) de Hoog & M.Th. Smith comb. nov., *Magnusiomyces capitatus* (de Hoog *et al.*) de Hoog & M.Th. Smith comb. nov., *M. magnusii* (Ludwig) de Hoog & M.Th. Smith comb. nov., *M. ovetensis* (Pelãez & Ramírez) de Hoog & M.Th. Smith comb. nov., *M. spicifer* (de Hoog *et al.*) de Hoog & M.Th. Smith comb. nov., *M. starmeri* (Phaff *et al.*) de Hoog & M.Th. Smith comb. nov., *M. tetrasperma* (Macy & Miller) de Hoog & M.Th. Smith comb. nov., *M. ingens* de Hoog & M.Th. Smith comb. nov., *Galactomyces pseudocandidus* de Hoog & M.Th. Smith sp. nov., *Gal. candidus* de Hoog & M.Th. Smith sp. nov., *Geotrichum europaeum* de Hoog & M.Th. Smith sp. nov., *Geo. restrictum* de Hoog & M.Th. Smith sp. nov.

Key words: ancient organisms, *Dipodascus*, *Galactomyces, Geotrichum, Magnusiomyces, Saprochaete, Hemiascomycetes*, phylogeny, ribosomal genes, taxonomy, yeast-like fungi.

INTRODUCTION

The taxonomy of the genus of filamentous yeast-like fungi, *Geotrichum* Link : Fr. (*Hemiascomycetes*) has been studied in depth by Smith and co-workers in a series of publications (de Hoog *et al*. 1986, Smith *et al*. 1995, 2000, Smith & Poot 1998, 2003, Naumov *et al*. 1999, Naumova *et al*. 2001). A review of the taxonomy of the fungi concerned was presented in Kurtzman & Fell (1998). Techniques used for genus-wide comparisons were morphology, physiology, cultural characteristics, nDNA reassociation, mol % G+C of genomic DNA and properties of the thermal denaturation curve, and mating type systems, while for smaller species complexes electrophoretic karyotyping, PCR-fingerprinting and multilocus enzyme electrophoresis were applied. At present, a total of 21 species are recognized, of which 13 have a teleomorph in *Dipodascus* de Lagerh., three in *Galactomyces* Redhead & Malloch, and for five the teleomorph is unknown.

Galactomyces comprises the generic type species of *Geotrichum*, *Geo. candidum* Link : Fr. (de Hoog *et al*. 1986). *Geotrichum* is morphologically characterized by the presence of arthroconidia that are liberated schizolytically in random order. Septal walls are perforated by micropores. In some species, such as *Geo. fermentans* (Diddens & Lodder) Arx and *Geo. capitatum* (Diddens & Lodder) Arx, additional holoblastic conidia are observed. Some species of the *Geo. capitatum* complex are remarkable by having blastic conidia arranged in a profusely branched conidial apparatus, each cell producing an elongate, regularly sympodial rachis. For this reason Salkin *et al*. (1985) classified *Geo. capitatum* in a separate genus, *Blastoschizomyces* Salkin *et al*. However, since similar though less pronounced structures are found in numerous *Dipodascus* species, this classification was not generally accepted (de Hoog *et al*. 2000).

The bipartition of *Geotrichum* along lines of their teleomorph morphology, either *Dipodascus* or *Galactomyces*, recently appeared not to be supported by ribosomal DNA data. Kurtzman & Robnett (1998)

found a bipartition in 26S rDNA sequence data, which grouped *Galactomyces* with several *Dipodascus* species far apart from the remainder of *Dipodascus*. A similar bipartition was reflected in 18S rDNA sequence data (Ueda-Nishimura & Mikata 2000), where a *Galactomyces* clustered as Group 1 with six *Dipodascus* species (including the generic type species, *D. albidus* de Lagerh.), while six remaining *Dipodascus* species (Group 2) were remote from this cluster. In addition, the SSU molecule of members of Group 2 appeared to be remarkable in the fungal Kingdom by comprising 1634–1636 bp only, as a result of large deletions in the V2, V3 and V8 variable domains. In both publications (Kurtzman & Robnett 1998, Ueda-Nishimura & Mikata 2000) it was noted that the distances between species were unexpectedly large, although de Hoog *et al*. (1986) has stressed the morphological and ecological unity of *Dipodascus*, regarding the pronounced phenotypes as variations on a single, basic theme. These remarkable findings and conflicts in visions prompted an in depth study of ribosomal sequence comparison comprising 18S, 26S and ITS regions of the molecule, with the aim to compare the results with those from nDNA reassociation, and morphological and physiological data generated during previous studies.

MATERIAL AND METHODS

List of strains

All strains examined previously and in this study, are presented in Table 1. The order is according to the phylogenetic relatedness (Ueda-Nishimura & Mikata 2000) and the taxonomy of the present study.

nDNA reassociation

Strains were grown for 2 d at 25 °C on a rotary shaker at 125 rpm in 2 L YM broth (Wickerham 1951) using 1 L flat-bottom flasks. Isolation and purification of DNA and determination of DNA base composition were done according the procedures cited before (Smith *et al*. 1995). DNA-DNA hybridization experiments were carried out according to the procedures described by Seidler & Mandel (1971) and modified by Kurtzman *et al.* (1980).

DNA extraction

About 1 g of mycelium was transferred to a 2 : 1 mixture of silicagel and Celite 545 with 300 µL cetyltrimethylammoniumbromide (CTAB) buffer added [Tris·HCl, 200 mM, pH 7.5; Na-EDTA (ethylenediaminotetraacetic acid sodium salt), 200 mM; NaCl 8.2 %; CTAB 2 %]. The material was ground with a micropestle (Eppendorf). After adding 200 µL CTAB-buffer and vigorously shaking the sample was incubated for 10 min in a 65 °C water bath; 500 µL chloroform was added, vortexed shortly and centrifuged for 5 min at 14000 r.p.m. After transferring the aqueous supernatant to a new Eppendorf tube, 2 volumes (~800 µL) ethanol 96 %, –20 °C were added and mixed gently. The DNA was precipitated at –20 °C for at least 30 min. The pellet, obtained by centrifugation for 5 min at 14000 r.p.m., was washed twice with 500 µL ethanol 70 % at –20 °C. DNA was dried overnight at room temperature and suspended in 97.5 µL TE-buffer (10 mM Tris, 10 mM Na-EDTA, pH 8.0) with 2.5 µL RNAse-solution (10 mg pancreatic RNAse 20 U/mg was added to 1 mL 0.01 M Na-acetate, heated at 100 °C during 15 min and cooled slowly to room temperature; the pH was adjusted to 7.4 by adding 100 µL Tris·HCl). The sample was incubated for 5–30 min at 37 °C and then stored in the refrigerator.

Sequencing

Amplicons V9G and LS 266 (De Hoog & Gerrits van den Ende 1998) were generated as above and purified using the Gel Band Purification Kit (Amersham Pharmacia, Roosendaal, The Netherlands). DNA was bound to GFX-columns, eluted according to protocols given by the supplier, and collected with TE-buffer. Concentrations of amplicons were estimated by comparison with SmartLadder markers (Eurogentec, Seraing, Belgium) on 1 % agarose gels. Sequencing primers were ITS1, ITS 4 and ITS 5; reactions (96 °C, 10 min; 50 °C, 5 min; 60 °C, 4 s; 25 cycles) were carried out with 15–50 ng of DNA for a 10 µL reaction mixture including 4 pmol primer and 4 µL BigDye RR Mix (Applied Biosystems, Nieuwerkerk a/d IJssel, The Netherlands). Subsequently DNA was precipitated with ethanol and sequenced using an ABI Prism™ 310 Genetic Analyzer (Applied Biosystems).

Alignment and phylogenetic analysis

Sequences were adjusted using SeqMan of Lasergene software (DNASTAR, Madison, Wisconsin). Alignment of the Internal Transcribed Spacer (ITS) region was done using BioNumerics (Applied Maths, Kortrijk, Belgium) guided by iterative production of trees based on Ward's averaging algorithm. Distance trees were constructed with neighbour-joining with Kimura-2 correction using the TREECON v. 1.3b software package (Van de Peer & De Wachter 1993), and phylogenetic trees using PAUP v. 4.0b8 with heuristic search option (data not shown). Bootstrap values were calculated from 100 resampled datasets. The 18S sequences were aligned with the ARB package developed by W. Ludwig (www.mikro.biologie.tu-muenchen.de/pub/ARB). Small Subunit (SSU) trees were reconstructed using positional variability with the neighbour-joining (not shown) and parsimony options in ARB. Alignment was optimized in a subfile of a database containing about 2500 near-complete fungal SSU sequences available at CBS.

Table 1. List of strains with their origin, arranged according ribosomal Groups 1 and 2 of Ueda-Nishimura & Mikata (2000).

Species	Strain	GenBank	Origin
Ribosomal Group 1			
1. *Gal. geotrichum*	CBS 772.71[T,a]		T of *Gal. geotrichum,* Homothallic, ex soil, Puerto Rico
	CBS 773.71[a]	AY788343	Ex soil, Puerto Rico
	CBS 774.71[a]	AY788344	Ex 773.71, MT of 775.71
	CBS 775.71[a]	AY788345	Ex 773.71, MT of 774.71
2. *Gal. reessii*	CBS 179.60[T,a]	AY788299	T of *Endomyces reessii*, ex cold-water retting of *Hibiscus cannabinus*
	CBS 295.84[a]		From E. Guého
	CBS 296.84[a]		Dominican Republic; from Inst. Pasteur
	CBS 564.97[a]		Ex soil, Costa Rica; from E.E. Butler
	CBS 565.87		Ex soil, Costa Rica; from E.E. Butler
3. *Gal. citri-aurantii*	CBS 228.38[a]		Ex *Citrus limonium*, Argentina; MT A1
	CBS 175.89[T]	AY788295	T of *Gal. citri-aurantii*, ex soil of orange orchard, Zimbabwe; MT A1
	CBS 176.89[T,a]	AY788296	T of *Gal. citri-aurantii*, ex soil of orange orchard, California, U.S.A.; MT A2
	CBS 604.85		Ex *Citrus paradisi*; MT A1
	CBS 605.85[a]	AY788331	Ex *Citrus paradisi*; MT A2
	CBS 246.96		Ex soil, Israel; from E.E. Butler; MT A1
	CBS 247.96		Ex soil, Israel; from E.E. Butler; MT A2
	CBS 248.96		Ex soil, Florida, U.S.A.; from E.E. Butler; MT A1
	CBS 249.96		Ex soil, Florida, U.S.A.; from E.E. Butler; MT A2
4. *Geo. europaeum*	CBS 866.68[T]	AY788351	T of *Geo. europaeum*, ex wheat-field soil, Germany
5. *Gal. pseudocandidus*	CBS 820.71[a]		Ex paper pulp, France; homothallic
	CBS 267.79[a]		Ex *Beta vulgaris*, The Netherlands
	CBS 626.83[T,a]	AY788334	T of *Geo. pseudocandidus*, ex stomach of elk, France
	CBS 100812	AY788288	Ex bark of beech log (*Fagus sylvatica*), Switzerland
	CBS 101161		UAMH 402, ex sewage treatment filter, England
	CBS 101162		Ex soil, near Lydenburg, South Africa
	CBS 101163		Ex soil, South Africa
6. *Gal. candidus*	CBS 109.12[a]		Ex milk, U.S.A.
	CBS 110.12[a]		Probably ex-type strain of *Oidium humi*, Inst. Pasteur
	CBS 121.22[a]		Ex bulb of *Hyacinthus orientalis*, The Netherlands
	CBS 122.22[a]		Authentic for *Oospora fragrans* var. *minuta*, ex *Musa* sp.
	CBS 114.23		Possibly ex-type strain of *Oidium nubilum*
	CBS 115.23		Ex decaying fruit of *Lycopersicon esculentum*
	CBS 116.23[a]		Authentic for *Oospora lactis* var. *parasitica*, ex fruit *Lycopersicon esculentum*, U.S.A.
	CBS 149.26[a]		Authentic for *Oidium asteroides*
	CBS 176.28[a]		Ex fruit *Durio zibethinus*
	CBS 178.30[T,a]		T of *Oospora lactis* var. *exuberans*, ex white slime flux in *Populus alba*, Germany
	CBS 180.33[a]		Authentic for *Geo. matelense* var. *chapmanii*; MT alpha
	CBS 181.33		Ex human nail, The Netherlands
	CBS 182.33[T,a]	AY788300	T of *Geo. javanense*, ex yoghurt, Italy
	CBS 193.34[T,a]		T of *Geo. versiforme*
	CBS 194.35	AY788304	Possibly a subculture of CBS 149.26
	CBS 195.35		Authentic for *Geotrichum matelense* var. *matelense*
	CBS 224.48		Ex fly in petroleum
	CBS 267.51		Sent by R. Ciferri as *Geotrichum pulmoneum*
	CBS 178.53[T,a]		T of *Endomyces lactis*, Germany
	CBS 184.56[a]		Ex human tongue, Germany
	CBS 185.56		Ex human sputum, The Netherlands
	CBS 240.62		Ex germinating grain of *Hordeum vulgare*, The Netherlands
	CBS 187.67		Ex Camembert cheese
	CBS 178.71[a]	AY788297	Ex soil polluted with oil, Germany; homothallic
	CBS 476.83[a]		Ex soil, Senegal; MT alpha
	CBS 557.83[T,a]	AY788327	T of *Geo. novakii*, ex fruit of *Prunus persica,* Egypt; MT a
	CBS 279.84[T,a]		T of *Geo. redaellii*
	CBS 299.84		Ex fruit of *Lycopersicon esculentum*, France
	CBS 607.84[a]		Ex industrial contaminant, Netherlands
	CBS 615.84[NT,a]		NT of *Geo. candidum*, ex Brie cheese, France, homothallic
	CBS 606.85		Ex *Drosophila* sp., Cameroun
	CBS 607.85		Ex *Drosophila* sp., Cameroun
	CBS 624.85[T,a]		T of *Trichosporon inulinum*
	CBS 357.86[a]		Unknown; MT a
	CBS 144.88		Ex fruit, The Netherlands
1. *D. albidus*	CBS 766.85	AY788342	Ex exudate of angiosperm tree, Japan

2. *D. australiensis*	CBS 625.74[T,b]		T of *D. australiensis*, Decaying cladode of *Opuntia inermis*, Australia
	CBS 666.79[b]		Ex necrosis in *Opuntia* sp., South Africa
	CBS 667.79		Ex necrosis in *Opuntia* sp., South Africa
	CBS 372.83[b]	AY788314	Ex *Euphorbia ingens*, South Africa
	UOFSY 0065		Unknown
3. *D. aggregatus*	CBS 175.53[T,b]	AY788294	T of *D. aggregatus*, ex pupal galleries of *Ips acuminatus* in *Pinus sylvestris*, Germany
	CBS 152.57[b]	AY788292	Ex *Ips pini* frass in root *of Pinus resinosa*, U.S.A.
	CBS 764.85[b]		Ex slime flux of *Pinus ponderosa*, U.S.A.
	CBS 284.86[b]		Unknown
	CBS 285.86		Unknown
4. *D. geniculatus*	CBS 184.80[T]	AY788301	T of *D. geniculatus*, ex pulp *Psidium guajava*, Maharastra, India
5. *Geo. fermentans*	CBS 409.34[c]	AY788315	Ex woodpulp mill
	CBS 439.83[T,c]	AY788318	T of *Geo. fermentans*, ex woodpulp, Sweden
	CBS 451.83[c]	AY788319	Ex tanned sheep skin, France
	CBS 452.83		Ex tanned sheep skin, France
	IGC 3556		Ex *Quercus kelloggii*
6. *D. armillariae*	CBS 165.29		Unknown
	CBS 817.71[T,b]	AY788348	T of *Geo. armillariae*, ex *Armillaria mellea*, Netherlands
	CBS 818.71[b]		ex *Armillaria mellea*, Netherlands
	CBS 834.71	AY788350	Ex gills of *Armillaria mellea*, Netherlands
	CBS 540.76		Ex gills of *Armillaria mellea*, Netherlands
	CBS 623.82		Ex *Armillaria* sp., Belgium
	CBS 624.82[b]		Ex *Armillaria mellea*, Belgium
	CBS 600.83		Ex *Armillaria* sp., Netherlands
7. *Geo. restrictum*	CBS 111234[T]		T of *Geo. restrictum*, ex *Picea abies* (endophytic), Sweden
8. *Geo. klebahnii*	CBS 179.30[T,c]	AY788298	T of *Geo. klebahnii*, ex slime flux *Taxus baccata*
	CBS 196.35[c]		Ex woodpulp
	CBS 627.74[T,c]	AY788335	T of *Trichosporon penicillatum*, ex flux of elm
	CBS 511.83[c]	AY788320	Ex sewage filter
	IGC 3715		Ex *Quercus kellogii*, California, U.S.A.
9. *D. macrosporus*	CBS 259.82[T,b]	AY788310	T of *D. macrosporus*, ex slime trail plasmodium of *Badhamia utricularis*, U.K.
	CBS 260.82[b]	AY788311	Ex slime trail plasmodium of *Badhamia utricularis*, U.K.
Ribosomal Group 2			
1. *M. starmeri*	CBS 780.96[T,b]	AY788346	T of *D. starmeri*, ex rotting saguaro plant, Arizona, U.S.A.
	CBS 781.96[b]	AY788347	Ex rotting cladode of *Opuntia ficus-indica*, Arizona, U.S.A.
2. *M. ovetensis*	CBS 192.55[T,b]	AY788303	T of *Endomyces ovetensis*, ex tannin concentrate, Spain
	CBS 634.85[b]		Ex slime flux in *Quercus* sp., Germany
	CBS 635.85[b]		Unknown, Germany
	CCY 30-2-6		Unknown
	CBS 749.85[b]	AY788337	T of *D. ambrosiae*, ex insect gallery, California, U.S.A.
3. *M. tetrasperma*	CBS 765.70[T]	AY788340	T of *Endomyces tetrasperma,* ex wet conveyer, California, U.S.A.
4. *M. magnusii*	CBS 107.12[b]		Unknown
	CBS 108.12	AY788289	Unknown, possibly ex-type strain of *Endomyces magnusii*.
	CBS 151.30[b]	AY788290	Ex slime flux in *Quercus* sp., Germany
	CBS 234.85[b]	AY788307	Ex slime flux in *Quercus alba*, Pennsylvania, U.S.A.
	CCY 42-1-2		Unknown
	CCY 42-1-3		Unknown
	CCY 42-1-4		Unknown
	CCY 42-1-5		Unknown
5. *M. spicifer*	CBS 244.85[T]	AY788308	T of *D. spicifer*, ex cactus rot, Arizona, U.S.A.
6. *M. capitatus*	CBS 197.35[d]	AY788305	Ex woodpulp, Sweden; MT a
	CBS 312.76[d]	AY788312	Ex sputum of human, Germany
	CBS 162.80[b,d]	AY788293	Ex bovine mastitis milk, U.K.
	CBS 571.82[LT,b,d]	AY788328	LT of *Trichosporon capitatum*, ex woodpulp, Sweden
	CBS 572.82		Ex woodpulp, Sweden
	CBS 573.82[b,d]		Ex yeastcake
	CBS 574.82		Ex sputum of human, Norway
	CBS 575.82[d]		Ex man, South Africa
	CBS 577.82[d]	AY788330	Ex sputum of human, Germany
	CBS 579.82		AUT of *Geotrichum linkii*
	CBS 580.82[d]		AUT of *Geotrichum linkii*, ex sputum of human; MT alpha
	CBS 207.83[b,d]	AY788307	T of S. *pseudotrichosporon,* ex sputum of human, U.S.A.
	CBS 598.83		Ex oral infection of human patient
	CBS 716.84[d]	AY788336	Ex digestive tube of pig, France

	CBS 327.86		Ex blood culture of human patient, U.S.A.
	VTTD-95458[d]		Unknown
	UAMH 375		Unknown
7. *S. suaveolens*	CBS 152.25[T,c]	AY788291	T of *Oidium suaveolens*, ex water brewery
	CBS 194.34[c]	AY788304	Ex mash of *Zea mays*
	CBS 188.38[c]		Ex effluent milk of factory
	CBS 127.76[T,c]		T of *Geo. fici*, ex fruit of *Ficus* sp., Japan
	CBS 610.85[c]		Ex pulmonary infection of female, India
	CBS 326.86[c]	AY788313	Ex human sputum, India
	CBS 785.86[c]		NRRL Y-699
	CBS 380.94[c]		Ex process-water from wheat-starch production plant, Belgium
	JCM 5223[c]		Ex extract soybean flake
8. *S. gigas*	CBS 140.25[T,a]		T of *Oospora gigas*, ex juice of *Arenga saccharifera*
	CBS 126.76[T,a]	AY838940	T of *Geo. rectangulatum*, ex oily debris, Japan
9. *S. chiloënsis*	CBS 8187[T]	AY788347	T of *Schizoblastosporion chiloënse*, ex rotten trunk of *Eucryphia cordifolia*, Chile
10. *S. clavata*	CBS 425.71[T,c]	AY788317	T of *Geotrichum clavatum*, ex human lung tissue, U.S.A.
	CBS 576.82	AY788329	Ex human patient with asthma
	CBS 758.85[c]	AY788339	Ex cactus rot, U.S.A.
	CBS 969.87	AY788352	Ex man, France
	CBS 970.87[c]		Ex condensation droplets brewery, Belgium
	CBS 489.88		Ex man, France
	IP 95662[c]		Ex man, France
11. *S. saccharophila*	CBS 252.91	AY788309	Ex bog pool, Sammlung von Algenkulturen, Göttingen, FRG
	CBS 412.95	AY788316	Ex plants in creek, Netherlands
12. *M. ingens*	CBS 518.90[b]		CBS 4115, ex phenolic waste
	CBS 519.90[b]	AY788322	CBS 4825, ex wine cellar, South Africa; MT a
	CBS 520.90		CBS 4826, ex wine cellar, South Africa
	CBS 521.90[T]	AY788323	T of *D. ingens*, CBS 4827, ex wine cellar, South Africa; MT a
	CBS 523.90[T,b]	AY788325	T of *D. ingens*, CBS 7197, unknown; MT alpha
	CBS 522.90[b]	AY788324	CBS 6057, ex asphalt-plant waste lagoon
	CBS 101346		CBS 1971, ex industrial sulphite waste
13. *S. quercus*	CBS 750.85[b]	AY788338	Ex slime flux *Quercus rubra*, Ontario, Canada
	CBS 751.85[b]		Ex slime flux *Quercus rubra*, Ontario, Canada
	CBS 752.85[T,b]		T of *S. quercus*, ex slime flux *Quercus rubra*, Ontario, Canada
14. *S. japonica*	CBS 100158	AY788287	JCM 2451, ex exudate of tree
15. *S. fungicola*	CBS 625.85[T]	AY788333	T of *S. fungicola*, ex *Nectria cinnabarina*, Russia
16. *S. psychrophila*	CBS 765.85[T]	AY788341	T of *S. psychrophila*, ex slime flux in *Pinus ponderosa*, U.S.A.
17. *S. ingens*	CBS 517.90[T,b]	AY788321	T of *Candida ingens*, CBS 4603, ex wine cellar, South Africa
	CBS 524.90[b]	AY788326	CBS 6787, mutant of 517.90, T of *Pichia humboldtii*

[T] = ex-Type strain; [LT] = ex-Lectotype strain; [NT] = ex-Neotype strain; MT = Mating type; [a] Strains used in intra-specific reassociations (Smith *et al*. 1995); [b] Strains used in intra-specific reassociations (Smith & Poot 2003); [c] Strains used in intra-specific reassociations (Smith *et al*. 2000); [d] Strains used in intraspecific reassociations (Smith & Poot, 1998); CBS = Centraalbureau voor Schimmelcultures, Utrecht, the Netherlands; CCY = Czechoslovak Collection of Yeasts, Institute of Chemistry, Slovakian Academy of Science, Bratislava, Slowakia; IGC = Centro de Biologia, Instituto Gulbenkian de Ciencia, Oeiras, Portugal; IP = Institut Pasteur, France; UAMH = University of Alberta, Microfungus Herbarium and Collection, Edmonton, Canada; UOFS = Department of Microbiology and Biochemistry, University of Orange Free State, Bloemfontein, South Africa; VTT = VTT, Biotechnology and Food Research, Espoo, Finland.

A total of 960 valid columns were used of a total of 2630 positions necessary for alignment, which corresponds to 1630 bases with reference to *Saccharomyces cerevisiae*. A total of 659 nucleotides were invariant. The Trichomycete genus *Smittium* (*Harpellales*) was chosen as outgroup. Bootstrap values were calculated from 100 resampled datasets.

RESULTS AND DISCUSSION

The phylogenetic position of *Geotrichum* and teleomorphs with respect to the *Hemiascomycetes* is elaborated in Fig. 1. Due to the large deviations between groups leading to an overabundance of non-comparable sites, a remarkable difference was found with neighbour-joining and parsimony algorithms. With both, species with *Geotrichum* anamorphs were found in two groups widely remote from each other. With NJ (data not shown) the group with an entire SSU gene (Group 1 of Ueda-Nishimura & Mikata 2000) was located within the *Hemiascomycetes*, though in a distant position close to *Schizosaccharomyces* and *Pneumocystis*, which have been united in the class *Archiascomycetes* by Nishida & Sugiyama (1994). Ribosomal Group 2 (Ueda-Nishimura & Mikata 2000) is remarkable in lacking several loops within the V2, V3 and V8 variable domains in the ribosomal operon. Additional deviations were large, such that SSU sequences partially are aligned with difficulty. With NJ Group 2 was found far outside the *Hemiascomycetes*. For this

reason the Trichomycete genus *Smittium* (*Harpellales*) was chosen as outgroup, where large thalli remarkably similar to those of *Geotrichum fragrans* and related species are known. The distance of ribosomal Group 2 to both *Ascomycota* and *Basidiomycota* was very large; it could therefore not be clearly attributed to any fungal higher category. Also alignment with members of *Zygomycota* was highly ambiguous. Recently another group more or less intermediate between the main categories *Ascomycota* and *Basidiomycota* was found in *Wallemia*, which was therefore raised to the class-level (P. Zalar *et al*., pers. comm.). Ribosomal Group 2 thus would deserve a similar status. However, this would be in conflict with the large similarities in morphology and ecology between members of the Groups 1 and 2 (see below). This resembles the situation in *Entomophthorales*, where extreme phylogenetic distances are found between fungi that are even difficult to distinguish at the generic level. It is obvious that with large-scale sequencing of fungal mavericks in yeasts and lower fungi where phylogenetic branches are much longer than in the *Ascomycota*, many more conflicts between cladograms and phenograms will emerge. We therefore refrain from formal introduction of such a category for *Geotrichum* ribosomal Group 2.

Deviations from existing classifications were considerably reduced when hypervariable regions were disregarded by the use of a parsimony algorithm (Fig. 1). Again the *Geotrichum* ribosomal Groups 1 and 2 were distinctly apart, but they were both located within a well-supported clade that could be referred to as the *Hemiascomycetes*. The *Archiascomycetes* were basal to this clade. *Basidiomycota* (including *Wallemia*) and *Zygomycota* were separated from both groups of *Ascomycota* at 99 % bootstrap support.

Six SSU sequences of *Galactomyces* species are available in GenBank and have been included in the tree (Fig. 1). Among these are two sequences labeled as *Gal. geotrichum* AB000647, based on strain IFO 9541 (Ueda-Nishimura & Mikata 2000) and X69842 based on strain MUCL 28959 (Wilmotte *et al*. 1993), and one labeled as *Endomyces geotrichum* U00947, based on a strain from the University of California (Berbee & Taylor 1993). There is some diversity within this group (Fig. 1). With the subdivision of the *Geotrichum candidum* complex (see below) we are unsure to which species these sequences are corresponding; for that reason we listed them as *Galactomyces* sp.

Specific borderlines between species have previously been determined by M.Th. Smith and coworkers (Smith *et al*. 1995, 2000, Smith & Poot 1998, 2003) using nDNA reassociation techniques and determination of mol % G+C of DNA. When G+C values were calculated from the derivative of the melting curve, patterns with one or two peaks were revealed that were highly characteristic of the species. Thus the species structure of the genus *Geotrichum* and its teleomorphs had already been determined prior to establishment of phylogenetic relationships on the basis of ITS sequence data, although the authors had refrained from formal introduction of taxa. A number of species showed marked phenetic deviations in nutritional physiology as well as derivative graph of the nDNA melting curve and were therefore not analyzed with nDNA reassociation. The present ITS study aimed to verify the taxa thus delimited on the basis of an independent data set.

Sequencing of the rDNA ITS region proved to be difficult. In some cases there were short stretches that repeatedly gave low peaks that were difficult to read. Because of such recalcitrant areas the sequences of particular sites remained unclear despite multiple sequencing efforts. In addition, occasionally unexpected degrees of variability within the same species (judging from nDNA reassociation data) were revealed, which shed doubt on the reliability of the sequencing result. Despite the very short length of the spacer domains, many strains were therefore sequenced with 4 to 6 runs.

Alignment was hampered by the occurrence of small to considerable mutations in the recognition sites of the otherwise highly conserved 5.8S and LSU rDNA domains. The two ribosomal groups recognized in LSU as well as in SSU data by Kurtzman & Robnett (1995) and Ueda-Nishimura & Mikata (2000) were also reflected in the ITS regions. ITS sequences of Groups 1 and 2 could not be evenly aligned, and are therefore presented in two separate trees based nearly exclusively on the 5.8S rDNA (Fig. 2). Even these 5.8S rDNA genes of the two groups showed considerable deviations.

As shown by Smith *et al*. (1995, 2000) the borderline between individual species is at > 80 % nDNA reassociation values (Figs 3, 4). Low values (< 20 %) had always been acknowledged to indicate separate species, but now also intermediate values (40–60 %) were taken as proof of separation. Consequently it was concluded that the *Galactomyces geotrichum / Geotrichum candidum* complex contained four separate species: *Gal. geotrichum sensu stricto*, and A, B and C, which are now classified as *Gal. candidum, Gal. pseudocandidus* and *Geo. europaeum,* respectively. These entities are separated from each other by 4.0–4.4 % ITS diversity (Fig. 3). Infra-specific ITS variability mostly is less than 1 %.

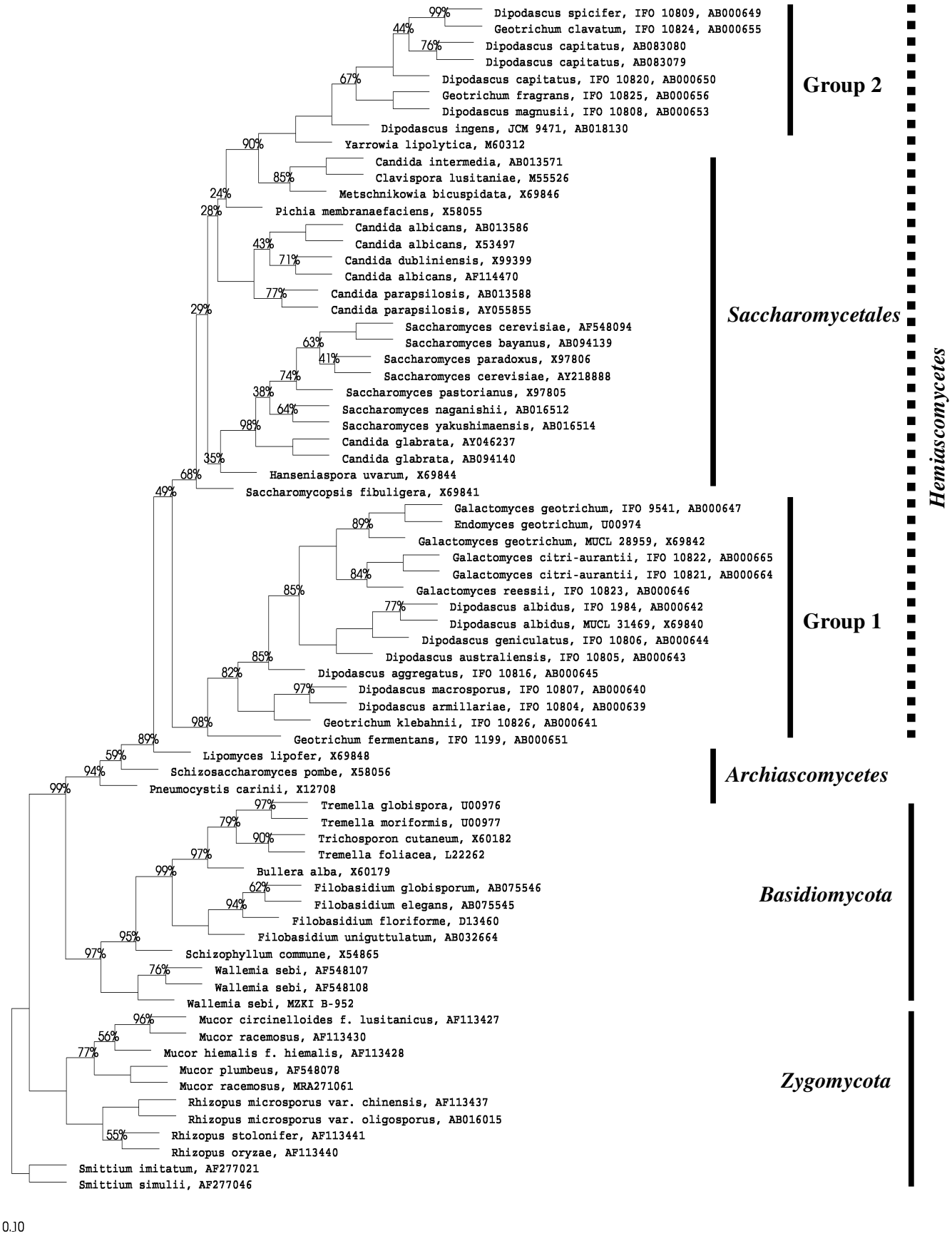

Fig. 1. SSU rDNA tree based on 72 near-complete sequences available from GenBank. The tree was constructed with the parsimony option in the ARB package; 2630 positions were taken into account, of which 960 were phylogenetically informative. Bootstrap values of 100 resampled data sets are shown with the branches. *Smittium imitatum*, AF 277021 was used as outgroup. The dotted line indicates the *Hemiascomycetes*.

Table 2. G+C percentages of strains belonging to Ribosomal Group 1: comparison of genomic values with those found in rDNA ITS.

		Mol % G+C of nDNA[1]		ITS1		5.8S		ITS2	
Strain	**Name**	**Low peak**	**High peak**	**Length**	**G+C %**	**Length**	**G+C %**	**Length**	**G+C %**
CBS 152.57	*Dipodascus aggregatus*	22.1	46.0	96	25.0	108	38.0	117	39.3
CBS 175.53^{T}		22.1	44.7	91	25.3	108	38.0	121	35.5
dH 12682		20.0	43.2	95	24.2	108	38.0	116	36.2
CBS 111234	*Geotrichum restrictum*	17.0	42.7	89	24.7	108	38.0	112	43.8
CBS 179.30^{T}	*Geotrichum klebahnii*	-	42.1	84	29.8	108	37.0	110	32.7
CBS 627.74		-	41.5	85	29.4	108	37.0	110	32.7
CBS 511.83		-	41.7	83	26.5	108	37.0	111	34.2
CBS 817.71^{T}	*Dipodascus armillariae*	13.5	41.7	84	28.6	108	37.0	114	32.5
CBS 834.71		14.3	41.6	84	28.6	108	37.0	114	32.5
CBS 624.82		15.4	43.6	84	28.6	108	37.0	114	32.5
CBS 260.82	*Dipodascus macrosporus*	21.4	42.7	97	21.7	108	37.0	133	27.9
CBS 259.82^{T}		20.5	42.8	98	21.4	108	37.0	134	28.4
CBS 409.34	*Geotrichum fermentans*	-	46.7	81	25.9	108	38.0	107	42.1
CBS 439.83^{T}		-	45.6	81	27.2	108	38.0	107	42.1
CBS 451.83		-	44.9	81	25.9	108	38.0	107	42.1
CBS 175.89^{T}	*Galactomyces citrii-aurantii*	34.3	42.0	73	15.1	108	38.0	110	33.6
CBS 176.89^{T}		32.4	40.0	72	15.3	108	38.0	110	33.6
CBS 605.85		34.5	40.6	73	15.1	108	38.0	109	33.9
CBS 179.60^{T}	*Galactomyces reessii*	29.9	41.9	78	20.5	108	38.0	111	36.0
CBS 557.83	*Galactomyces candidus*	39.7	41.7	74	21.6	108	38.0	111	36.0
CBS 182.33		-	43.5	78	18.0	108	38.0	111	36.9
CBS 178.71		37.2	41.1	77	19.5	108	38.0	114	35.1
CBS 775.71	*Galactomyces geotrichum*	35.3	42.9	76	18.4	108	38.0	112	35.7
CBS 773.71		34.1	41.9	76	18.4	108	38.0	112	35.7
CBS 774.71		34.9	42.5	76	18.4	108	38.0	112	35.7
CBS 866.68^{T}	*Geotrichum europaeum*	36.6	43.0	75	16.0	108	38.0	110	37.3
CBS 626.83^{T}	*Galactomyces pseudocandidus*	36.7	43.4	78	14.1	108	38.0	110	36.4
CBS 100812		nd	nd	79	13.9	108	38.0	110	37.3
CBS 184.80^{T}	*Dipodascus geniculatus*	25.2	42.2	107	18.7	108	35.2	125	26.4
CBS 372.83	*Dipodascus australiensis*	-	36.8	120	20.0	108	35.2	126	30.2
CBS 766.85	*Dipodascus albidus*	20.4	37.4	161	19.9	108	36.1	420	19.1

LT = ex-lectotype strain; T = ex-type strain.

[1]Values from derivatives of melting curves. Data taken from Smith *et al.* (1995, 2000) and Smith & Poot (1998, 2003).

Table 2. (continued). G+C percentages of strains belonging to Ribosomal Group 2: comparison of genomic values with those found in rDNA ITS domains.

		Mol % G+C of DNA[1]		ITS1		5.8S		ITS2	
Strain	**Name**	**Low peak**	**High peak**	**Length**	**G+C %**	**Length**	**G+C %**	**Length**	**G+C %**
CBS 151.30[T]	*Magnusiomyces magnusii*	33.8	41.6	97	17.5	104	44.2	109	33.9
CBS 108.12		nd	nd	97	17.5	104	44.2	109	33.9
CBS 234.85		31.4	38.8	97	17.5	104	44.2	109	35.8
CBS 194.34	*Saprochaete suaveolens*	-	39.6	81	17.3	104	44.2	109	33.9
CBS 326.86		-	40.3	97	17.5	104	44.2	110	33.6
CBS 188.38		-	41.7	81	17.3	104	44.2	110	33.6
JCM 2450		-	nd	97	17.5	104	44.2	110	33.7
CBS 152.25[T]		-	39.4	98	16.3	104	44.2	110	32.7
CBS 100158[T]	*Saprochaete japonica*	-	42.2	95	17.9	104	44.2	106	33.0
CBS 765.70[T]	*Magnusiomyces tetrasperma*	35.4	43.4	96	17.7	104	44.2	106	34.0
CBS 625.85	*Saprochaete fungicola*	-	41.3	99	18.2	104	44.2	110	31.8
CBS 252.91[T]	*Saprochaete saccharophila*	-	nd	94	20.2	104	44.2	107	34.6
CBS 412.95		-	39.7	94	20.2	104	44.2	107	34.6
CBS 523.90[T]	*Magnusiomyces ingens*	-	44.0	97	15.5	104	44.2	107	35.5
CBS 522.90		-	43.7	97	15.5	104	44.2	108	35.2
CBS 521.90[T]		-	44.0	97	15.5	104	44.2	107	35.5
CBS 519.90		-	43.5	98	16.3	104	43.3	107	35.5
CBS 524.90	*Saprochaete ingens*	25.9	37.4	95	15.8	104	44.2	106	34.0
CBS 517.90[T]		25.9	39.7	98	16.3	104	44.2	106	34.0
CBS 126.76	*Saprochaete gigas*	-	41.3	91	16.5	104	44.2	113	34.5
CBS 8187[T]	*Saprochaete chiloënsis*	34.6	40.6	97	16.5	104	44.2	114	36.0
CBS 781.96	*Magnusiomyces starmeri*	27.6	42.3	97	17.5	104	43.3	123	31.7
CBS 780.96[T]		27.9	42.5	97	17.5	104	43.3	124	31.5
CBS 192.55[T]	*Magnusiomyces ovetensis*	21.5	47.5	90	18.9	104	43.3	106	34.9
CBS 749.85		19.4	47.5	91	18.7	104	43.3	106	34.9
CBS 750.85	*Saprochaete quercus*	39.9	48.0	93	18.3	104	43.3	109	32.1
CBS 576.82	*Saprochaete clavata*	27.1	39.0	105	14.3	104	45.2	178	28.7
CBS 969.87		26.6	38.5	105	14.3	104	45.2	179	28.5
CBS 758.85		26.7	38.6	105	14.3	104	45.2	177	28.8
CBS 425.71[T]		26.5	38.6	105	14.3	104	45.2	178	28.7
CBS 765.85[T]	*Saprochaete psychrophila*	-	43.4	105	14.3	104	45.2	177	28.8
CBS 244.85[T]	*Magnusiomyces spicifer*	29.2	38.3	107	15.0	104	45.2	180	28.3
CBS 197.35	*Magnusiomyces capitatus*	30.0	39.4	104	15.4	104	45.2	169	30.8
CBS 312.76		27.0	37.0	104	15.4	104	45.2	169	30.8
CBS 577.82		28.2	37.9	104	15.4	104	45.2	169	30.8
CBS 716.84		28.3	38.2	104	15.4	104	45.2	169	30.8
CBS 571.82[LT]		30.0	39.3	104	15.4	104	45.2	169	30.8
CBS 162.80		26.9	38.7	104	15.4	104	45.2	169	30.8
CBS 207.83		30.0	39.4	104	15.4	104	45.2	169	30.8

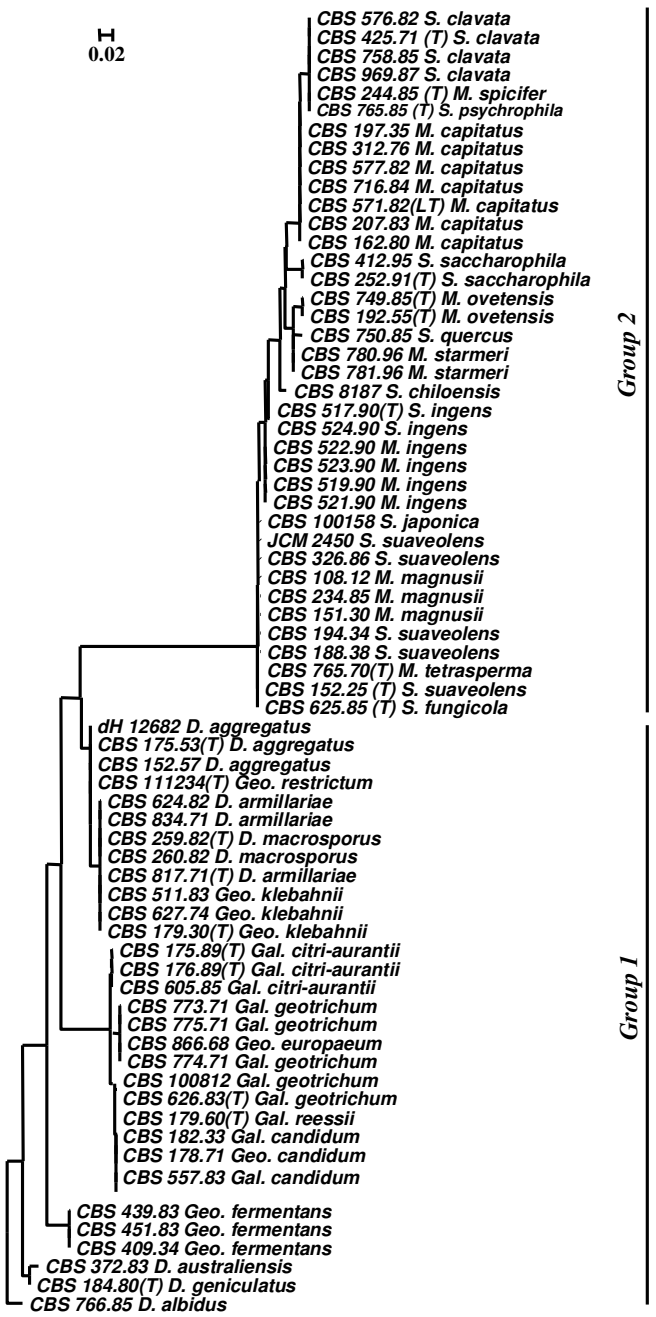

Fig. 2. Distance tree of ribosomal Groups (1) and (2) generated with the neighbour-joining algorithm in the TREECON package mainly based on complete alignment of rDNA 5.8 sequences; 112 positions were taken into account, of which 32 were phylogenetically informative. Kimura 2-parameter correction was used. *Dipodascus albidus*, CBS 766.85 was used as outgroup.

As established in a series of publications by Smith and co-workers (Smith *et al.* 1995, 2000, Smith & Poot 1998, 2003), species are also distinguished by significant differences in genomic G+C percentages (Table 2). The first derivative of the melting curve shows one or two peaks. In ribosomal Group 1, the first peak varies between 13.5 and 37.2 %, and in ribosomal Group 2 between 19.4 and 39.9 %. Similarly, the second peak, which is invariably present, varies between 36.8 and 46.7 % in Group 1, and between 37 and 48 % in Group 2.

The ITS spacer domains in all species analyzed are remarkably short, and have a striking AT bias. The 5.8S rDNA gene is 104–108 bp. The ITS1 region varies between 68 and 161 bp, ITS2 is 107–220 bp long (Table 2). Ribosomal G+C percentages of ITS1 are very low: in Group 1 between 13.9 and 29.8 % (av. 21.2), and in Group 2 between 14.3 and 20.2 % (av. 16.6; Table 2). The ITS2 in Group 1 has G+C % between 19.1 and 42.1 % (av. 34.7), and in Group 2 it is between 28.3 and 36.0 % (av. 32.6). All values of G+C % in ITS in Group 1 are consistently higher, though very large variations are noted between individual species. Den Bakker *et al.* (2004) found similar AT bias in either ITS1 or ITS2 of closely related members of the mushroom genus *Leccinium*. The AT-rich stretches in that genus composed minisatellites of repetititive elements, which were thought to come about by gene conversion and unequal crossing over. Due to this process, different members of the same species might differ considerably in base composition. In our data set, large differences within species such as *G. klebahnii* were not strikingly based on an overabundance of AT. This was, however, the case in *Dipodascus albidus*, where both ITS1 and ITS2 were significantly elongated compared to remaining members of ribosomal Group 1, due to insertion of AT-rich stretches.

The ecology of species of the genus *Geotrichum* show a rather unexpected degree of consistency given the large phylogenetic distances between species. Over larger species complexes, ecological similarities can be observed: association with slime flux of trees is a recurrent phenomenon in the genus. Species may also be found in pupal galleries of bark beetles. The shared ecology is found in ribosomal Groups 1 and 2 of Ueda-Nishimura & Mikata (2000): *Dipodascus aggregatus* (1), *D. albidus* (1), *D. magnusii* (2), and *D. ovetensis* (2). Note that *D. magnusii* and *D. ovetensis,* although these species are widely apart in all taxonomic respects, are both consistently found in *Quercus* slime flux on two different continents. A somewhat similar substratum is juicy plant tissue rich in carbohydrates, such as rotting cactus cladodes, or various types of pulp. The following species belong to this category: *D. australiensis* (1), *D. geniculatus* (1), *D. capitatus* (2), *D. spicifer* (2), and *D. starmeri* (2). In some cases material such as wood pulp is self-heated; the species found in this environment, *D. capitatus*, is also encountered as an etiologic agent of opportunistic infections in humans. A third category is occurrence as a hyperparasite on other fungi, which is the case for *D. armillariae* (1) and *D. macrospora* (1), while in the present paper also *Saprochaete saccharophila* (2) seems to belong in this category. In our new taxonomy presented in this paper particularly the species of *Dipodascus* with a known teleomorph show a strong predilection for a specific substrate.

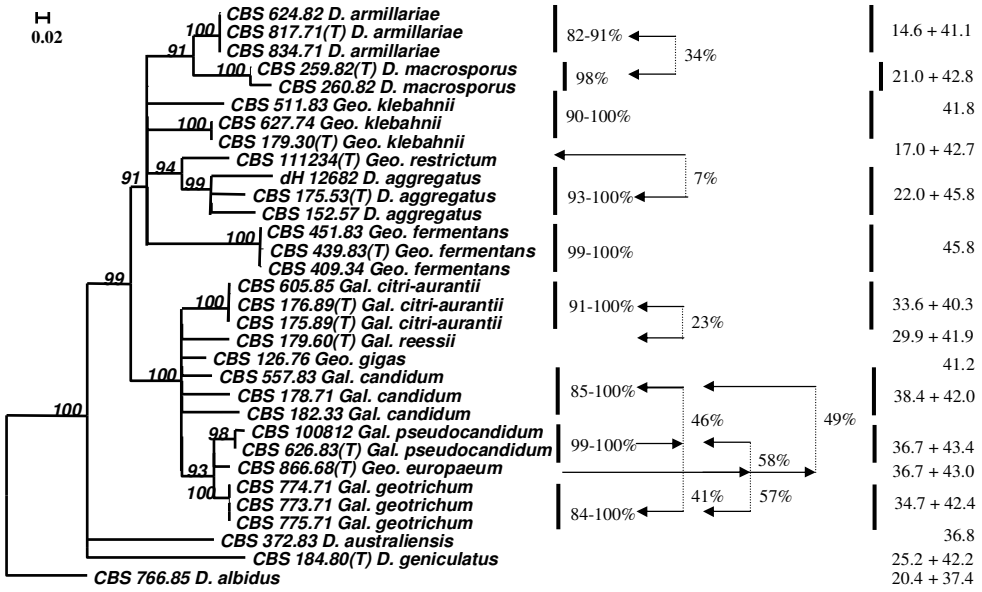

Fig. 3. ITS rDNA phylogeny of ribosomal Group (1) (*Galactomyces-Dipodascus-Geotrichum* clade) generated with the neighbour-joining algorithm in the TREECON package based on complete alignment of ITS sequences including the 5.8S rDNA region. Kimura 2-parameter correction was used. Bootstrap values > 80 of 100 replications are shown. *Dipodascus albidus*, CBS 766.85 was used as outgroup. Intra-species (solid bars only) and inter-species (dotted bars with arrows) genomic DNA/DNA reassociation values are given. Average nDNA mol % G+C (low and high peaks based on derivatives of melting curves) are provided for each species.

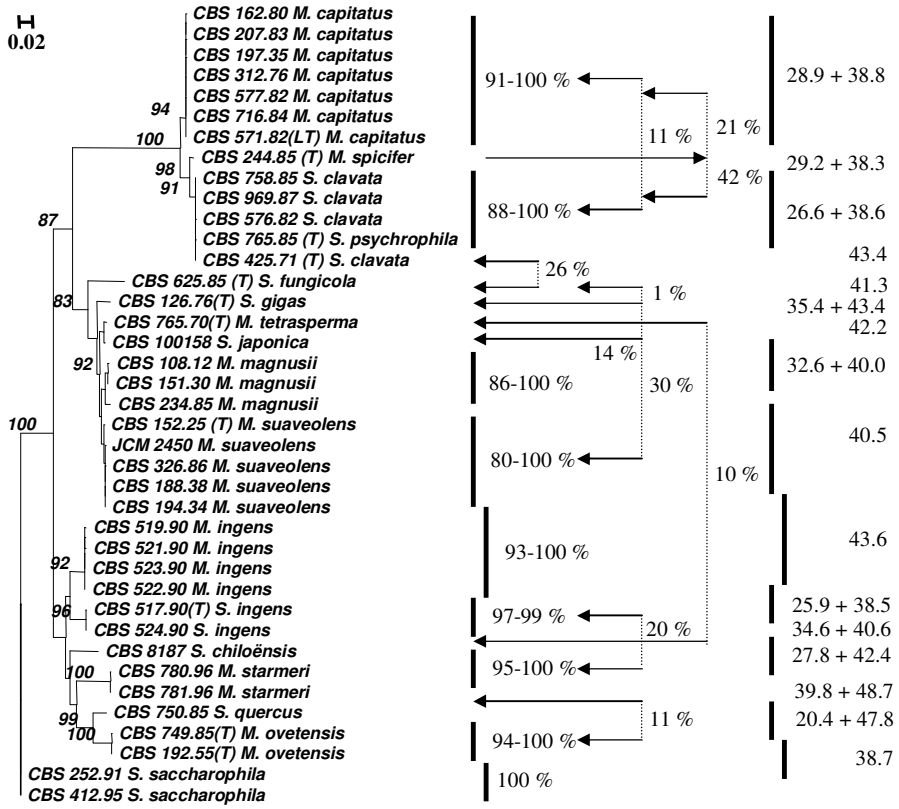

Fig. 4. ITS rDNA phylogeny of ribosomal Group (2) (*Magnusiomyces-Saprochaete* clade) generated with the neighbour-joining algorithm in the TREECON package based on complete alignment of ITS sequences including the 5.8S rDNA region. Kimura 2-parameter correction was used. Bootstrap values > 80 of 100 replications are shown. *Saprochaete saccharophila*, CBS 252.91 was used as outgroup. Intra-species (solid bars only) and inter-species (dotted bars with arrows) genomic DNA/DNA reassociation values are indicated. Average nDNA mol % G+C (low and high peaks based on derivatives of melting curves) are provided for each species.

The genus *Galactomyces* (1) seems less pronounced in its choice of habitat, as strains are found on all kinds of soft plant tissues, in soil, and occasionally in the digestive tract of humans.

The rDNA ITS region was very difficult to align; remarkably different signatures are even observed in the normally highly conserved 5.8S gene. For this reason, the genus *Magnusiomyces* (with anamorph *Saprochaete*) is introduced herewith for species having a Group 2 ribosomal structure, while *Dipodascus* (with *Geotrichum* anamorphs) is retained for species of Group 1 (Figs 3, 4). Species of ribosomal Groups 1 and 2 share some special and basic characteristics despite their phylogenetic distance. This would have been viewed as coincidental homoplasy if the morphology of ana- and teleomorphs of *Dipodascus* and *Magnusiomyces* were not so strikingly similar and at the same time so outstanding in the fungal Kingdom.

The preponderantly filamentous, arthroconidial anamorphs, with pronounced hyphae and septa perforated by numerous micropores (Kreger-van Rij & Veenhuis 1972, 1973, Van der Walt & von Arx 1985, Moore 1987), and the large bipodal asci with ascospores covered by an even, thick gelatinous sheath (de Hoog *et al*. 1986), are quite unique. The morphology of anamorphs is sometimes very striking, arthroconidia being found in both groups. Group 1 is consistent in assimilating D-xylose, but this property is also found in some members of Group 2 (Table 3). The recurrent similarities between such remote organisms strongly suggest that the two main Groups 1 and 2 have not evolved recently, but rather are the remains of a very large, widely radiated group of fungi of which the great majority of species has become extinct. If this hypothesis is correct, particularly the genera *Dipodascus* and *Galactomyces* (ribosomal Group 1) should be regarded as 'living fossils'. The two genera seem to have gone through an evolution separate from each other. If this is correct, the SSU signature of Group 1, i.e., without deletions, must be regarded as being ancestral. This group has also the highly conserved beginning of the 5.8S gene, AACTTTTAAC, widely distributed in the fungal Kingdom, while Group 2 has AACCTCCAAC, which is otherwise, as far as we are aware, unknown in the fungal Kingdom. Group 1 lacks some bases at the otherwise highly conserved beginning of the LSU rDNA domain, which starts with AACCTC rather than GGTTGACCTC in most filamentous fungi. Group 2 is even extremely different, with ACTCC followed by 14 mutations at the beginning of LSU. We conclude that the genera *Galactomyces* and *Dipodascus* are among the very ancient fungi, and that *Magnusiomyces* has branched off from *Dipodascus* at an early state and has shown little evolutionary development other than molecular clock mutations in regions without evolutionary constraints. Independent genes should be sequenced in order to verify this hypothesis.

If we compare interspecific ITS distances between species of *Dipodascus, Magnusiomyces* and *Galactomyces* within ribosomal Groups 1 and 2 with those seen in most genera of filamentous Euascomycetes, remarkable differences are observed. For example, unambiguously separate species of hypocrealean genera such as *Fusarium* and *Trichoderma* may have identical ITS sequences (Lieckfeldt & Seifert 2000). In black yeasts the specific borderline lies around 1 % deviation (de Hoog *et al.* 2003). Within ribosomal Group 1 the maximum ITS span is 30.8 % and within Group 2 the maximum is 23.0 %; distances between nearest species are 1.2–19.9 % within *Dipodascus* (Group 1), 0.7–8.0 % within *Galactomyces* (Group 1) and 2.1–4.8 % within *Magnusiomyces* (Group 2). Alignments partially are highly ambiguous, even within a single genus. Lengths of the ITS1 and ITS2 regions are very variable (Table 2) and very short when compared with filamentous fungi, where the ribosomal spacer region is mostly between 450 and 800 bp (Platas *et al.* 2004). Nevertheless considerable length variation can be observed among related fungi (de Hoog *et al*. 1999): ITS1 was noted to vary between 134 and 260 bp in the order *Dothideales*. In *Geotrichum* ITS1 is frequently well below 100 bp in length (Table 2). ITS spacers (excluding the 5.8S gene) may have a remarkable AT-bias, with GC % sometimes as low as 13.9 % (Table 2). This feature might also be interpreted to indicate a high phylogenetic age, since similar short, AT-rich ITS domains are known in lower fungi such as *Zygomycetes* and *Chytridiomycetes*. However, other phylogenetic mavericks in the fungal Kingdom, such as *Wallemia* (P. Zalar *et al*., pers. comm.), do not show significant AT-bias. As yet we do not have an explanation for these phenomena.

Clades (1) and (2) being phylogenetically widely apart cannot remain without taxonomic consequences. Kurtzman & Robnett (1998) noted earlier in species currently classified in *Dipodascus* Group 2, that despite their high degree of overall morphological similarity to those of Group 1 they all have smaller, 4-spored asci, without subsequent mitoses. In contrast, members of Group 1 have 8-spored asci, or may go through several additional mitoses leading to asci with a maximum of 128 ascospores. Thus there is one consistent phenetic character by which the two *Dipodascus s. l.* groups can be defined. We therefore propose to acknowledge the large phylogenetic distance between Groups 1 and 2 by erecting a new genus for *Dipodascus* Group 2 with the name *Magnusiomyces.* For similar reasons the anamorph genus *Geotrichum* should be subdivided along the same lines. The generic type species, *Geo. candidum*, is a member of Group 1 and is associated with *Galactomyces* and *Dipodascus*; the anamorph generic name *Saprochaete* (Coker & Shanor 1939) is available for members of Group 2. A number of species has not been studied by Ueda-Nishimura & Mikata (2000); Figs 1 and 2 clearly show that the subdivision is reflected in all ribosomal genes and spacers.

Table 3. Physiological characters and mol % G+C of nDNA of novel species of *Galactomyces, Geotrichum, Dipodascus*, *Magnusiomyces* and *Saprochaete* of ribosomal Group 1 and ribosomal Group 2 (Ueda-Nishimura & Mikata 2000).

	Ribosomal Group 1				**Ribosomal Group 2**					
	Geo. europ.	***Gal. pseudo.***	***Gal. candid.***	***Geo. restrict.***	***M. ingens***	***S. quercus***	***S. japon.***	***S. fungic.***	***S. psychr.***	***S. sacch.***
Numbers in physiology	1	7	36	1	8	3	1	1	1	2
Numbers in reassociations	1	4	22	1	4	3	1	1	1	2
D-Glucose	+	+	+	+	+	+	+	+	+	+
D-Galactose	+	+	+	+	+	+	+	+	+	–
L-Sorbose	+	+	+	v	+	v	+	+	+	+/w
D-Glucosamine	–	–	–	+	–	–	–	–	–	–
D-Ribose	–	–	–	–	–	–	–	v	–	–
D-Xylose	+	+	+	+	–	–	–	+	+	–
L-Arabinose	–	–	–	–	–	–	–	–	–	–
D-Arabinose	–	–	–	–	–	–	–	–	–	–
L-Rhamnose	–	–	–	–	–	–	–	–	–	–
Sucrose	–	–	–	–	–	–	–	–	–	–
Maltose	–	–	–	–	–	–	–	–	–	–
α-Trehalose	–	–	–	–	–	–	–	–	–	–
Me-α-glucoside	–	–	–	–	–	–	–	–	–	–
Cellobiose	–	–	–	–	–	–	–	–	–	–
Salicin	–	–	–	–	–	–	–	–	–	–
Arbutin	–	–	–	–	–	–	–	–	–	–
Melibiose	–	–	–	–	–	–	–	–	–	–
Lactose	–	–	–	–	–	–	–	–	–	–
Raffinose	–	–	–	–	–	–	–	–	–	–
Melezitose	–	–	–	–	–	–	–	–	–	–
Inulin	–	–	–	–	–	–	–	–	–	–
Sol. starch	–	–	–	–	–	–	–	–	–	–
Glycerol	+	+	+	+	+	+	+	+	+	+
Erythritol	–	–	–	–	–	–	–	–	–	–
Ribitol	–	–	v	–	–	–	–	+	+	–
Xylitol	–	–	–	–	–	–	–	+	–	–
L-Arabinitol	–	–	–	–	–	–	–	–	–	–
D-Glucitol	+	+	+	+	–	–	+	+	+	–
D-Mannitol	–	–	+	+	–	–	+	+	+	–
Galactitol	–	–	–	–	–	–	–	–	–	–
myo-Inositol	–	–	–	–	–	–	–	–	–	–
D-Glucono-1,5-lactone	–	–	+	nd	–	–	+	+	v	–
D-Gluconate	–	–	w	–	–	–	–	–	–	–
D-Glucuronate	–	–	–	–	–	–	–	–	–	–
D-Galacturonate	+	+	+	nd	–	–	–	–	–	–
Dl-Lactate	+	+	+	–	+	+	+	+	+/w	–
Succinate	+	+	+	nd	+	+	+	+	+/w	+
Citrate	–	v	v	nd	–	–	+	+	v	–
Methanol	–	–	–	nd	–	–	–	–	–	–
Ethanol	+	+	+	nd	+	+	+	+	+	+
Propane 1,2 diol	+	+	+	nd	+	+	+	+	+	+
Butane 2,3 diol	+	+	+	nd	+	+	+	+	+	+
Nitrate	–	–	–	–	–	–	–	–	–	–
Ethylamine	+	+	+	+	+	+	+	+	+	+
L-Lysine	+	+	+	+	+	+	+	+	+	+
Cadaverine	+	+	+	+	+	+	+	+	+	+
w/o vitamins	+	+	+	–	+	+	–	–	–	–
25 °C	+	+	+	+	+	+	+	+	+	+
30 °C	+	+	+	–	+	+	+	+	–	+
35 °C	–	–	+	–	+	–	–	+	–	–
37 °C	–	–	v	–	+	–	–	w	–	–
40 °C	–	–	–	–	–	–	–	–	–	–
Range expansion growth / 7 days in mm radius	21 –26	20 –32	15 –43	2 –3	3 –7	5 –9	9	7 –8	4	3.5 6.0
Mol% G+C low[a]	36.7	36.5	38.4	42.7	43.6	39.8	42.2	41.3	43.4	38.7
Mol% G+C high[a]	43.0	42.7	41.6	–	–	48.7	–	–	–	–

[a] Calculated from derivative of melting curves; w = weak; v = variable; nd = not determined.

TAXONOMY

Teleomorph and anamorph species belonging to the *Galactomyces* clade

Galactomyces Redhead & Malloch, Canad. J. Bot. 55: 1708. 1977.
Anamorph: *Geotrichum* Link : Fr.

= *Mycoderma* Pers., Mycol. Eur. p. 96. 1822 (lectotype species: *M. malti-juniperini* Desm. = *Geotrichum candidum*).
= *Oosporoidea* Sumst., Mycologia 5: 53, 1913 [type species: *O. lactis* (Fresen.) Sumst. = *Geotrichum candidum*].
= *Geotrichum* Link : Fr. subg. *Eugeotrichum* Verona & Cif., Mycopath. Mycol. Appl. 1 222. 1938/39.
= *Geotrichum* Link : Fr. subg. *Pseudomycoderma* Verona & Cif., Mycopath. Mycol. Appl. 1 222. 1938/39.
For a discussion of the doubtful and invalid synonyms *Glucomyces, Polymorphomyces, Fermentotrichum* and *Ascotrichosporon*, see de Hoog et al. (1986).

Thallus growth mostly initiating from a single, inflated cell. *Hyphae* branched at broad or right angles, with rounded apices, disarticulating into arthroconidia. Some additional sympodial conidiogenesis from undifferentiated cells may be present. *Chlamydospores* may be produced. *Gametangia* formed with 1–2(–4) on opposite sides of hyphal septa in areas of dense septation, spherical to clavate, soon fusing at apex and forming an ascus. *Asci* subhyaline, subspherical to broadly ellipsoidal, containing one, rarely two ascospores. *Ascospores* broadly ellipsoidal, subhyaline, with a rough inner wall and an irregular exosporium wall, often with a hyaline equatorial furrow. *Ascospores* liberated by dissolution of the entire ascus wall. Cell walls 3-layered, without xylose or fucose. Septa perforated by micropores. Xylose mostly assimilated. Nitrate not assimilated. Urease not present. Extracellular starch not produced. Diazonium blue B reaction negative. SSU rDNA without deletions, ribosomal Group 1 (Ueda-Nishimura & Mikata 2000).

Type: *Galactomyces geotrichum* (E.E. Butler & L.J. Petersen) Redhead & Malloch (teleomorph); *Geotrichum candidum* Link : Fr. (anamorph).

1. *Galactomyces geotrichum* (E.E. Butler & L.J. Petersen) Redhead & Malloch, Canad. J. Bot. 55: 1708. 1977.
Anamorph: unnamed *Geotrichum* species.

≡ *Endomyces geotrichum* E.E. Butler & L.J. Petersen, Mycologia 64: 367. 1972.
≡ *Dipodascus geotrichum* (E.E. Butler & L.J. Petersen) Arx, Antonie van Leeuwenhoek 43: 336. 1977.

Illustration: de Hoog *et al*. 1986: 81.

Asci are produced on wide hyphae, which locally become densely septate and develop gametangia at both sides of a septum. The gametangia fuse at the top and produce an ascus containing a single ascospore. *Ascospores* are broadly ellipsoidal, 6–9 × 7–10 µm, with an echinate inner wall and an irregular exosporium wall, often with a hyaline equatorial furrow. Note that the anamorph *Geo. candidum* has been attributed to this species (Butler & Peterson 1972, de Hoog *et al*. 1986), but this complex has now been subdivided, with the name *Geo. candidum* being maintained for the most prevalent species; see below.

Type: CBS 772.71 (ex-**holotype**), homothallic, ex soil, Puerto Rico. The indication of this strain as neotype for *Geo. candidum* (de Hoog *et al*. 1986: 85) is no longer applicable.

2. *Galactomyces reessii* (van der Walt) Redhead & Malloch, Canad. J. Bot. 55: 1708. 1977.
Anamorph: unnamed *Geotrichum* species.

≡ *Endomyces reessii* van der Walt, Antonie van Leeuwenhoek 25: 458. 1959.
≡ *Galactomyces reessii* (van der Walt) Redhead & Malloch, Canad. J. Bot. 55: 1708. 1977.
≡ *Dipodascus reessii* (van der Walt) Arx, Antonie van Leeuwenhoek 43: 338. 1977.

Illustration: de Hoog *et al*. 1986: 95.

Type: CBS 179.60 (ex-**holotype**), ex cold water, retting of *Hibiscus cannabinis*.

3. *Galactomyces citri-aurantii* E.E. Butler, Mycotaxon 33: 200. 1988.
Anamorph: *Geotrichum citri-aurantii* (Ferraris) E.E. Butler, Mycotaxon 33: 201. 1988.

Illustration: Butler *et al*. 1988: 203, 205.

Type: CBS 175.89 (ex-**holotype**) (MT A1), ex soil of orange orchard, **Zimbabwe**, × CBS 176.89 (MT A2), ex soil of orange orchard, California, **U.S.A.**

4. *Teleomorph*: unknown.
Anamorph: ***Geotrichum europaeum*** de Hoog & M.Th. Smith, **sp. nov.** MycoBank MB500127. Fig. 5.

Entity described for nDNA homology group C (Smith *et al*. 1995).

Geotricho candido simile. Hyphis ad 8 µm crassis, apicibus rotundatis. Ramis perpendicularibus in athroconidia rectangularia fragmentata. Conidia non vel parce inflata. Chlamydosporae ellipsoideae, hyalinae. Characteres physiologici et G+C acidi deoxyribonucleinici in Tabula 3. Typus vivus et exsiccatus CBS 866.68 (CBS H-13662) in CBS praeservatus.

Micromorphology similar to that of *Geo. candidum*. Expanding *hyphae* up to 8 µm wide, with rounded apices, bearing much narrower hyphae at nearly right angles, soon disarticulating into rectangular arthroconidia. *Conidia* mostly showing none or little inflation after liberation. Ellipsoidal, hyaline chlamydospore-like cells may be present.

Physiologically, the species is indistinguishable from *Gal. pseudocandidus*. However, it can be separated from the remaining species by positive growth on D-xylose and growth in the absence of vitamins and by lacking growth on maltose, cellobiose, soluble starch and D-mannitol as well as at 37 °C. The expansion growth ranges from 21–25 mm radius in 7 d. The G+C values determined from the derivative graph of the melting curve (two peaks) are 36.6 and 43.0 mol %.

Type: CBS 866.68 (CBS H-13662, **holotype**).

Ecology: the single strain was isolated from wheat-field soil in Germany.

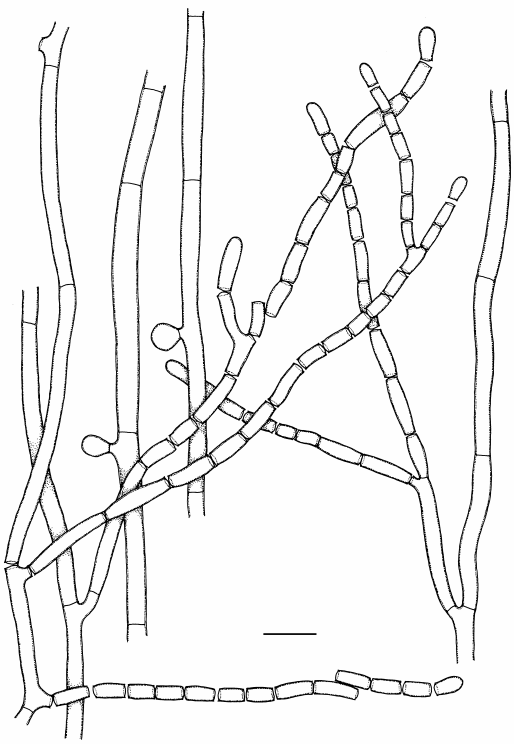

Fig. 5. *Geotrichum europaeum* (CBS 866.68). Hyphae with branching at more or less right angles and disarticulating into rectangular cells. Scale bar = 10 µm.

5. *Galactomyces pseudocandidus* de Hoog & M.Th. Smith, **sp. nov.** MycoBank MB500128. Fig. 6.
Anamorph: *Geotrichum pseudocandidum* Saëz, Mycopath. Mycol. Appl. 34: 363. 1968.

Entity described for nDNA homology group B (Smith *et al*. 1995).

Hyphis ad 5 µm crassis, apicibus rotundatibus. Ramis perpendicularibus in athroconidia rectangularia fragmentatis. Conidia non vel parce inflata. Chlamydosporae ellipsoideae, hyalinae. Asci ellipsoidei, subhyalini, monospori, crassitunicati. Ascosporae subglobosae, 5-6 µm diam, verrucosae, exosporio irregulariter inflato. Characteres physiologici et G+C acidi deoxyribonucleinici in Tabula 3. Typus vivus et exsiccatus CBS 820.71 (CBS H-13663) in CBS praeservatus.

Expanding *hyphae* up to 5 µm wide, with rounded apices, bearing much narrower hyphae at nearly right angles, soon disarticulating into rectangular arthroconidia. *Conidia* mostly showing none or little inflation after liberation. Ellipsoidal, hyaline chlamydospore-like cells may be present, which may develop into asci. *Asci* monopodal, ellipsoidal, often somewhat asymmetric, with firm, subhyaline walls, mostly remaining immature, or developing a single ascospore which is subspherical, 5–6 µm wide, rough-walled, with an irregular exosporium.

The species, with an anamorph indistinguishable from *Geotrichum europaeum*, can be distinguished from the remaining taxa by growth on D-xylose, by growth without vitamins, and by absence of growth on maltose, cellobiose, soluble starch and D-mannitol and at 37 °C. The expansion growth ranges from 20–32 mm radius in 7 d. The G+C values determined from the derivative graph of the melting curve (two peaks) are 36.7 and 43.4 mol %.

Type: CBS 820.71 (dried as CBS H-13663, **holotype**) (*teleomorph,* homothallic); CBS 626.83, ex-type strain of *Geo. pseudocandidum* (*anamorph*).

Ecology: soil, wood.

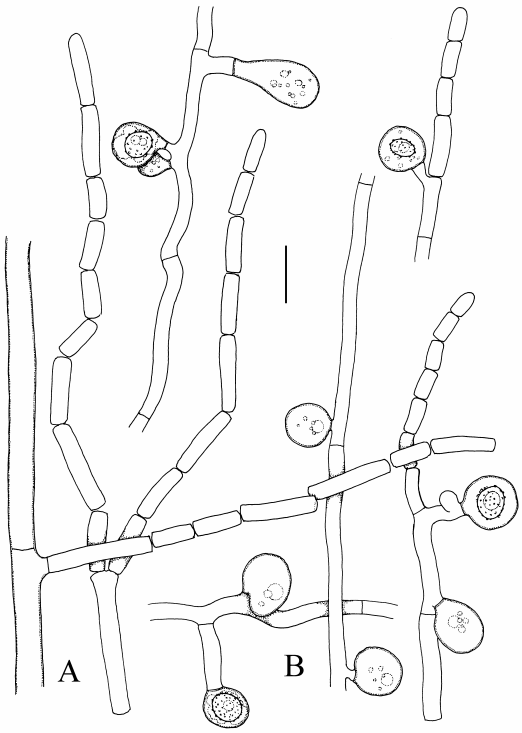

Fig. 6. *Galactomyces pseudocandidus*, CBS 820.71. A. Hyphae disarticulating into arthroconidia. B. Monopodal asci with immature and mature ascospores. Scale bar = 10 µm.

6. *Galactomyces candidus* de Hoog & M.Th. Smith, **sp. nov.** MycoBank MB500129. Fig. 7.
Anamorph: *Geotrichum candidum* Link : Fr., Mag. Naturf. Freunde, Berlin 9: 17. 1809; Syst. Mycol. 3: 420. 1832.

≡ *Geotrichum candidum* Link, Mag. Naturf. Freunde, Berlin 9: 17. 1809.
≡ *Botrytis geotricha* Link, Linn. Spec. Pl. 6-1: 53. 1824 (name change).
≡ *Acrosporium candidum* (Link) Sprengel, Syst. Veg. 16, ed. 4, p. 556. 1827.
≡ *Torula geotricha* (Link) Corda in Sturm, Deutschl. Fl. Abt. 3, 8: 79. 1829.
= *Mycoderma malti-juniperini* Desm., Ann. Sci. Nat. Bot. 10: 62. 1826.
= *Oidium lactis* Fresen., Beitr. Mykol 1: 23. 1850.
≡ *Oospora lactis* (Fresen.) Sacc., Syll. Fung. 4: 15. 1886.
≡ *Oosporoidea lactis* (Fresen.) Sumst., Myco-logia 5: 53. 1913.
= *Oidium lactis* Fresen. var. *luxurians* Reiss, Klotsch, Herb. Mykol. No. 1885. 1854.
= *Oospora lactis* (Fresen.) Sacc. var. *obtusa* (Thümen) Sacc., Syll. Fung. 4: 15. 1866.
≡ *Oidium obtusum* Thümen, Mycoth. Univ. No. 289. 1875.
= *Oidium nubilum* Weigmann & Wolff, Zentbl. Bakt. ParasitKde, Abt. 2, 22: 668. 1909.
≡ *Oospora nubila* (Weigmann & Wolff) Berkh., Schimmelgesl. *Monilia*, *Oidium*, *Oospora* en *Torula* p. 50. 1923.
= *Oidium humi* Mazé, Annls Inst. Pasteur 24: 407. 1910.
≡ *Oospora humi* (Mazé) Berkh., Schimmelgesl. *Monilia*, *Oidium*, *Oospora* en *Torula* p. 48. 1923.
= *Monilia asteroides* Castell., J. Trop. Med. Hyg. 17: 307. 1914.
≡ *Oidium asteroides* (Castell.) Castell. & Chalm., Man. Trop. Med., ed. 3, p. 1095. 1919.
≡ *Mycoderma asteroides* (Castell.) Brumpt, Précis Parasitol., ed. 3, p. 1076. 1922.
≡ *Geotrichum asteroides* (Castell.) Basgal, Contrib. Estudo Blast. Pulm., p. 48. 1931.
= *Oidium matelense* Castell., Lect. Higher Fungi Rel. Hum. Path., R. Coll. Phys., Lond. 1915.
≡ *Oospora matelensis* (Castell.) Berkh., Schimmelgesl. *Monilia*, *Oidium*, *Oospora* en *Torula* p. 46. 1923.
≡ *Mycoderma matelense* (Castell.) Brumpt, Précis Parasitol., ed. 3, p. 1084. 1922.
≡ *Pseudomycoderma matelensis* (Castell.) Cif., Arch. ProtistenKde 71: 436. 1930.
≡ *Geotrichum matelense* (Castell.) Castell., J. Trop. Med. Hyg. 35: 278. 1932.
≡ *Pseudomonilia matelensis* (Castell.) C.W. Dodge, Med. Mycol., p. 295, 1935.
≡ *Endomyces lactis* Windisch var. *matelensis* (Castell.) Windisch, Beitr. Biol. Pfl. 28: 123. 1951.
≡ *Trichosporon matelense* (Castell.) Cif., Anais Soc. Biol. Pernambuco 30: 140. 1955.
= *Oospora lactis* (Fresen.) Sacc. var. *parasitica* Pritchard & Porte, J. Agric. Res. 24: 898. 1923.
= *Oidium suaveolens* Krzemecki var. *minuta* Berkh., Versl. Vergad. Wis- en Natuurk. Afd. Kon. Akad. Wet. 32: 119. 1923.
≡ *Oospora fragrans* Berkhout var. *minuta* Berkh., Schimmelgesl. *Monilia*, *Oidium*, *Oospora* en *Torula* p. 47. 1923.
= *Oospora lactis* (Fresen.) Sacc. var. *exuberans* Stautz, Phytopath. Z. 3: 189. 1931.
= *Geotrichum matelense* (Castell.) Castell. var. *chapmanii* Castell., J. Trop. Med. Hyg. 35: 279. 1932.
= *Geotrichum javanense* Verona, Boll. Ist. Agr. Pisa 9: 480. 1933.
= *Geotrichum versiforme* M. Moore, Annls Mo. Bot. Gard. 21: 361. 1934.
= *Geotrichum redaellii* Negroni & Fischer, Revta Argent. Dermatosifil. 24: 147. 1940.
= *Endomyces lactis* Windisch, Beitr. Biol. Pfl. 28: 124. 1951.

= *Geotrichum novakii* El-Masry & Zsolt, Acta Biol., Szeged, N. Ser. 12: 69. 1966.

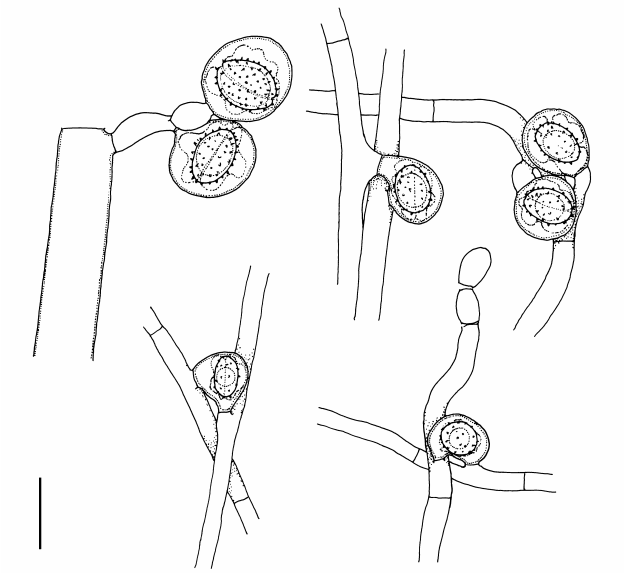

Fig. 7. *Galactomyces candidus*, CBS 180.33 × CBS 557.83. Predominantly bipodal asci with mature ascospores. Scale bar = 10 µm.

Entity described for nDNA homology group A (Smith *et al*. 1995).

Illustration: de Hoog *et al*. 1986: 82.

Asci ellipsoidei, subhyalini, monospori, crassitunicati. Ascosporae late ellipsoideae, 4.0-5.5 × 6-8 µm, verrucosae, exosporio irregulariter inflato, saepe zona hyalina cinctae. Characteres physiologici et G+C acidi deoxyribonucleinici in Tabula 3. Typus vivus et exsiccatus CBS 178.71 (CBS H-13664) in CBS praeservatus.

Asci are produced on undifferentiated hyphae without conspicuous septation, or on the same hypha; gametangia irregular. The gametangia fuse at the top and produce an ascus containing a single ascospore. *Ascospores* are broadly ellipsoidal, 4.0–5.5 × 6–8 µm, with an echinate inner wall and an irregular exosporium wall, often with a hyaline equatorial furrow. The species is homo- or heterothallic.

The species can be differentiated from all other arthroconidial species by growth at 35 °C and in vitamin-free medium, and by assimilation of D-xylose and D-mannitol. Growth is absent at 40 °C and on maltose, cellobiose and soluble starch. The expansion growth ranges from 15–43 mm radius in 7 d. The G+C values determined from the derivative graph of the melting curve (2 peaks) are 35.2 and 42.9 mol %.

This species represents the most common and widely distributed species of the *Geotrichum candidum* complex, known from a variety of substrates. The name *Geo. candidum* has become in general use for the anamorph particularly since the publications of Morenz (1963, 1964); it encompasses a number of well-known synonyms such as *Oidium lactis* and *Geotrichum javanense*.

Types: CBS 178.71 (CBS H-13664, homothallic, ex-**holotype** strain of teleomorph), from soil polluted with oil, **Germany**; CBS 615.84 (CBS H-13685, **neotype** of anamorph designated here), brie cheese, **France**.

Ecology: ubiquitous species in slurries rich in fat such as pig fodder, milk and milk-related products (Prillinger *et al*. 1999). It is occasionally encountered as an opportunist e.g. in the human digestive tract (de Hoog *et al*. 2000).

Teleomorph and anamorph species belonging to the *Dipodascus* clade

Dipodascus de Lagerheim, Jahrb. Wiss. Bot. 24: 561. 1892.
Anamorph: *Geotrichum* Link : Fr.

Thallus growth mostly initiating from a single, inflated cell. *Hyphae* branched at broad or right angles, with rounded apices, disarticulating into arthroconidia. Some additional sympodial conidiogenesis from undifferentiated cells may be present. *Chlamydospores* may be produced. *Gametangia* formed with on opposite sides of hyphal septa, clavate, soon fusing at apex and forming ascus. *Asci* hyaline, broadly cylindrical to tubular or acicular, containing eight to more than 100 ascospores. *Ascospores* broadly ellipsoidal or cylindrical, hyaline, smooth-walled, each with an even, thick gelatinous coat. Ascospores liberated through apical rupture of the firm ascus wall. Cell walls 3-layered, without xylose or fucose. Septa perforated by micropores. Xylose assimilated. Nitrate not assimilated. Urease not present. Extracellular starch not produced. Diazonium blue B reaction negative. SSU rDNA without deletions, ribosomal Group 1 (Ueda-Nishimura & Mikata 2000).

Type: *Dipodascus albidus* de Lagerheim (teleomorph). Associated anamorph genus: *Geotrichum.*

1. *Dipodascus albidus* de Lagerheim, Jahrb. Wiss. Bot. 24: 561. 1892.
Anamorph: unnamed *Geotrichum* species.

Illustration: de Hoog *et al*. 1986: 18–19.

Type: CUP 20140, **neotype**; *representative strain*: CBS 766.85, exudates of angiosperm tree, Japan.

2. *Dipodascus australiensis* Arx & Barker, Antonie van Leeuwenhoek 43: 335. 1977.
Anamorph: unnamed *Geotrichum* species.

Illustration: de Hoog *et al*. 1986: 22–23.

Type: CBS 625.74 ex-**holotype**, decaying cladode of *Opuntia inermis*, **Australia**.

3. *Dipodascus aggregatus* Francke-Grosmann, Med. Stat. SkogsvFör. Inst. 41: 30. 1952.
Anamorph: unnamed *Geotrichum* species.

Illustration: de Hoog *et al.* 1986: 29–30.

Type: CBS 175.53, ex-**holotype**, pupal galleries of *Ips acuminatus* in *Pinus sylvestris,* **Germany**.

4. *Dipodascus geniculatus* de Hoog, M.Th. Smith & Guého, Stud. Mycol. 29: 25. 1986.
Anamorph: none.

Illustration: de Hoog *et al*. 1986: 26.

Type: CBS 184.80, ex-**holotype**, pulp *Psidium guajava,* Maharashtra, **India**.

5. *Teleomorph*: unknown.
Anamorph: ***Geotrichum fermentans*** (Diddens & Lodder) Arx, Stud. Mycol. 14: 32. 1977.

≡ *Trichosporon fermentans* Diddens & Lodder, Anaskospor. Hefen, 2. Hälfte, p. 488, 1942
≡ *Fermentotrichon fermentans* (Diddens & Lodder) Novák & Zsolt, Acta Bot. Hung. 7: 131, 1961.

Illustration: de Hoog *et al*. 1986: 44.

Type: CBS 439.83, ex-**holotype**, woodpulp mill, **Sweden**.

6. *Dipodascus armillariae* W. Gams, Sydowia 36: 50. 1983.
Anamorph: *Geotrichum decipiens* (L. Tul. & R. Tul.) W. Gams.

Illustrations: Gams 1983: 50; de Hoog *et al.* 1986: 33–34.

Types: CBS 817.71 (ex-**holotype** of teleomorph), *Armillaria mellea*, **Netherlands**; dried specimen in herb. in PC (Prague) (**holotype** of anamorph); *representative strain*: CBS 817.71.

7. *Teleomorph*: unknown.
Anamorph: ***Geotrichum restrictum*** de Hoog & M.Th. Smith, **sp. nov.** MycoBank MB500130. Fig. 8.

Geotricho klebahnii simile. Hyphis ad 5 µm latis, apicibus rotundatis, parce ramosis, in athroconidia cylindrica fragmentata. Conidia saepe inflata. Characteres physiologici et G+C acidi deoxyribonucleinici in Tabula 3. Typus vivus et exsiccatus CBS 111234 (CBS H-13665) in CBS praeservatus.

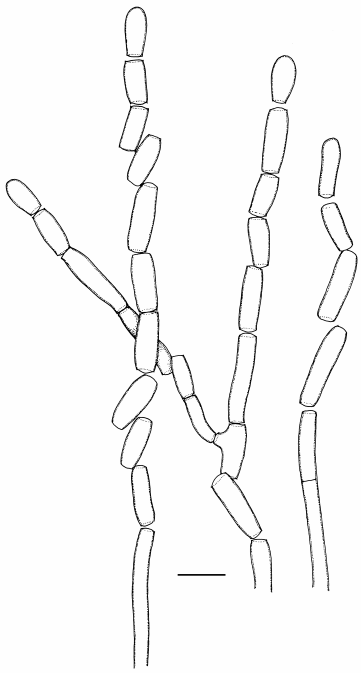

Fig. 8. *Geotrichum restrictum* (CBS 111234). Disarticulating hyphae. Scale bar = 10 µm.

Micromorphology of anamorph similar to that of *Geo. klebahnii*. Expanding hyphae up to 5 µm wide, with rounded apices, poorly branched, soon disarticulating into cylindrical arthroconidia. Conidia mostly inflating after liberation.

Differentiation of this species from all other arthroconidial taxa is by absence of growth at 30 ^{0}C and by assimilation of D-xylose. Growth is absent on maltose, cellobiose and soluble starch, DL-lactate and in vitamin-free medium. The expansion growth is limited, reaching 2–3 mm radius in 7 d. The G+C values determined from the derivative graph of the melting curve (two peaks) are 17.0 and 42.7 mol %.

Type: CBS 111234 (CBS H-13665, **holotype**), ex *Picea abies* (endophytic), **Sweden**.

Ecology: known from a single strain that was isolated as an endophyte of *Picea abies* in Sweden.

8. *Teleomorph*: unknown.
Anamorph: ***Geotrichum klebahnii*** (Stautz) Morenz, Mykol. Schriftenreihe 2: 36. 1964.

≡ *Trichosporon klebahnii* Stautz, Phytopath. Z. 3: 189. 1931
≡ *Endomyces lactis* (Fresen.) Windisch var. *klebahnii* (Stautz) Windisch, Beitr. Biol. Pfl. 28: 125, 1951
= *Trichosporon penicillatum* do Carmo Sousa, Antonie van Leeuwenhoek 31: 153. 1965.
≡ *Geotrichum penicillatum* (do Carmo Sousa) Arx, Stud. Mycol. 14: 32. 1977.

Illustration: de Hoog *et al*. 1986: 65.

Type: CBS 179.30 (ex-**holotype**), slime flux of *Taxus baccata*.

9. *Dipodascus macrosporus* Madelin & Feest, Trans. Br. Mycol. Soc. 79: 331. 1982.
Anamorph: none.

Illustration: de Hoog *et al*. 1986: 13–14.

Type: CBS 259.82 (ex-**holotype**), *Badhamia utricularis,* slime trail plasmodium, **U.K.**

Teleomorph and anamorph species belonging to the *Magnusiomyces* clade

Magnusiomyces Zender, Bull. Soc. Bot. Genève, Ser. 2, 17: 41. 1925.
Anamorph: *Saprochaete* Coker & Shanor ex D.T.S. Wagner & Dawes, Mycologia 62: 794. 1970.

= *Endyllium* Clements, Gen. Fungi p. 245. 1931 (name change for *Magnusiomyces*).
= *Zendera* Redhead & Malloch, Canad. J. Bot. 55: 1707. 1977 (type: *Endomyces tetrasperma*).
= *Blastoschizomyces* Salkin, Gordon, Samsonoff & Rieder, Mycotaxon 22: 503. 1985 (type: *B. pseudotrichosporon* = *S. capitata*).

Thallus growth initially mostly pseudomycelial with inflated cells. *Hyphae* branched at acute angles, with acuminate apices, disarticulating into arthroconidia. Additional sympodial and some annellidic conidiogenesis may be present. *Chlamydospores* mostly absent. *Gametangia* formed on opposite sides of hyphal septa, broadly ellipsoidal, soon fusing entirely and being transformed to an ascus. *Asci* hyaline, subspherical to broadly ellipsoidal, containing four ascospores. *Ascospores* ellipsoidal to broadly ellipsoidal, hyaline, smooth-walled, each with an even, thick gelatinous coat. Ascospores liberated through apical rupture of the firm ascus wall. Cell walls 3-layered, without xylose or fucose. Septa perforated by micropores. Xylose mostly not assimilated. Nitrate not assimilated. Urease not present. Extracellular starch not produced. Diazonium blue B reaction negative. SSU rDNA with deletions in V2, V3 and V8: ribosomal Group 2 (Ueda-Nishimura & Mikata 2000).

Types: *Endomyces magnusii* (*Magnusiomyces* teleomorph); *Saprochaete saccharophila* (*Saprochaete* anamorph).

1. *Magnusiomyces starmeri* (Phaff, Blue, Hagler & Kurtzman) de Hoog & M.Th. Smith, **comb. nov.** MycoBank MB500131.
Anamorph: unnamed *Saprochaete* species.

≡ *Dipodascus starmeri* Phaff, Blue, Hagler & Kurtzman, Int. J. Syst. Bacteriol. 47: 309. 1997 (basionym).

Illustration: Phaff *et al*. 1997: 310.

Type: CBS 780.96 (ex-**holotype**), rotting saguaro plant, Arizona, **U.S.A.**

2. *Magnusiomyces ovetensis* (Pelãez & C. Ramírez) de Hoog & M.Th. Smith, **comb. nov.** MycoBank MB500132.
Anamorph: ***Saprochaete sericea*** (Stautz) de Hoog & M.Th. Smith, **comb. nov.** MycoBank MB500133.

= *Endomyces ovetensis* Pelãez & C. Ramírez, Microbiol. Espagn. 9: 191. 1956 (basionym of teleomorph).
  ≡ *Endomycopsis ovetensis* (Pelãez & C. Ramírez) Kreger-van Rij, Taxon. Stud. Gen. *Endomycosis, Pichia* and *Debaryomyces* p. 48. 1964.
  ≡ *Zendera ovetensis* (Pelãez & C. Ramírez) Redhead & Malloch, Canad. J. Bot. 55: 1017. 1977.
  ≡ *Dipodascus ovetensis* (Pelãez & C. Ramírez) von Arx, Antonie van Leeuwenhoek 43: 338. 1977.
= *Dipodascus ambrosiae* de Hoog, M.Th. Smith & Guého, Stud. Mycol. 29: 47. 1989.
= *Oospora sericea* Stautz, Phytopath. Z. 3: 193. 1931 (basionym of anamorph).
  ≡ *Trichosporon sericeum* (Stautz) Diddens & Lod, der, Anaskosp. Hefen, 2. Hälfte p. 448. 1942.
  ≡ *Ascotrichosporon sericeum* (Stautz) Kocková-Kratochvílová, Sláviková, Zemek & Kuniak, Proc. 5th Int. Spec. Symp. Yeasts, Bratislava p. 9. 1977.
  ≡ *Geotrichum sericeum* (Sautz) de Hoog, M.Th. Smith & Guého, Stud. Myol. 29: 36. 1986.

Illustrations: de Hoog *et al*. 1986: 37 (teleomorph), de Hoog *et al*. 1986: 38 (anamorph).

Types: CBS 192.55 (ex-**holotype** of teleomorph), tannin concentrate, **Spain**; CBS 634.85 (ex-**holotype** of anamorph), slime flux in *Quercus* sp., **Germany**.

3. *Magnusiomyces tetrasperma* (Macy & M.W. Miller) de Hoog & M.Th. Smith, **comb. nov.** MycoBank MB500134.
Anamorph: none.

= *Endomyces tetrasperma* Macy & Miller, J. Bact. 105: 638. 1971 (basionym).

≡ *Zendera tetrasperma* (Macy & Miller) Redhead & Malloch, Can. J. Bot. 55: 1707. 1977.
≡ *Dipodascus tetrasperma* (Macy & Miller) Arx, Antonie van Leeuwenhoek 43: 338. 1977.

Illustration: de Hoog *et al*. 1986: 68.

Type: CBS 765.70 (ex-**holotype**), wet conveyer, California, U.S.A.

4. *Magnusiomyces magnusii* (F. Ludwig) de Hoog & M.Th. Smith, **comb. nov.** MycoBank MB500135.
Anamorph: *Saprochaete ludwigii* (Hansen) de Hoog & M.Th. Smith, **comb. nov.** MycoBank MB500136.

= *Endomyces magnusii* F. Ludwig, Ber. Dt. Bot. Ges. 4: 17. 1886 (basionym of teleomorph).
≡ *Endyllium magnusii* (F. Ludwig) Clements in Clements & Shear, Gen. Fung. p. 245. 1931.
≡ *Magnusiomyces magnusii* (F. Ludwig) Redhead & Malloch, Can. J. Bot. 55: 1708. 1977.
≡ *Dipodascus magnusii* (F. Ludwig) Arx, Antonie van Leeuwenhoek 43: 336. 1977.
= *Oidium ludwigii* E.C. Hansen, Zentbl. Bakt. ParasitKde, Abt. 2, 7: 185.1901 (basionym of anamorph).
≡ *Oospora ludwigii* (E.C. Hansen) Sacc. & D. Sacc., Syll. Fung. 18: 500. 1906.
≡ *Geotrichum ludwigii* (E.C. Hansen) S. Fang, T.C. Yen & J.C. Yen, Acta Microbiol. Sin. 12: 69. 1966.
= *Oospora magnusii* Stautz, Phytopath. Z. 3: 185. 1931.

Illlustration: de Hoog *et al*. 1986: 71-72.

Types: CBS 108.12, possibly ex-**holotype** of teleomorph, but unknown; type of anamorph unknown, *representative culture* CBS 108.12.

5. *Magnusiomyces spicifer* (de Hoog, M.Th. Smith & Guého) de Hoog & M.Th. Smith, **comb. nov.** MycoBank MB500137.
Anamorph: unnamed *Saprochaete* species.

≡ *Dipodascus spicifer* de Hoog, M.Th. Smith & Guého, Stud. Mycol. 29: 60. 1986 (basionym).

Illustration: de Hoog *et al.* 1986: 61.

Type: CBS 244.85 (ex-**holotype**), cactus rot, Arizona, **U.S.A.**

6. *Magnusiomyces capitatus* (de Hoog, M.Th. Smith & Guého) de Hoog & M.Th. Smith, **comb. nov.** MycoBank MB500138.
Anamorph: *Saprochaete capitata* (Diddens & Lodder) de Hoog & M.Th. Smith, **comb. nov.** MycoBank MB500139

= *Dipodascus capitatus* de Hoog, M.Th. Smith & Guého, Stud. Mycol. 29: 51. 1986 (basionym of teleomorph).
= *Trichosporon capitatum* Diddens & Lodder, Die anaskosporogenen Hefen, 2. Hälfte, p. 488. 1942 (basionym of anamorph).
≡ *Geotrichum capitatum* (Diddens & Lodder) von Arx *in* von Arx, Rodrigues de Miranda, M.Th. Smith & Yarrow, Stud. Mycol. 14: 32. 1977.
≡ *Ascotrichosporon capitatum* (Diddens & Lodder) Kocková-Kratochvilová, Sláviková, Zemek & Kuniak, Proc. 5th Int. Spec. Symp. Yeasts, Bratislava p. 9. 1977 (invalid).
≡ *Blastoschizomycese capitatus* (Diddens & Lodder) Salkin, Gordon, Samsonoff & Rieder, Mycotaxon 22: 378. 1985.
= *Sporotrichum spicatum* Delitsch, Syst. Schimmelp. p. 106. 1943.
= *Geotrichum linkii* Vöros-Felkai, Acta Microbiol. Hung. 8: 95. 1961 (nom. inval.).
= *Blastoschizomyces pseudotrichosporon* Salkin, Gordon, Samsonoff & Rieder, Mycotaxon 22: 503. 1985.

Illustration: de Hoog *et al.* 1986: 52.

Types: CBS 162.80 (= CBS H-14215, **holotype** of teleomorph), bovine mastitis milk, **U.K.**; tester strains CBS 197.35 (MT a), woodpulp, Sweden, and CBS 580.82 (MT α), sputum of man; CBS 571.82 (ex-**lectotype** strain of anamorph), woodpulp, **Sweden**.

7. *Teleomorph*: unknown.
Anamorph: ***Saprochaete suaveolens*** (Krzemecki) de Hoog & M.Th. Smith, **comb. nov.** MycoBank MB500140.

≡ *Oidium suaveolens* Krzemecki, Zentbl. Bakt. ParasitKde, Abt. 2, 38: 577. 1913 (basionym).
≡ *Geotrichum suaveolens* (Krzemecki) S. Fang, TC. Yen & J.C. Yen, Acta Microbiol. Sin. 12: 68. 1966 (invalid; Art. 64 ICBN).
≡ *Oospora fragrans* Berkh., Schimmelgesl. *Monilia, Oidium, Oospora* en *Torula* p. 47. 1923 (name change; non *Oospora suaveolens*).
≡ *Cylindrium fragrans* (Berkh.) Burns, Iowa St. Coll. J. Sci. 7: 436. 1933.

≡ *Endomyces lactis* (Fresen.) Windisch var. *fragrans* (Berkh.) Windisch, Beitr. Biol. Pfl. 28: 125. 1951.

≡ *Geotrichum fragrans* (Berkh.) Morenz, Mykol. SchrReihe 1: 69, 1963.

= *Geotrichum fici* Goto, Yamakawa & Yokotsuka, J. Agric. Chem. Soc. Japan 49: 522. 1975.

Type: CBS 152.25 (ex-**holotype**), from water in brewery.

8. *Teleomorph*: unknown.
Anamorph: ***Saprochaete gigas*** (J. Smit & L. Meyer) de Hoog & M.Th. Smith, **comb. nov.** MycoBank MB500141. Fig. 9.

≡ *Oospora gigas* J. Smit & L. Meyer, Ned. Tijdschr. Microbiol. Serol. 2: 86. 1928 (basionym).

≡ *Geotrichum gigas* (J. Smit & L. Meyer) M.Th. Smith & Poot, Antonie van Leeuwenhoek 77: 77. 2000.

= *Geotrichum magnum* Saëz, Microbiol. Espagn. 21: 203. 1968.

= *Geotrichum rectangulatum* Goto, Yamakawa & Yokotsuka, J. Agr. Chem. Soc. 49: 523. 1975.

Illustration: de Hoog *et al*. 1986: 76.

Micromorphology similar to that of S. *fragrans*. Thallus very large; expanding hyphae straight with narrower lateral hyphae at acute angles, with acuminate apices. Main hyphae up to 12 µm wide, remotely septate, disarticulating. Cells mostly germinate through the scar, sometimes repeatedly, leading to some vague annellations.

Type: CBS 140.25 (ex-**holotype**), juice of *Arenga saccharifera.*

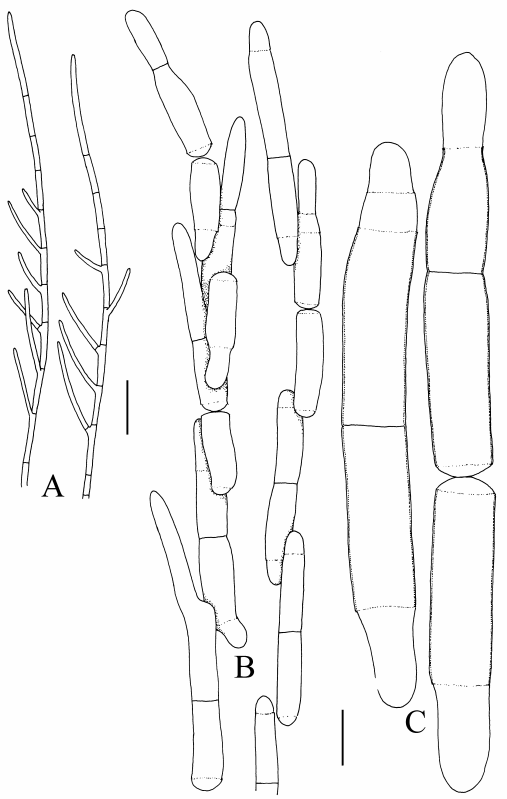

Fig. 9. *Saprochaete gigas* (CBS 140.25). A. Developing thallus with expanding hyphae with branches at acute angles. Scale bar = 100 µm. B. Hyphae disarticulating into separate cells. Scale bar = 25 µm. C. Detail of individual arthroconidia with vague annellations. Scale bar = 10 µm.

9. *Teleomorph*: unknown.
Anamorph: ***Saprochaete chiloënsis*** (C. Ramírez & González) Kurtzman, Robnett, de Hoog & M.Th. Smith, **comb. nov.** MycoBank MB500142. Fig. 10.

≡ *Schizoblastosporion chiloënse* C. Ramírez & González, Mycopathologia 88: 168. 1984 (basionym).

Micromorphology of anamorph similar to that of S. *ingens*. Thallus large, profusely branched at acute angles, pseudomycelial, consisting of series of ellipsoidal cells connected at narrow isthmi; disarticulation with reluctance. Central cells up to 15 × 8 µm, terminal cells much smaller. Chains expanding by apical, blastic growth.

Type: CBS 8187 (ex-**holotype**), rotten trunk of *Eucryphia cordifolia*, **Chile**.

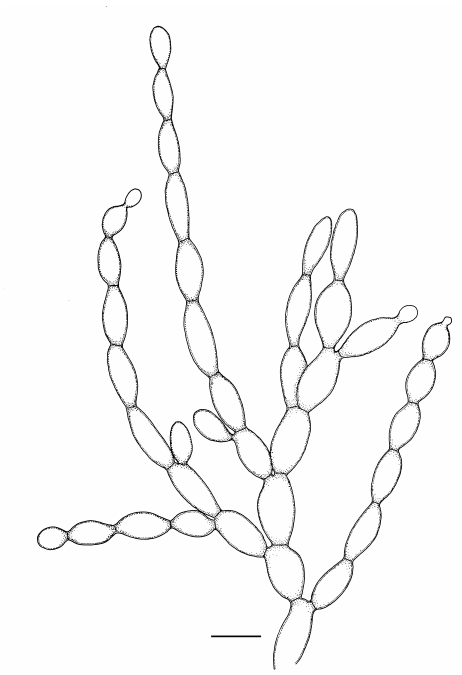

Fig. 10. *Saprochaete chiloënsis* (CBS 8187). Strongly branched pseudohyphal system. Scale bar = 10 µm.

10. *Teleomorph*: unknown.
Anamorph: ***Saprochaete clavata*** (de Hoog, M.Th. Smith & Guého) de Hoog & M.Th. Smith, **comb. nov.** MycoBank MB500143.

≡ *Geotrichum clavatum* de Hoog, M.Th. Smith & Guého, Stud. Mycol. 29: 57. 1986 (basionym).

Illustration: de Hoog *et al*. 1986: 58.

Type: CBS 425.71 (ex-**holotype**), human lung tissue, **U.S.A.**

11. *Teleomorph*: unknown.
Anamorph: ***Saprochaete saccharophila*** Coker & Shanor ex D.T.S. Wagner & Dawes. Fig. 11.

≡ *Saprochaete saccharophila* Coker & Shanor, J. Elisha Mitchell Sci. Soc. 55: 163. 1939 (without Latin diagnosis).
≡ *Saprochaete saccharophila* Coker & Shanor ex D.T.S. Wagner & Dawes, Mycologia 62: 794. 1970 (basionym).

Micromorphology similar to that of *S. fragrans*. Thallus very large, often initiating with short series of inflated cells; expanding hyphae straight with narrower, repeatedly branched lateral hyphae at acute angles, with acuminate apices. Main hyphae up to 10 µm wide, remotely septate, reluctantly disarticulating. Cells may germinate up to 3 times next to or through the scar, leading to short sympodula or some vague annellations.

Lectotype: Coll. L. Shanor (UNC), stream near sawdust pile, North Carolina, **U.S.A.**, 1938; *representative strain*: CBS 252.91.

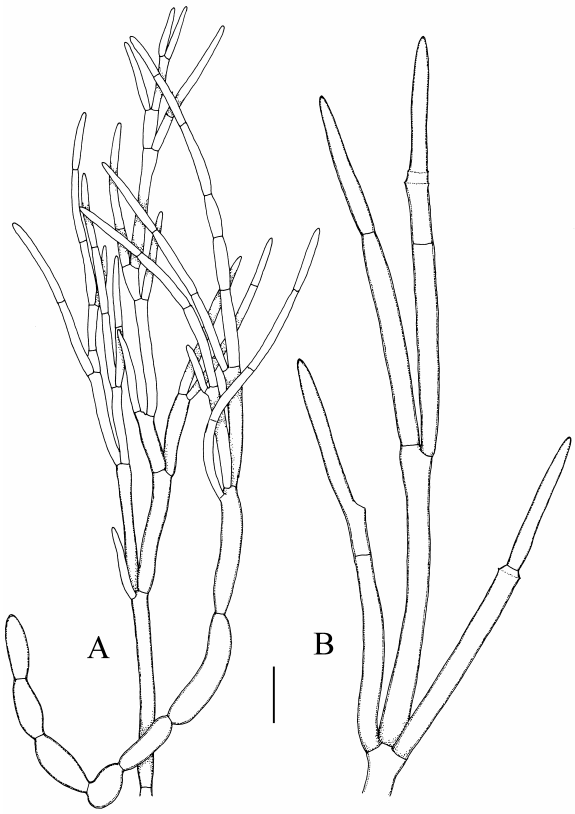

Fig. 11. *Saprochaete saccharophila* (CBS 252.91). A. Hyphal system. Scale bar = 25 µm. B. Detail of hyphal cells with percurrent growth. Scale bar = 10 µm.

The species *Saprochaete saccharophila* was invalidly described by Coker & Shanor (1939) as a saprophytic fungoid alga in spite of absence of chlorophyll and any trace of plastids. Judging from morphological properties such as type of branching and rhizoidal attachment system and cell wall composition, the organism was classified close to the *Chaetophoraceae*. Original isolates of Coker and Shanor have not been preserved. However, according Von Stosch (1966), the clear description of the species by Coker & Shanor (1939) made identification of fresh isolates possible. Von Stosch (1967) reported the rediscovery of *S. saccharophila* in a bog pool in Fulda (Hessen, Germany). These isolates were examined by Dawes (1969) for their ultrastructure, X-ray diffraction patterns and cell wall composition. From these observations it appeared that *S. saccharophila* is probably a fungus rather than an alga, because mitochondria, nuclei, lipid bodies, endoplasmic reticulum, and microtubules were similar to those of fungal cells. Wagner & Dawes (1970) validated the species name based on Coker & Shanor's (1939) original material by providing a Latin diagnosis, without further taxonomic assignment to any fungal group. On basis of ITS sequence data, the isolate CBS 252.91 originating from Von Stosch was found in the present paper to match *Magnusiomyces* ribosomal Group 2 (Ueda-

Nishimura & Mikata 2000). Not being reported in previous publications, the physiological characteristics are listed in Table 3.

Fig. 12. *Saprochaete saccharophila,* Sample locality of CBS 412.95, on plants in creek, Groeneveld, Baarn, The Netherlands; W. Gams.

12. ***Magnusiomyces ingens*** (de Hoog, M.Th. Smith & Guého) de Hoog & M.Th. Smith, **comb. nov.** MycoBank MB500144.

Anamorph: unnamed *Saprochaete* species (non *Saprochaete ingens*, see below).

≡ *Dipodascus ingens* de Hoog, M.Th. Smith & Guého, Mycotaxon 63: 345. 1997 (basionym).

Illustration: de Hoog *et al*. 1986: 41, as *D. ingens*.

Entity described for *Dipodascus ingens* reassociation group A (Smith & Poot 2003).

The name is introduced for *Dipodascus ingens* de Hoog *et al*. that was described for the supposed teleomorph of *Candida ingens* van der Walt & van Kerken (de Hoog *et al*. 1997). In the present study these taxa appeared to be separate.

The species can be separated physiologically from the remaining taxa by assimilation of L-sorbose, growth at 35 °C and in the absence of vitamins, while growth is absent on D-xylose, sucrose, raffinose, D-glucitol, D-mannitol and at 37 °C. The expansion growth ranges from 3–7 mm in 7 d. The G+C value determined from the derivative graph of the melting curve (single peak) is 43.6 mol %.

Type: CBS 521.90 = CBS 4827 MT a, wine cellar, **South Africa**, × CBS 523.90 = CBS 7197 MT alpha, unknown (ex-**holotype**).

Ecology: species was found in a wine cellar in South Africa.

13. *Teleomorph*: unknown.

Anamorph: ***Saprochaete quercus*** de Hoog & M.Th. Smith, **sp. nov.** MycoBank MB500145. Fig. 13.

Entity described for *Dipodascus ovetensis* reassociation group A (Smith & Poot 2003).

Hyphis ad 5 µm latis, regulariter ramosis, in athroconidia cylindrica fragmentatis. Conidia paulo inflata. Characteres physiologici et G+C acidi deoxyribonucleinici in Tabula 3. Typus vivus et exsiccatus CBS 752.85 (CBS H-13666) in CBS, Utrecht, praeservatus.

Micromorphology: regularly branched hyphae, up to 5 µm wide, gradually disarticulating into smaller entities, with slight inflation of liberated cells in a later stage.

This species is physiologically similar to *Magnusiomyces ingens* from which it can be distinguished by absence of growth at 35 °C. It differs from the remaining taxa by growth at 30 °C and in the absence of vitamins, while no growth is observed on D-xylose, sucrose, raffinose, D-glucitol, D-mannitol and at 35 °C. The expansion growth ranges from 5–9 mm diam in 7 d. The G+C values determined from the derivative graph of the melting curve (2 peaks) are 39.8 and 48.7 mol %.

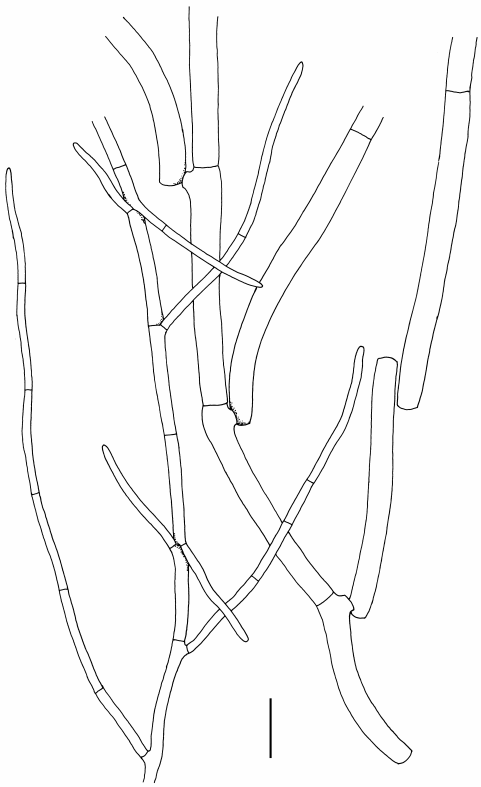

Fig. 13. *Saprochaete quercus*, CBS 752. 85. Hyphae reluctantly disarticulating into separate cells. Scale bar = 10 µm.

Type: CBS 752.85 (CBS H-13666, **holotype**), slime flux of *Quercus rubra,* Ontario, **Canada**.

Ecology: strains have been isolated from slime flux of *Quercus rubra*.

14. *Teleomorph*: unknown.
Anamorph: ***Saprochaete japonica*** de Hoog & M.Th. Smith, **sp. nov.** MycoBank MB500146. Fig. 14.

Entity described for *Geotrichum* sp. B (Smith *et al.* 2000).

Saprochaeti suaveolenti similis. Thallus latus, undulatus. Hyphis ad 10 µm crassis, apicibus acuminatibus. Ramis bifurcatis, semper in athroconidiis ellipsoideis dissolutis. Conidia inflata, apicibus rotundatibus. Caractera fysiologica et G+C acidi deoxyribonucleati presentes in Tabula 3. Typus vivus et exsiccatus CBS 100158 (CBS H-13667) in CBS, Utrecht, praeservatur.

Micromorphology similar to that of *S. suaveolens*. Thallus large, consisting of minutely undulate expanding hyphae, bearing narrower lateral hyphae at acute angles, with acuminate apices. Main hyphae up to 10 µm wide, quickly disarticulating and becoming pseudomycelial, developing profuse backward branching. Scars inconspicuous, annellations very rare. Cells rounding off after liberation.

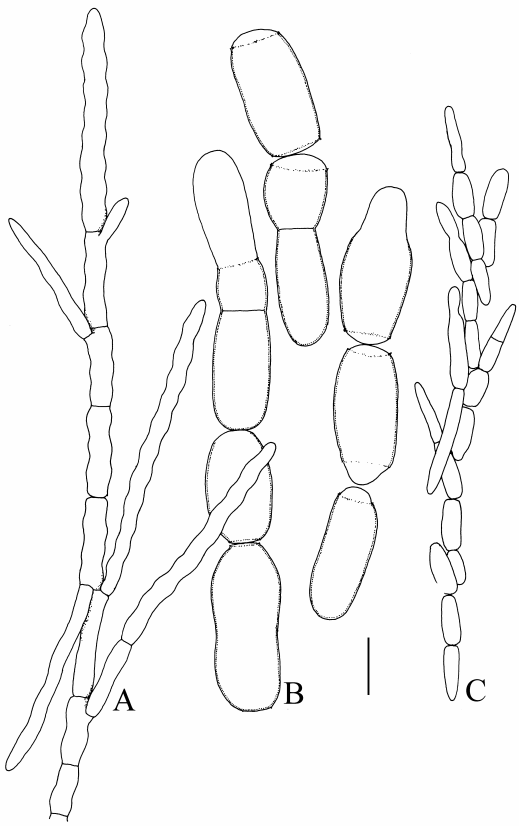

Fig. 14. *Saprochaete japonica*, CBS 100158. A. Young hyphal system. B, C. Disarticulating pseudomycelium at different magnifications. Scale bar = 25 µm (for A, C) and 10 µm (for B).

The species can be separated physiologically from the remaining taxa by assimilation of D-glucitol, D-mannitol and citrate, while growth is absent on D-xylose, sucrose, raffinose and at 35 °C. The expansion growth ranges from 8–10 mm in 7 d. The G+C value determined from the derivative graph of the melting curve (single peak) is 42.2 mol %.

Type: CBS 100158 (CBS H-13667, **holotype**), exudates of tree.

Ecology: exudate of trees.

15. *Teleomorph*: unknown.
Anamorph: ***Saprochaete fungicola*** de Hoog & M.Th. Smith, **sp. nov.** MycoBank MB500147. Fig. 15.

Entity described for *Dipodascus aggregatus* reassociation group B (Smith & Poot 2003).

Geotricho klebahnii similis. Hyphis ad 3 µm latis, ad apices rotundatis, parce ramosis, in athroconidia rectangularia fragmentatis. Conidia inflata, late ellipsoidea. Characteres physiologici et G+C acidi deoxy-ribonucleinici in Tabula 3. Typus vivus et exsiccatus CBS 625.85 (CBS H-13668) in CBS, Utrecht, praeservatur.

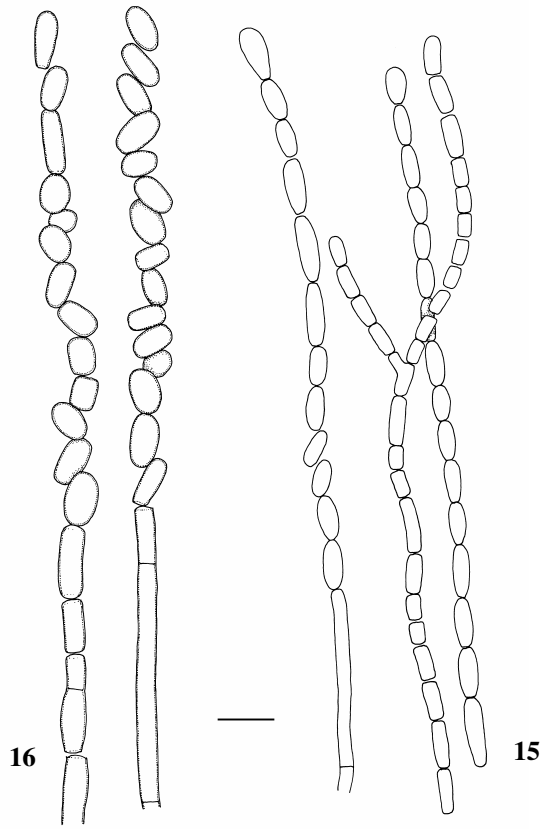

Fig. 15 (right). *Saprochaete fungicola*, CBS 625.85. Disarticulating hyphae. Bar = 10 µm.
Fig. 16 (left). *Saprochaete psychrophila*, CBS 765.85. Disarticulating hyphae. Bar for both figures = 10 µm.

Micromorphology similar to that of *Geo. klebahnii.* Expanding hyphae up to 3 µm wide, with rounded apices, poorly branched, soon disarticulating into rectangular arthroconidia that tend to inflate to broadly ellipsoidal.

The species can be recognized by assimilation of D-xylose and D-mannitol and growth at 35 °C and by absence of growth on maltose, soluble starch, cellobiose, at 40 °C and in vitamin-free medium. The expansion growth ranges from 7–8 mm radius in 7 d. The G+C value determined from the derivative graph of the melting curve (single peak) is 41.3 mol %.

Type: CBS 625.85 (CBS H-13668, **holotype**), *Nectria cinnabarina,* **Russia**.

Ecology: isolated from carpophores of *Nectria cinnabarina*.

16. *Teleomorph*: unknown.
Anamorph: ***Saprochaete psychrophila*** de Hoog & M.Th. Smith, **sp. nov.** MycoBank MB500148. Fig. 16.

Entity described for *Dipodascus aggregatus* reassociation group A (Smith & Poot 2003).

Saprochaeti fungicolae similis, sed hyphae vulgo latiores, ad 5 µm, ad apices rotundatae, parce ramosae, in athroconidia cylindrica fragmentatae. Conidia inflata, late ellipsoidea. Characteres physiologici et G+C acidi deoxyribonucleinici in Tabula 3. Typus vivus et exsiccatus CBS 765.85 (CBS H-13669) in CBS, Utrecht, praeservatur.

Micromorphology similar to that of *S. fungicola* but significantly larger. Expanding hyphae up to 5 µm wide, with rounded apices, poorly branched, soon disarticulating into cylindrical arthroconidia that tend to inflate to broadly ellipsoidal.

This species is physiologically similar to *Saprochaete fungicola* from which it can be distinguished by absence of growth at 30 C. The species is separated from remaining species by assimilation of D-xylose and DL-lactate and by absence of growth on maltose, soluble starch, cellobiose, at 30 ^{0}C and in vitamin-free medium. The expansion growth ranges from 4–5 mm radius in 7 d. The G+C value determined from the derivative graph of the melting curve (single peak) is 43.4 mol %.

Type: CBS 765.85 (CBS H-13669, **holotype**), slime flux in *Pinus ponderosa,* **U.S.A.**

Ecology: slime flux of *Pinus ponderosa.*

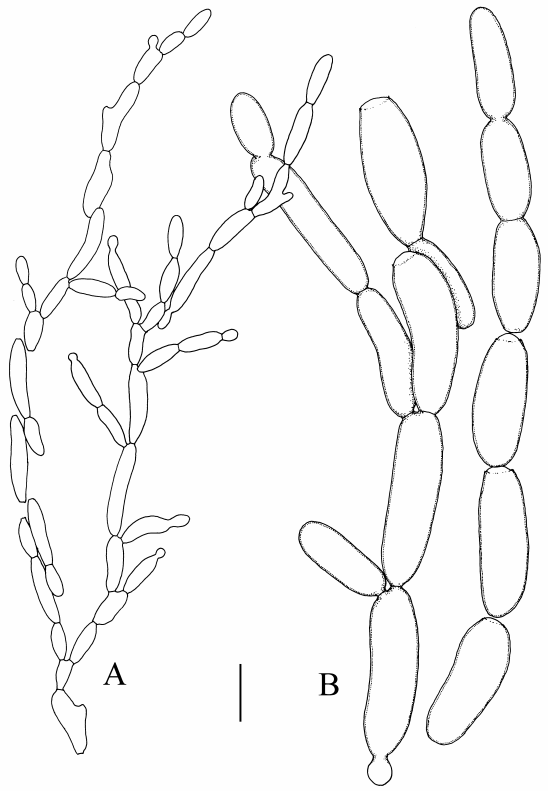

Fig. 17. *Saprochaete ingens* (CBS 517.90). A, B. Disarticulating pseudomycelium at different magnifications. Scale bar = 25 µm (for A), and 10 µm (for B).

17. *Teleomorph*: unknown.
Anamorph: ***Saprochaete ingens*** (van der Walt & van Kerken) de Hoog & M.Th. Smith, **comb. nov.** MycoBank MB500149. Fig. 17.

≡ *Candida ingens* van der Walt & van Kerken. Antonie van Leeuwenhoek 27: 285. 1961 (basionym).
≡ *Geotrichum ingens* (van der Walt & van Kerken) de Hoog, M.Th. Smith & Guého, Mycotaxon 63: 346. 1997.
= *Pichia humboldtii* Rodrigues de Miranda & Török, Antonie van Leeuwenhoek 42: 343. 1976.

Type: CBS 517.90 (ex-**holotype**), wine cellar, **South Africa**.

Key to species

1. a. Growth on D-xylose 2
   b. No growth on D-xylose 18

2. a. Growth at 40 °C 3
   b. No growth at 40 °C 4

3. a. Growth on cellobiose , salicin and arbutin ....***Magnusiomyces spicifer***
   b. No growth on cellobiose, salicin and arbutin .....

.....***Dipodascus australiensis***

4. a. Growth on maltose and soluble starch .....
.....***Dipodascus geniculatus***
b. No growth on maltose and soluble starch 5
5. a. Growth on cellobiose 6
b. No growth on cellobiose 8

6. a. Growth onL-sorbose 7
b. No growth on L-sorbose
.....***Dipodascus armillariae***
7. a. Growth at 30 °C.....***Geotrichum fermentans***
b. No growth at 30 °C.... ***Dipodascus macrosporus***

8. a. Growth without vitamins..... 9
b. No growth without vitamins 12

9. a. Expansion growth radius >10 mm/7 d 10
b. Expansion growth radius < 10 mm/7 d
..... ***Geotrichum klebahnii***

10.a. Growth on mannitol 11
b. No growth on mannitol .....
.....***Galactomyces pseudocandidus*** or
..... ***Geotrichum europaeum***

11.a. Growth at 35 °C.....***Galactomyces candidus***
b. No growth at 35 °C .. ***Galactomyces geotrichum***

12.a. Growth at 30 °C..... 13
b. No growth at 30 °C..... 16

13.a. Growth on D-mannitol 14
b. No growth on D-mannitol . ***Galactomyces reessii***

14.a. Growth at 35 °C.....***Saprochaete fungicola***
b. No growth at 35 °C..... 15

15 a. Expansion growth radius > 13 mm/7 d .....
.....***Galactomyces citri-auranti***
b. Expansion growth radius < 13 mm/7 d .....
..... ***Dipodascus aggregatus***

16 a. Expansion growth radius > 10 mm/7 d .....
..... ***Dipodascus albidus***
b. Expansion growth radius < 10 mm/7 d 17

17.a. Growth on DL-lactate .....
..... ***Saprochaete psychrophila***
b. No growth on DL-lactate .....
.....***Geotrichum restrictum***

18.a. Growth at 40 °C..... 19
b. No growth at 40 °C..... 22

19.a. Growth on cellobiose, salicin and arbutin .....
..... ***Saprochaete clavata***
b. No growth on cellobiose, salicin and arbutin .. 20

20.a. Growth on D-sorbitol .....
.....***Magnusiomyces tetrasperma***
b. No growth on D-sorbitol 21

21.a. Expansion growth radius > 5 mm/7 d.....
.....***Magnusiomyces capitatus***
b. Expansion growth radius < 5 mm/7 d .....
.....***Magnusiomyces starmeri***

22.a. Growth on sucrose and raffinose.....
.....***Magnusiomyces magnusii***
b. No growth on sucrose and raffinose.....23

23.a. Growth on D-glucitol and D-mannitol 24
b. No growth on D-glucitol and D-mannitol 28

24.a. Growth at 35 °C 25
b. No growth at 35 °C 27

25.a. Expansion growth radius < 10 mm/7 d.....
.....***Magnusiomyces ovetensis***
b. Expansion growth radius > 10 mm/7 d 26

26.a. Growth at 37 °C .....***Saprochaete suaveolens***
b. Growth at 37 °C.....***Saprochaete gigas***

27.a. Growth on citrate.....***Saprochaete japonica***
b. No growth on citrate.....
.....***Saprochaete suaveolens***

28.a. Growth without vitamins 29
b. No growth without vitamins 32

29.a. Growth at 35 °C 30
b. No growth at 35 °C 31

30.a. Growth on L-sorbose.....***Magnusiomyces ingens***
b. No growth on L-sorbose.....
.....***Saprochaete ingens***

31.a. Growth at 30 °C .....***Saprochaete quercus***
b. No growth at 30 °C .....
.....***Saprochaete chiloënsis***

32.a. Growth at 35 °C .....***Saprochaete suaveolens***
b. No growth at 35 °C 33

33.a. Growth on DL-lactate ***Magnusiomyces ovetensis***
b. No growth on DL-lactate.....
.....***Saprochaete saccharophila***

ACKNOWLEDGEMENTS

We are indebted to K. Ueda-Nishimura for advice concerning SSU data, and to C.P. Kurtzman, A. Aptroot, W. Gams and A.W.A.M. de Cock for useful

comments. A.H.G. Gerrits van den Ende, K.F. Luijsterburg, G. Poot and K. Luijk are thanked for technical assistance.

REFERENCES

Bakker HC den, Gravendeel B, Kuyper TW (2004). An ITS phylogeny of *Leccinium* and an analysis of the evolution of minisatellite-like sequences within ITS1. *Mycologia* **96**: 102–118.

Berbee ML, Taylor JW (1993). From 18S ribosomal sequence data to evolution of morphology among the fungi. *Canadian Journal of Botany* **73** Suppl. 1: S677–S683.

Butler EE, Fogle D, Miranda M (1988). *Galactomyces citriaurantii*, a newly found teleomorph of *Geotrichum citriaurantii*, the cause of sour rot of citrus fruit. *Mycotaxon* **33**: 197–212.

Butler EE, Petersen LJ (1972). *Endomyces geotrichum* a perfect state of *Geotrichum candidum*. *Mycologia* **64**: 365–374.

Coker WC, Shanor L (1939). A remarkable saprophytic fungoid alga. *Journal of the Elisha Mitchell Scientific Society* **55**: 152–166.

Dawes CJ (1969). *Saprochaete saccharophila*: ultrastructure, X-ray diffraction and chitin assay of cell wall as aids in evaluating taxonomic position. *Transactions of the American Microscopical Society* **88**: 572–581.

Gams W (1983). Two species of mycoparasitic fungi. *Sydowia* **36:** 46–*52*.

Hoog GS de, Gerrits van den Ende AHG (1998). Molecular diagnostics of clinical strains of filamentous Basidiomycetes. Mycoses 41: 183–189.

Hoog GS de, Guarro J, Figueras MJ, Gené J (2000). *Atlas of clinical fungi,* 2nd ed., 1126 pp. Centraalbureau voor Schimmelcultures / Universitat Rovira I Virgili, Utrecht / Reus.

Hoog GS de, Guého E, Smith MTh (1997). Nomenclatural notes on some arthroconidial yeasts. *Mycotaxon* **63**: 345–347.

Hoog GS de, Poonwan N, Gerrits van den Ende AHG (1999). Taxonomy of *Exophiala spinifera* and its relationship to *E. jeanselmei*. *Studies in Mycology* **43**: 133–142.

Hoog GS de, Smith MTh, Guého E (1986). A revision of the genus *Geotrichum* and its teleomorphs. *Studies in Mycology* **29**: 1–131.

Hoog GS de, Vicente V, Caligiorne RB, Kantarcioglu S, Tintelnot K, Gerrits van den Ende AHG, Haase G (2003). Species diversity and polymorphism in the *Exophiala spinifera* clade containing opportunistic black yeast-like fungi. *Journal of Clinical Microbiology* **41**: 4767–4778.

Kreger-van Rij NJW, Veenhuis M (1972). Some features of vegetative and sexual reproduction in *Endomyces* species. *Canadian Journal of Botany* **50**: 1691–1695.

Kreger-van Rij NJW, Veenhuis M (1973). Spores and septa in the genus *Dipodascus*. *Canadian Journal of Botany* **52**: 1335–1338.

Kurtzman CP, Fell JW (eds) (1998). *The yeasts, a taxonomic study*. 4th ed. Elsevier, Amsterdam, the Netherlands.

Kurtzman CP, Robnett CJ (1995). Molecular relationships among hyphal ascomycetous yeasts and yeastlike taxa. *Canadian Journal of Botany* **73** Suppl. 1: S824–S830.

Kurtzman CP, Smiley MJ, Johnson CJ, Wickerham LJ (1980). Two new closely related heterothallic species, *Pichia amylophila* and *Pichia mississipiensis*: characterization by hybridization and deoxyribonucleic acid reassociation. *International Journal Systematic Bacteriology* **30**: 206–216.

Lieckfeldt E, Seifert KA (2000). An evaluation of the use of ITS sequences in the taxonomy of the *Hypocreales*. *Studies in Mycology* **45**: 35–44.

Moore RT (1987). Micromorphology of yeasts and yeastlike fungi and its taxonomic implications. In: *The expanding realm of yeast-like fungi* (GS de Hoog *et al*., eds). *Studies in Mycology* **30**: 203–226.

Morenz, J. (1963). *Geotrichum candidum* Link. Taxonomie, Diagnose und medizinische Bedeutung. *Mykologische Schriftenreihe* **1**: 1–79.

Morenz, J. (1970). Geotrichosis. *Handbuch der Spezielle Pathologie und anatomische Histologie* **3**: 919-952.

Naumov GI, Smith MTh, Hoog GS de (1999). Genetic interpretation of speciation and life cycle in *Galactomyces* fungi. *Microbiology, Moscow* **68**: 362–364.

Naumova ES, Smith MTh, Boekhout T, Hoog GS de, Naumov GI (2001). Molecular differentiation of sibling species in the *Galactomyces geotrichum* complex. *Antonie van Leeuwenhoek* **80**: 263–273.

Nishida H, Sugiyama J (1994). *Archiascomycetes*: detection of a major new lineage within the *Ascomycota*. *Mycoscience* **35**: 361–366.

Peer Y Van de, Wachter R De (1993). Treecon: a software package for the construction and drawing of evolutionary trees. *Computer Applications in the Biosciences* **9**: 177–182.

Phaff HJ, Blue J, Hagler AN, Kurtzman CP (1997). *Dipodascus starmeri* sp. nov., a new species of yeast occurring in cactus necroses. *International Journal of Systematic Bacteriology* **47**: 307–312.

Platas G, Ruibal C, Collado J (2004). Size and sequence heterogeneity in the ITS1 of *Xylaria hypoxylon* isolates. *Mycological Research* **108**: 71–75.

Prillinger H, Molnár O, Eliskases-Lechner F, Lopandic K (1999). Phenotypic and genotypic identification of yeasts from cheese. *Antonie van Leeuwenhoek* **75**: 267–283.

Salkin IF, Gordon MA, Samsonoff WA, Rieder CL (1985). *Blastoschizomyces capitatus*, a new combination. *Mycotaxon* **22**: 375–380.

Seidler RJ, Mandel M (1971). Quantitative aspects of deoxyribonucleic acid renaturation: base composition, site of chromosome replication, and polynucleotide homologies. *Journal of Bacteriology* **106**: 608–614.

Smith MTh, Cock AWAM de, Poot GA, Steensma HY (1995). Genome comparisons in the yeastlike fungal genus *Galactomyces* Redhead et Malloch. *International Journal of Systematic Bacteriology* **45**: 826–831.

Smith MTh, Poot GA (1998). *Dipodascus capitatus, Dipodascus spicifer* and *Geotrichum clavatum*: genomic

characterization. *Antonie van Leeuwenhoek* **74**: 229–235.

Smith MTh, Poot GA (2003). Genome comparisons in the genus *Dipodascus* de Lagerheim. *FEMS Yeast Research* **3**: 301–311.

Smith MTh, Poot GA, Cock AWAM de (2000). Re-examination of some species of the genus *Geotrichum* Link:Fr. *Antonie van Leeuwenhoek* **77**: 71–81.

Stosch HA von (1966). Eine algologische Seltenheit, *Saprochaete saccharophila* Coker u. Shanor, in Hessen. *Hessische Floristische Briefe* **15**: 21-28.

Stosch HA von (1967). Bemerkungen zur Physiologie und Morphologie der pigmentfreien Alga *Saprochaete saccharophila* Coker und Shanor. *Le Botaniste* **50**: 437–455.

Ueda-Nishimura K, Mikata K (2000). Two distinct 18S rRNA secondary structures in *Dipodascus* (Hemiascomycetes). *Microbiology, Reading* **146**: 1045–1051.

Wagner DTS, Dawes CJ (1970). Revision of the systematic position of *Saprochaete saccharophila*. *Mycologia* **62**: 791–796.

Walt JP van der, Arx JA von (1985). Use of ultrastructural characters in yeast systematics. *South African Journal of Science* **81**: 156–159.

Wickerham LJ (1951). Taxonomy of yeasts. *Technical Bulletin 1029. US Department of Agriculture*, Washington DC.

Wilmotte AM, Peer Y van de, Goris A, Chapelle S, Baere R de, Nelissen B, Neefs JM, Hennebert GL, Wachter R de (1993). Evolutionary relationships among higher fungi inferred by small ribosomal subunit RNA sequence analysis. *Systematic and Applied Microbiology* **16**: 436–444.