International Journal of Systematic Bacteriology (1999), 49, 907–913 Printed in Great Britain

Cystofilobasidiales, a new order of basidiomycetous yeasts

Jack W. Fell,[1] Henri Roeijmans[2] and T. Boekhout[3]

Author for correspondence: Jack W. Fell. Tel: +1 305 361 4603. Fax: +1 305 361 4600.
e-mail: jfell@rsmas.miami.edu

[1] Rosenstiel School of Marine and Atmospheric Science, University of Miami, 4600 Rickenbacker Causeway, Key Biscayne, FL 33149, USA

[2] Centraalbureau voor Schimmelcultures, Oosterstraat 1, 3742 SK Baarn, The Netherlands

[3] Centraalbureau voor Schimmelcultures, Yeast Division, Julianalaan 67, 2628 BC Delft, The Netherlands

The order Cystofilobasidiales is described for teleomorphic basidiomycetous yeasts with holobasidia and teliospores. Their septa have dolipores, but lack parenthesomes. D-Glucuronate, nitrate and nitrite are assimilated and *myo*-inositol is usually assimilated. Coenzyme Q has 8 or 10 isoprenologues. 25S and 18S rDNA sequence analysis indicates a monophyletic branch within the Tremellomycetidae of the Hymenomycetes. *Cystofilobasidium* is the type genus.

Keywords: phylogeny, basidiomycetes, yeast, rDNA, Cystofilobasidiales

INTRODUCTION

The assessment of relationships among basidiomycetous yeasts has been hampered by the absence of phylogenetically informative characters; affinity among these organisms was inferred using morphological, biochemical and physiological characters (von Arx *et al*., 1977; Boekhout *et al*., 1993; van der Walt, 1987; Kurtzman & Fell, 1998). Features considered useful were cell wall composition (Prillinger *et al*., 1993; Roeijmans *et al*., 1998; Sugiyama *et al*., 1985; Weijman & Golubev, 1987; Weijman & Rodrigues de Miranda, 1988), septal ultrastructure (Boekhout *et al*., 1992; McLaughlin *et al*., 1995; Moore, 1987; Suh *et al*., 1993), morphology of the basidium (Bandoni, 1995; Boekhout *et al*., 1993; Oberwinkler, 1987), killer sensitivity patterns (Golubev & Boekhout, 1995) and specific physiological traits such as assimilation of *myo*-inositol and D-glucuronate (Golubev, 1989), production of extracellular starch-like compounds (Boekhout *et al*., 1993; Sampaio & Fonseca, 1995), assimilation of nitrate and nitrite, vitamin requirements (Sampaio & Fonseca, 1995) and ubiquinone composition (Sugiyama *et al*., 1985; Yamada *et al*., 1987). As a result, both anamorphic and teleomorphic basidiomycetous yeasts were divided into two or three main groups (Boekhout *et al*., 1993; Sampaio & Fonseca, 1995; van der Walt, 1987).

Recent molecular phylogenetic studies of basidiomycetous yeasts, using 18S rDNA sequences, indicated the presence of three classes of Basidiomycota, namely Hymenomycetes, Urediniomycetes and Ustilaginomycetes (Swann & Taylor, 1995a, b). Based on studies of 25S rDNA sequences, yeast stages were found to occur in all of these classes (Boekhout *et al*., 1995; Fell *et al*., 1995). A monophyletic cluster of species occurs within the Tremellomycetidae of the Hymenomycetes (Swann & Taylor, 1995c), that we propose to recognize as the new order Cystofilobasidiales.

METHODS

Strains. Species and strains investigated are shown in Table 1. Wherever possible, type strains were investigated. Many filamentous species, such as species of *Tremella* and *Sirobasidium*, do not have designated living type strains.

Carbohydrate patterns and coenzyme Q measurement. Carbohydrate patterns were determined by analysis of whole-cell hydrolysates with GLC/MS (Roeijmans *et al*., 1998). Coenzyme Q (CoQ) was extracted with hexane after saponification of intact cells and purification by TLC. Coenzyme Q systems were identified by reverse-phase TLC (Yamada & Kondo, 1973; Hiraishi *et al*., 1984) and by MS. In the latter technique, 3 µl crude extract was inserted in the quartz sample tube of the direct inlet probe and probe temperature was programmed from 50 to 400 °C. The CoQ components evaporated when the probe temperature exceeded 300 °C. Mass spectra were recorded with an ionizing energy of 70 eV (electron impact) and the CoQ systems were identified by their molecular mass.

Sequencing and sequence analysis. Forward and reverse strands of the ~ 650 bp region at the 5′ end of the lrDNA were sequenced with an Li-Cor Automated Sequencer using

---

The GenBank accession numbers for the sequences reported in this paper are AF075463–AF075526.

Table 1. List of strains examined

Species	CBS strain no.*	GenBank accession no.
Bullera crocea Buhagiar	6714^{T}	AF075508
Bullera globispora Johri & Bandoni	6981^{T}	AF075509
Bullera oryzae Nakase & Suzuki	7194^{T}	AF075511
Bullera pseudoalba Nakase & Suzuki	7227^{T}	AF075504
Bullera unica Hamamoto & Nakase	8290^{T}	AF075524
Bulleromyces albus Boekhout & Fonseca	501	AF075500
Cryptococcus aerius (Saito) Skinner	155^{T}	AF075486
Cryptococcus albidus (Saito) Skinner	142^{T}	AF075474
Cryptococcus antarcticus Vishniac & Kurtzman	7687^{T}	AF075488
Cryptococcus aquaticus (Jones & Slooff) Rodrigues de Miranda & Weijman	5443^{T}	AF075470
Cryptococcus cellulolyticus Nakase *et al.*	8294^{T}	AF075525
Cryptococcus diffluens Zach (Lodder & Kreger-van Rij)	160^{T}	AF075502
Cryptococcus dimennae Fell & Phaff	5770^{T}	AF075489
Cryptococcus feraegula Saëz & Rodrigues de Miranda	7201	AF075487
Cryptococcus flavus (Saito) Phaff & Fell	331^{T}	AF075497
Cryptococcus friedmannii Vishniac	7160^{T}	AF075478
Cryptococcus fuscescens Golubev	7189^{T}	AF075472
Cryptococcus heveanensis (Groenewege) Baptist & Kurtzman	569^{T}	AF075467
Cryptococcus hungaricus (Zsolt) Phaff & Fell	4214^{T}	AF075503
Cryptococcus laurentii (Kufferath) Skinner	139^{T}	AF075469
Cryptococcus luteolus Skinner	943^{T}	AF075482
Cryptococcus macerans (Frederiksen) Phaff & Fell	2425^{MT}	AF075477
Cryptococcus podzolicus (Babjeva & Reshetova) Golubev	6819^{T}	AF075481
Cryptococcus skinneri Phaff & do Carmo-Sousa	7890†	AF075494
Cryptococcus terreus di Menna	1895^{T}	AF075479
Cryptococcus vishniacii Vishniac & Hempfling	7110^{T}	AF075473
Cystofilobasidium bisporidii (Fell *et al.*) Oberwinkler & Bandoni	6347^{MT}	AF075464
Cystofilobasidium capitatum (Fell *et al.*) Oberwinkler & Bandoni	6358^{T}	AF075465
Cystofilobasidium infirmominiatum (Fell *et al.*) Hamamoto *et al.*	323^{T}	AF075505
Cystofilobasidium lari-marini (Saëz & Nguyen) Fell & Statzell-Tallman	7420^{T}	AF075466
Fellomyces fuzhouensis (Yue) Yamada & Banno	6133	AF075506
Filobasidiella neoformans var. *bacillispora* Kwon-Chung	6289^{MT}	AF075526
Filobasidiella neoformans var. *neoformans* Kwon-Chung	132^{T}	AF075484
Filobasidium capsuligenum Rodrigues de Miranda	4736^{T}	AF075501
Filobasidium floriforme Olive	6241^{T}	AF075498
Filobasidium globisporum Bandoni & Oberwinkler	7642	AF075495
Filobasidium uniguttulatum Kwon-Chung	1730^{T}	AF075468
Kockovaella thailandica Nakase *et al.*	7552^{T}	AF075516
Mrakia frigida (Fell *et al.*) Yamada & Komagata	5270^{T}	AF075463
Rhodotorula diffluens (Ruinen) von Arx & Weijman	5233^{T}	AF075485
Rhodotorula foliorum (Ruinen) Rodrigues de Miranda & Weijman	6370	AF075499
Rhodotorula philyla (van der Walt *et al.*) Rodrigues de Miranda & Weijman	6272^{T}	AF075471
Sirobasidium intermedium Kundalkar & Patil (yeast phase)	7805	AF075492
Sirobasidium magnum Boedijn	6803	AF075475
Sporobolomyces falcatus Santa Marìa	7368^{T}	AF075490
Sterigmatosporidium polymorphum Kraepelin & Schulze	8088^{T}	AF075480
Tremella fuciformis Berkeley (yeast state)	6970	AF075476
Tremella mesenterica Retzius: Fries (yeast state)	6973	AF075518
Tremella moriformis Smith & Sowerby (yeast state)	7810^{T}	AF075493
Trichosporon aquatile Hedrick & Dupont	5973^{T}	AF075520
Trichosporon asteroides (Rischin) Ota	2481^{T}	AF075513
Trichosporon brassicae Nakase	6382^{T}	AF075521
Trichosporon cutaneum (de Beurmann *et al.*) Ota	2466^{T}	AF075483
Trichosporon domesticum Sugita *et al.*	8280^{T}	AF075512
Trichosporon dulcitum (Berhout) Weijman	8257^{T}	AF075517
Trichosporon laibachii (Windisch) Guého & Smith	5790^{T}	AF075514
Trichosporon loubieri (Morenz) Weijman	7065^{T}	AF075522
Trichosporon mucoides Guého & Smith	7625^{T}	AF075515
Trichosporon ovoides Behrend	7556^{T}	AF075523
Trimorphomyces papilionaceus Oberwinkler & Bandoni	445.92	AF075491
Udeniomyces megalosporus (Nakase & Suzuki) Nakase & Takematsu	7236^{T}	AF075510
Udeniomyces puniceus (Komagata & Nakase) Nakase & Takematsu	5689^{T}	AF075519
Udeniomyces pyricola (Stadelmann) Nakase & Takematsu	6754^{T}	AF075507
Xanthophyllomyces dendrorhous Golubev	7918^{T}	AF075496

* CBS, Centraalbureau voor Schimmelcultures, The Netherlands. ^{T}, Type strain; ^{MT}, mating type.

† Note added in proof. CBS 7890 was originally identified as *Cryptococcus skinneri* in the CBS database. Subsequent examination of CBS 5029^{T}, the type strain of *C. skinneri*, demonstrated that the two strains are phylogenetically distinct. CBS 5029^{T} occurs in the same clade as *C. luteolus* in Fig. 1. CBS 7890 represents an undescribed species.

a standard protocol with forward primer 5′ GCATATCAATAAGCGGAGGAAAAG and reverse primer 5′ GGTCCGTGTTTCAAGACG. Alignments were prepared with MegAlign (DNAStar) and visually corrected. Phylogenetic trees were computed with PAUP (test version 4.0d61, used with the permission of D. Swofford) using parsimony analysis (heuristic search, random stepwise addition, nearest-neighbour interchange, 100 maximum trees). Bootstrap analysis was based on 1000 replicates.

RESULTS AND DISCUSSION

Yeasts classified in the Tremellomycetidae (Hymenomycetes) are characterized by the presence of dolipore septa, xylose in the cell walls and, with some exceptions, utilization of *myo*-inositol and D-glucuronate, and production of starch-like compounds. Fig. 1 is a representative analysis of the Tremellomycetidae; the number of species in this figure was limited to increase the visual clarity of the tree. There are two major branches, one of which contains genera of the Tremellales/Filobasidiales and the other branch consists of the teleomorphic genera *Mrakia* and *Cystofilobasidium*, the anamorphic genus *Udeniomyces* and two species of *Cryptococcus*. The Tremellales and Filobasidiales are characterized by a sexual cycle that lacks a teliosporic probasidium and by the morphology of the septal pores. Species of *Bulleromyces*, *Sirobasidium*, *Sterigmatosporidium*, *Tremella* and relatives, and *Filobasidium* have Tremellales-type dolipores with tubular, vesiculate or 'U'-shaped parenthesomes (Boekhout *et al.*, 1993; Kwon-Chung *et al.*, 1995; McLaughlin *et al.*, 1995; Moore & Kreger-van Rij, 1972; Oberwinkler & Bandoni, 1982). The reported absence of a parenthesome in *Filobasidium floriforme* can be questioned, as a Tremellales-type parenthesome appears to be present in Fig. 4 of Moore & Kreger-van Rij (1972). Parenthesomes are generally considered as modified compartments of the endoplasmic reticulum (Moore, 1985), which may play an important role in intercellular signalling and maintenance of homeostasis (Mueller *et al.*, 1998).

There are two sub-branches within the Tremellales/Filobasidiales: one sub-branch, which is weakly supported (58%) by bootstrap analysis, consists of species of *Tremella*, *Bulleromyces*, *Filobasidiella*, etc. The anamorphic genus *Trichosporon*, which is included in this cluster, is supported (bootstrap value 81%) as a cohesive group. This genus, which is characterized by the presence of arthroconidia, was studied in detail by Guého *et al.* (1992). The other sub-branch, which has a bootstrap support of 90%, includes *Filobasidium* and several species of *Cryptococcus*. These data suggest that the Filobasidiales are phylogenetically distinct from the Tremellales. The sequence analysis also demonstrates a separate Cystofilobasidiales clade within the Tremellomycetidae of the Hymenomycetes, with a bootstrap value of 87% (Fig. 1). Two subclades can be discerned: (1) *Mrakia frigida*, *Cryptococcus aquaticus* and *Udeniomyces* spp. (90% support); and (2) *Cryptococcus feraegula*, *Cryptococcus macerans* and *Cystofilobasidium* spp. (100%). These results agree with 18S rDNA data, which support the *Cystofilobasidium–Mrakia* clade with 98% bootstrap values (Suh & Nakase, 1995; Swann & Taylor, 1995a, b). The two teleomorphic genera in these clades, *Mrakia* and *Cystofilobasidium*, produce teliospores, a characteristic not found among other hymenomycetous yeasts. Metabasidia differ between the two genera: teliospore germination in *Mrakia* results in short (~ 8 × 5 μm) holo- or phragmometabasidia; whereas, most species of *Cystofilobasidium* produce a long, thin (up to 80 × 2·7 μm) holobasidium with a capitate apex (Kwon-Chung, 1998).

The distribution of other character states among the species of Cystofilobasidiales is given in Table 2. Two different CoQ systems occur: CoQ 8 occurs in all species except for *Udeniomyces* spp., which contain CoQ 10 (Boekhout *et al.*, 1992; Guého *et al.*, 1993; Nakase & Suzuki, 1986; Nakase & Takematsu, 1992; Suh & Sugiyama, 1993; Sugiyama *et al.*, 1985; Yamada & Komagata, 1987; Yamada & Kondo, 1972, 1973; Yamada & Matsumoto, 1989). The different genes involved in the biosynthesis of distinct CoQ molecules (CoQ 7, 8, 9 or 10) may be present in a single organism. This possibility is suggested by the report of the occurrence of minor CoQ fractions in concert with one or two major CoQ fractions (Billon-Grand, 1988; Kuraishi *et al.*, 1985) and by the observation that the relative proportions of these different CoQ fractions are influenced by environmental conditions (Billon-Grand, 1989). If this supposition is true, the presence of two different major CoQ molecules among the Cystofilobasidiales may be the result of differences in gene expression with limited phylogenetic significance. It should be noted, however, that CoQ 10 is confined to the ballistoconidia-forming species and CoQ 8 to the teliospore-forming yeasts and closely related anamorphs.

All species of Cystofilobasidiales contain xylose in their cell walls (von Arx & Weijman, 1979; Boekhout *et al.*, 1992; Elinov *et al.*, 1974; Golubev, 1995; Gorin & Spencer, 1970; Roeijmans *et al.*, 1989; Sugiyama *et al.*, 1985; Suh & Sugiyama, 1993; Weijman, 1979; Weijman & Rodrigues de Miranda, 1988). D-Glucuronate, nitrate and nitrite are assimilated by all species, *myo*-inositol is used by most species, except *Cryptococcus aquaticus*, most species are dependent on thiamine and biotin, and fermentative capabilities occur in *Cystofilobasidium lari-marini* (Barnett *et al.*, 1990; Kwon-Chung, 1998). Hyphal septa of investigated species of the Cystofilobasidiales, namely *Mrakia frigida* and *Cystofilobasidium* spp., have dolipores without parenthesomes (Oberwinkler *et al.*, 1983; Suh & Sugiyama, 1993; Suh *et al.*, 1993). The argument can be presented that dolipores without parenthesomes are plesiomorphic when compared with dolipores with vesiculate, tubular or 'U'-shaped,

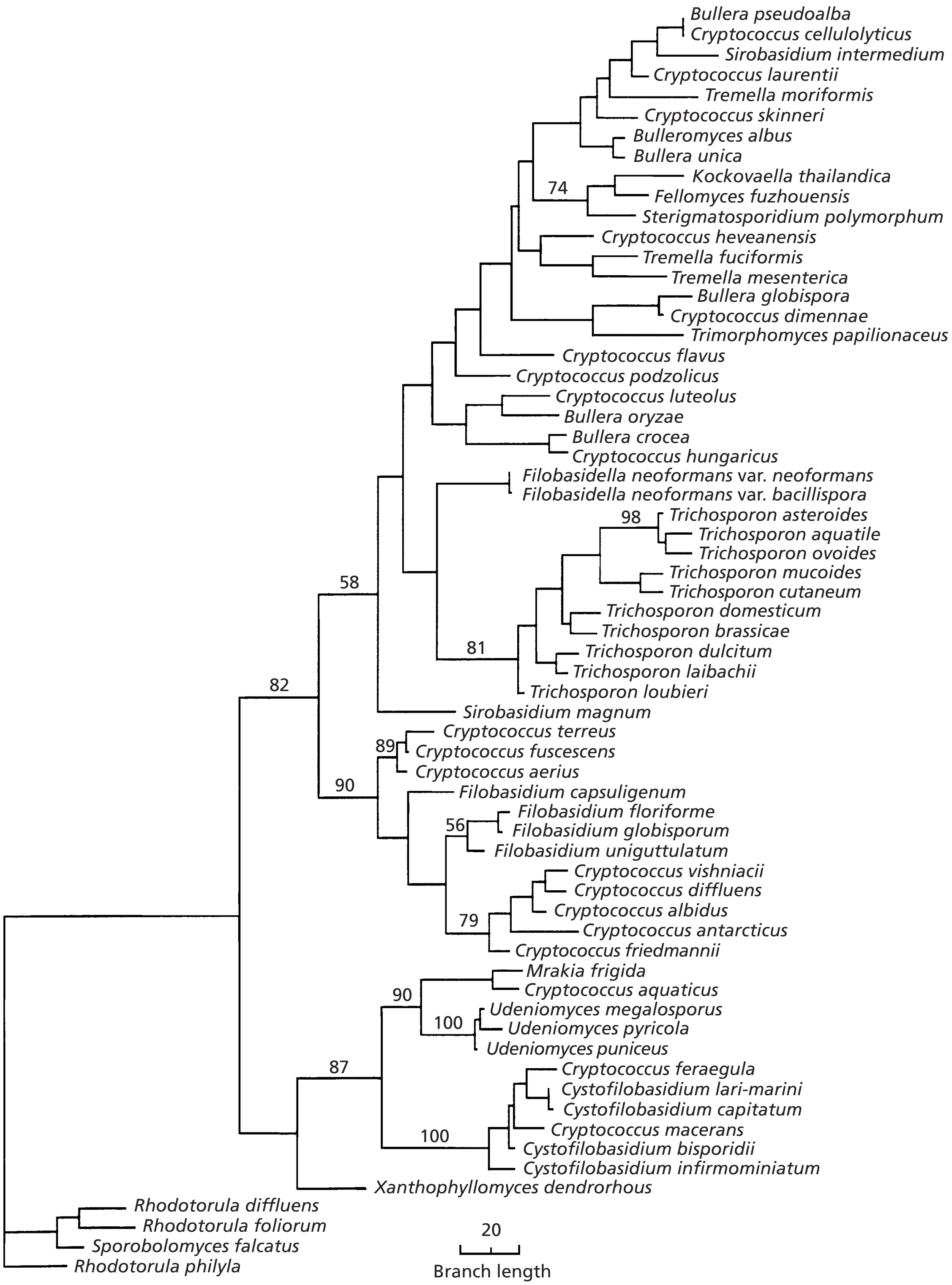

Fig. 1. Phylogenetic tree of yeasts of the Tremellomycetidae derived from parsimony analysis (heuristic search, random stepwise addition, nearest-neighbour interchange, 100 maximum trees). Branch lengths are proportional to the number of nucleotide differences. The numbers given on branches are the frequencies (percentages) that a given branch appeared in 1000 bootstrap replicates (PAUP test version 4.0d61, used with the permission of D. Swofford). Species of Urediniomycetes (*Rhodotorula* and *Sporobolomyces*) were used as an outgroup.

Table 2. Summary of salient characteristics of Cystofilobasidiales species

CoQ, Coenzyme Q; Xylose, presence of xylose in cell-free extract; Telio, formation of teliospore; Basidia, absence of basidium (−) or presence of holometabasidium (HOLO); Septa, septal type unknown (?) or presence of dolipore (DOLI); Inositol, assimilation of *myo*-inositol; D-Gluc, assimilation of D-glucuronate; Starch, production of starch-like compounds; NO_3 and NO_2, assimilation of nitrate and nitrite; w/o Thiamine/Biotin, growth in the absence of either of those vitamin sources. V, Variable reaction; W, weak reaction, D, delayed; *, new data. Other data from Barnett *et al.* (1990) and Kurtzman & Fell (1998).

Species	CoQ	Xylose	Telio	Basidia	Septa	Inositol	D-Gluc	Starch	NO_3	NO_2	w/o Thiamine	w/o Biotin
Cryptococcus aquaticus	8*	+	−	−	?	−	+	+	+	+	−	−
Cryptococcus feraegula	8*	+*	−	−	?	+	+	+	+	+	−	−
Cryptococcus macerans	8*	+	−	−	?	+	+	+	+	+	−	−
Cystofilobasidium bisporidii	8	+	+	HOLO	DOLI	+	+	V	+	+	−	−
Cystofilobasidium capitatum	8	+	+	HOLO	DOLI	+	+	+	+	+	−	V
Cystofilobasidium infirmominiatum	8	+	+	HOLO	DOLI	+	+	+	+	+	−	−
Cystofilobasidium lari-marini	8	+	+	HOLO	DOLI	V	+	+	+	+	−	−,D
Mrakia frigida	8	+	+	HOLO	DOLI	V	+	+	+	+	−	−,D
Udeniomyces megalosporus	10	+	−	−	?	−	+	+	+	+	−	−
Udeniomyces pyricola	10	+	−	−	?	+	+	W	+	+	−	−
Udeniomyces puniceus	10	+	−	−	?	+	+	+	+	+	−	−

plate-like or perforate parenthesomes. However, detailed studies on the biogenesis, differentiation and function of the septum–parenthesome complex in different groups of hymenomycetous fungi, e.g. Auriculariales, Cystofilobasidiales, Filobasidiales, Exidiales, Tremellales and higher fungi (e.g. Agaricales), are necessary to understand the evolutionary importance of these structures. Preparation techniques, such as high-pressure freezing, freeze substitution and cryo-electron microscopy, are essential to avoid artifacts (Mueller *et al.*, 1995; Orlovich & Ashford, 1994) which may hamper correct interpretations. This requirement is illustrated by conflicting results in different studies on the septal ultrastructure of *Filobasidiella depauperata* (Kwon-Chung *et al.*, 1995; Rhodes *et al.*, 1981).

Our analysis of rDNA sequences demonstrates that three species of *Cryptococcus* are members of the Cystofilobasidiales (Fig. 1). Based on this apparent relationship, the hypothesis can be offered that *Cryptococcus macerans* and *Cryptococcus feraegula* may have a *Cystofilobasidium*-type life cycle, whereas the life cycle of *Cryptococcus aquaticus* may be similar to *Mrakia*. *Xanthophyllomyces dendrorhous* (*Phaffia rhodozyma*) may be related to Cystofilobasidiales as indicated by 18S data (Swann & Taylor, 1995c); however, inclusion in this clade is not supported by bootstrap analysis (Fig. 1) using 25S rDNA data. Parsimony analysis indicated that two other species, *Trichosporon pullulans* and *Itersonila perplexans*, may be weakly related to the Cystofilobasidiales (Fell *et al.*, 1995), but bootstrap analysis did not support their inclusion. Both species differ from the Cystofilobasidiales by the presence of CoQ 9. As more data accumulates regarding peripherally related species, our vision of Cystofilobasidiales may alter. Similarly, the present definitions of the Tremellales and Filobasidiales must be evaluated to consider such problems as the phylogenetic placement of *Filobasidiella*.

Latin diagnosis of Cystofilobasidiales Boekhout & Fell

Cystofilobasidiales, order novus Basidiomycetum, species plerumque zymosae; taxa meiosporica, heterothallica vel ipsifertilia. Teliosporae vulgo formatae; basidia si praesentia, unicellularia. Coenzymum Q 8 vel 10. Parietes cellularum xylosium continentes. Septa hypharum plerumque doliporis parenthesomate carentibus praedita, sed nonnumquam imperforata. D-*Glucuronatum, nitratum et nitritum assimilantur. Sequentiae rDNA positionem in hymenomycetibus indicantes. Genus typicum Cystofilobasidium* Oberwinkler & Bandoni.

Description of Cystofilobasidiales Boekhout & Fell

Basidiomycetous fungi, usually with a free-living yeast phase. Meiotic taxa are heterothallic or self-sporulating (secondarily homothallic?). Teliospores are present. Basidia, if present, are unicellular. CoQ 8 or 10 is present. Cell walls contain xylose. Hyphal septa usually have dolipores without a parenthesome. D-Glucuronate, nitrate and nitrite are assimilated and *myo*-inositol is usually assimilated. 25S rDNA sequences form a monophyletic branch within the Tremellomycetidae. Type genus is *Cystofilobasidium* Oberwinkler & Bandoni.

The following meiotic taxa are included in the Cystofilobasidiales: *Cystofilobasidium*, including *Cystofilobasidium bisporidii* (Fell *et al.*) Oberwinkler & Bandoni, *Cystofilobasidium capitatum* (Fell *et al.*) Oberwinkler & Bandoni, *Cystofilobasidium infirmominiatum* (Fell *et al.*) Hamamoto *et al.*, *Cystofilobasidium lari-marini* (Saëz & Nguyen) Fell & Statzell-Tallman; *Mrakia*, including *Mrakia frigida* (Fell *et al.*) Yamada & Komagata. The following taxa are only known in their mitotic state, but 25S rDNA sequences indicate that they belong to this order: *Cryptococcus*, including *Cryptococcus aquaticus* (Jones

& Slooff) Rodrigues de Miranda & Weijman (*Mrakia* branch), *Cryptococcus feraegula* Saëz & Rodrigues de Miranda, *Cryptococcus macerans* (Frederiksen) Phaff & Fell (*Cystofilobasidium* branch); *Udeniomyces*, including *Udeniomyces megalosporus* (Nakase & Suzuki) Nakase & Takematsu, *Udeniomyces puniceus* (Komagata & Nakase) Nakase & Takematsu, *Udeniomyces pyricola* (Stadelmann) Nakase & Takematsu (*Udeniomyces* branch).

ACKNOWLEDGEMENTS

Funding to J.W.F. was provided by the National Science Foundation (Biological Oceanography and Systematic Biology). Gina Blatt, Gloria Scorzetti and Adele Statzell-Tallman (University of Miami) provided invaluable assistance in developing and preparing the sequence data.

REFERENCES

von Arx, J. A. & Weijman, A. C. M. (1979). Conidiation and carbohydrate composition in some *Candida* and *Torulopsis* species. *Antonie Leeuwenhoek* **45**, 547–555.

von Arx, J. A., Rodrigues de Miranda, L., Smith, M. T. & Yarrow, D. (1977). The genera of the yeasts and the yeast-like fungi. *Stud Mycol* **14**, 1–42.

Bandoni, R. J. (1995). Dimorphic heterobasidiomycetes: taxonomy and parasitism. *Stud Mycol* **38**, 13–27.

Barnett, J. A., Payne, R. W. & Yarrow, D. (1990). *Yeasts: Characteristics and Identification*, 2nd edn. Cambridge: Cambridge University Press.

Billon-Grand, G. (1988). *Contribution la taxonomie et la phylogenie des levures. Apport du critère 'Coenzyme Q'*. PhD thesis, Université Claude Bernard, Lyon, France.

Billon-Grand, G. (1989). Influence on minor peaks of coenzyme Q of the glucose concentration in the culture medium, the stage of the growth cycle, and the duration of the coenzyme Q extraction: required conditions for determining the minor coenzymes Q. *J Gen Appl Microbiol* **35**, 261–268.

Boekhout, T., Yamada, Y., Weijman, A. C. M., Roeijmans, H. J. & Batenburg-van der Vegte, W. H. (1992). The significance of coenzyme Q, carbohydrate composition and septal ultrastructure for the taxonomy of ballistoconidia-forming yeasts and fungi. *Syst Appl Microbiol* **15**, 1–10.

Boekhout, T., Fonseca, A., Sampaio, J.-P. & Golubev, W. I. (1993). Classification of heterobasidiomycetous yeasts: characteristics and affiliation of genera to higher taxa. *Can J Microbiol* **39**, 276–290.

Boekhout, T., Fell, J. W. & O'Donnell, K. (1995). Molecular systematics of some yeast-like anamorphs belonging to the Ustilaginales and Tilletiales. *Stud Mycol* **38**, 175–183.

Elinov, N. P., Vitovskaya, G. A., Kaloshin, V. G. & Kolotinskaya, T. M. (1974). Structural polysaccharides of some species of *Rhodotorula* and *Cryptococcus*. *Biokhimiya* **39**, 787–792.

Fell, J. W., Boekhout, T. & Freshwater, D. W. (1995). The role of nucleotide sequence analysis in the systematics of the yeast genera *Cryptococcus* and *Rhodotorula*. *Stud Mycol* **38**, 129–146.

Golubev, W. I. (1989). Catabolism of i-inositol and taxonomic value of D-glucuronate assimilation in yeasts. *Mikrobiologiya* **58**, 276–283.

Golubev, W. I. (1995). Perfect state of *Rhodomyces dendrorhous* (*Phaffia rhodozyma*). *Yeast* **11**, 101–110.

Golubev, W. I. & Boekhout, T. (1995). Sensitivity to killer toxins as a taxonomic tool among heterobasidiomycetous yeasts. *Stud Mycol* **38**, 47–58.

Gorin, P. A. J. & Spencer, J. F. T. (1970). Proton magnetic resonance spectroscopy as an aid in identification and chemotaxonomy of yeasts. *Adv Appl Microbiol* **13**, 25–89.

Guého, E., Smith, M. Th., de Hoog, G. S., Billon-Grand, G., Christen, R. & Batenburg-van der Vegte, W. H. (1992). Contributions to a revision of the genus *Trichosporon*. *Antonie Leeuwenhoek* **61**, 289–316.

Guého, E., Improvisi, L., Christen, R. & de Hoog, G. S. (1993). Phylogenetic relationships of *Cryptococcus neoformans* and some related basidiomycetous yeasts determined from partial large subunit rRNA sequences. *Antonie Leeuwenhoek* **63**, 175–189.

Hiraishi, A., Hoshino, Y. & Kitamura, H. (1984). Isoprenoid quinone composition in the classification of Rhodospirillaceae. *J Gen Appl Microbiol* **30**, 197–210.

Kuraishi, H., Katayama-Fujimura, Y., Sugiyama, J. & Yokoyama, T. (1985). Ubiquinone systems in fungi I. Distribution of ubiquinones in the major families of ascomycetes, basidiomycetes, and deuteromycetes, and their taxonomic implications. *Trans Mycol Soc Jpn* **26**, 383–395.

Kurtzman, C. P. & Fell, J. W. (1998). *The Yeasts: a Taxonomic Study*, 4th edn. Amsterdam: Elsevier.

Kwon-Chung, K. J. (1998). *Cystofilobasidium* Oberwinkler, Bandoni, Blanz, Kishimova-Horovitz. In *The Yeasts: a Taxonomic Study*, 4th edn, pp. 650–657. Edited by C. P. Kurtzman & J. W. Fell. Amsterdam: Elsevier.

Kwon-Chung, K. J., Chang, Y. C., Bauer, R., Swann, E. C., Taylor, J. W. & Goel, R. (1995). The characteristics that differentiate *Filobasidiella depauperata* from *Filobasidiella neoformans*. *Stud Mycol* **38**, 67–80.

McLaughlin, D. J., Frieders, M. E. & Lue, H. (1995). A microscopist's view of heterobasidiomycete phylogeny. *Stud Mycol* **38**, 91–109.

Moore, R. T. (1985). The challenge of the dolipore/parenthesome septum. In *Developmental Biology of Higher Fungi*, pp. 175–212. Edited by D. Moore, L. A. Casselton, D. A. Wood & J. A. Frankland. Cambridge: Cambridge University Press.

Moore, R. T. (1987). Micromorphology of yeasts and yeast-like fungi and its taxonomic implications. *Stud Mycol* **30**, 203–226.

Moore, R. T. & Kreger-van Rij, N. J. W. (1972). Ultrastructure of *Filobasidium*. *Can J Microbiol* **18**, 1949–1951.

Mueller, W. H., van Aelst, A. V., van der Krift, T. & Boekhout, T. (1995). Novel approaches to visualize the septal pore cap. *Stud Mycol* **38**, 111–117.

Mueller, W. H., Humbel, B. M., van Aelst, A. C., van der Krift, T. & Boekhout, T. (1998). Intercellular exchange through perforate septal pore caps in homobasidiomycetes. In *Plasmodesmata: Nanochannels with Megatasks*. Edited by A. van Bel. Berlin: Springer (in press).

Nakase, T. & Suzuki, M. (1986). The ubiquinone system in strains of species in the ballistospore-forming yeast genera *Sporidiobolus*, *Sporobolomyces* and *Bullera*. *J Gen Appl Microbiol* **32**, 251–258.

Nakase, T. & Takematsu, A. (1992). *Udeniomyces*, a new ballistosporous anamorphic yeast genus in the Cryptococcaceae proposed for three *Bullera* species which produce large bilaterally symmetrical ballistospores. *FEMS Microbiol Lett* **100**, 497–502.

Oberwinkler, F. (1987). Heterobasidiomycetes with ontogenetic yeast-stages. Systematic and phylogenetic aspects. *Stud Mycol* **30**, 61–74.

Oberwinkler, F. & Bandoni, R. J. (1982). *Trimorphomyces*: a new genus in the Tremellaceae. *Syst Appl Microbiol* **4**, 105–113.

Oberwinkler, F., Bandoni, R. J. & Kisimova-Horovitz, L. (1983). *Cystofilobasidium*: a new genus in the Filobasidiaceae. *Syst Appl Microbiol* **4**, 114–122.

Orlovich, D. A. & Ashford, A. E. (1994). Structure and development of the dolipore septum in *Pisolithus tinctorius*. *Protoplasma* **178**, 66–80.

Prillinger, H., Oberwinkler, F., Umile, C., Tlachac, K., Bauer, R., Dörfler, C. & Taufratzhofer, E. (1993). Analysis of cell wall carbohydrates (neutral sugars) from ascomycetous and basidiomycetous yeasts with and without derivatization. *J Gen Appl Microbiol* **39**, 1–34.

Rhodes, J. C., Kwon-Chung, K. J. & Popkin, T. J. (1981). Ultrastructure of the septal complex in hyphae of *Cryptococcus laurentii*. *J Bacteriol* **145**, 1410–1412.

Roeijmans, H. J., van Eijk, G. W. & Yarrow, D. (1989). Some name changes necessitated by the redefinition of the genus *Candida*. *Mycotaxon* **35**, 405–406.

Roeijmans, H., Prillinger, H., Umile, C., Sugiyama, J., Nakase, T. & Boekhout, T. (1998). Analysis of carbohydrate composition of cell walls and extracellular carbohydrates. In *The Yeasts: a Taxonomic Study*, 4th edn, pp. 103–105. Edited by C. P. Kurtzman & J. W. Fell. Amsterdam: Elsevier.

Sampaio, J. P. & Fonseca, A. (1995). Physiological aspects in the systematics of heterobasidiomycetous yeasts. *Stud Mycol* **38**, 29–46.

Sugiyama, J., Fukagawa, M., Chiu, S. W. & Komagata, K. (1985). Cellular carbohydrate composition, DNA base composition, ubiquinone systems, and diazonium blue B colour test in the genera *Rhodosporidium*, *Leucosporidium*, *Rhodotorula* and related basidiomycetous yeasts. *J Gen Appl Microbiol* **31**, 519–550.

Suh, S. O. & Nakase, T. (1995). Phylogenetic analysis of the ballistosporous anamorphic genera *Udeniomyces* and *Bullera*, and related basidiomycetous yeasts, based on 18S rDNA sequence. *Microbiology* **141**, 901–906.

Suh, S. O. & Sugiyama, J. (1993). Septal pore ultrastructure of *Leucosporidium lari-marini*, a basidiomycetous yeast, and its taxonomic implications. *J Gen Appl Microbiol* **39**, 257–260.

Suh, S. O., Hirata, A., Sugiyama, J. & Komagata, K. (1993). Septal ultrastructure of basidiomycetous yeasts and their taxonomic implications with observations on the ultrastructure of *Erythrobasidium hasegawianum* and *Sympodiomycopsis paphiopedili*. *Mycologia* **85**, 30–37.

Swann, E. C. & Taylor, J. W. (1995a). Phylogenetic perspectives on basidiomycete systematics: evidence from the 18S rRNA gene. *Can J Bot* **73**, S862–S868.

Swann, E. C. & Taylor, J. W. (1995b). Phylogenetic diversity of yeast-producing basidiomycetes. *Mycol Res* **99**, 1205–1210.

Swann, E. C. & Taylor, J. W. (1995c). Toward a phylogenetic systematics of the Basidiomycota: integrating yeasts and filamentous basidiomycetes using 18S rRNA gene sequences. *Stud Mycol* **38**, 147–161.

van der Walt, J. P. (1987). The yeasts – a conspectus. *Stud Mycol* **30**, 19–31.

Weijman, A. C. M. (1979). Carbohydrate composition of *Geotrichum*, *Trichosporon* and allied genera. *Antonie Leeuwenhoek* **45**, 119–127.

Weijman, A. C. M. & Golubev, W. I. (1987). Carbohydrate patterns and taxonomy of yeasts and yeast-like fungi. *Stud Mycol* **30**, 361–371.

Weijman, A. C. M. & Rodrigues de Miranda, L. (1988). Carbohydrate patterns of the genera *Candida*, *Rhodotorula*, and *Cryptococcus*. *Antonie Leeuwenhoek* **54**, 535–543.

Yamada, Y. & Komagata, K. (1987). *Mrakia* gen. nov., a heterobasidiomycetous yeast genus for the Q8-equipped, self-sporulating organism which produces a unicellular metabasidium, formerly classified in the genus *Leucosporidium*. *J Gen Appl Microbiol* **33**, 455–457.

Yamada, Y. & Kondo, K. (1972). Taxonomic significance of the coenzyme Q system in yeasts and yeast-like fungi. Proceedings of the 4th International Fermentation Symposium, Kyoto, Japan. *Ferment Technol Today* 781–784.

Yamada, Y. & Kondo, K. (1973). Coenzyme Q system in the classification of the yeast genera *Rhodotorula* and *Cryptococcus*, and the yeast-like genera *Sporobolomyces* and *Rhodosporidium*. *J Gen Appl Microbiol* **19**, 59–77.

Yamada, Y. & Matsumoto, A. (1989). An electrophoretic comparison of enzymes in strains of species in the genus *Mrakia* Yamada et Komagata (Filobasidiaceae). *J Gen Appl Microbiol* **34**, 201–208.

Yamada, Y., Banno, I., von Arx, J. A. & van der Walt, J. P. (1987). Taxonomic significance of the coenzyme Q system in yeasts and yeast-like fungi. *Stud Mycol* **30**, 299–308.