Fungal Taxonomy: New Developments in Medically Important Fungi

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Our understanding of the causative agents of fungal diseases has changed considerably in recent years due to molecular studies that compare DNA across a wide range of fungi, including human and animal pathogens. In many cases, what had once been understood as traditional species were found to be species complexes. Importantly, members of such complexes may differ in pathogenicity and susceptibility to antifungals, which suggests a need for accurate identification to provide optimal patient care. This article presents a few striking examples from Zygomycetes, Ascomycetes, and Basidiomycetes.

Introduction

Taxonomic concepts of clinically relevant fungi have changed considerably in the past decade (Fig. 1). This is mainly due to the large-scale introduction of molecular studies that compare one or more stretches of DNA across a wide range of fungi, including human and animal pathogens [1•]. As a result, a novel higher-level taxonomy has been proposed for the fungal kingdom [2]. One of the most striking discoveries was the recognition that *Pneumocystis* is a fungus, as this organism was previously considered a parasite. Importantly, the number of human pathogenic species have increased considerably in almost all studied genera. In several cases, significant conceptual changes have resulted. For instance, in the case of *Malassezia*, *M. restricta* and *M. globosa* are involved in many skin disorders, and not necessarily only *M. furfur*, which had been believed to be the primary pathogen. A further consequence of these developments in modern medical mycology is that many newly recognized, clinically relevant species may differ in susceptibility to commonly used antifungals. Therefore, correct identification of the new pathogens is highly important for proper patient management. Unfortunately, most clinical microbiology laboratories do not have the capacity or knowledge to identify new species recognized in these so-called species complexes. This need can, however, be fulfilled by research and/or reference laboratories in many countries. This article discusses some of these taxonomy-related trends and their consequences for clinical practice, such as therapeutic choices.

Zygomycetes

Recent phylogenetic studies based on data from several loci favor a polyphyletic origin of the Zygomycota (Fig. 1) [1•,2]. Two subphyla of Zygomycota, namely Mucoromycotina and Entomophthoromycotina, contain agents of human zygomycosis. The Entomophthoromycotina, with the single order Entomophthorales, include the clinically relevant *Conidiobolus*, whereas the clinically important genus *Basidiobolus*, previously listed in the same order, was excluded and is now assigned to the order Basidiobolus. However, Basidiobolus is not yet classified as a higher taxon because of its unresolved position close to the flagellated genus *Olpidium* [1•,2].

The following genera or species have been associated with infections in humans: *Rhizopus*, *Mucor*, *Rhizomucor*, *Cokeromyces recurvatus*, *Cunninghamella bertholletia*, *Apophysomyces elegans*, *Saksenaea vasiformis*, *Syncephalastrum racemosum*, *Lichtheimia*, all belonging to the *Mucorales* (Mucoromycotina), *Conidiobolus* (Entomophthoromycotina), and *Basidiobolus* (Basidiobolaceae).

Rhizopus species, particularly *R. arrhizus* (*R. oryzae*), are the most common zygomycetous opportunists [3]. Species reported from clinical cases are *R. arrhizus*, *R. homothallicus*, *R. microsporus*, *R. schipperae*, and *R. stolonifer.* As a result of recent taxonomic studies, the names of some clinically important *Rhizopus* species have changed (Table 1) [4]. Three varieties are recognized within *R. arrhizus*: var. *arrhizus*, var. *tonkinensis*, and var. *delemar*, which are all involved in infections of vertebrates (Walther et al., unpublished data) [4]. Recently, *R. homothallicus* has been described from two cases in India to cause cavitary pulmonary zygomycosis (Chakrabarti et al., unpublished data).

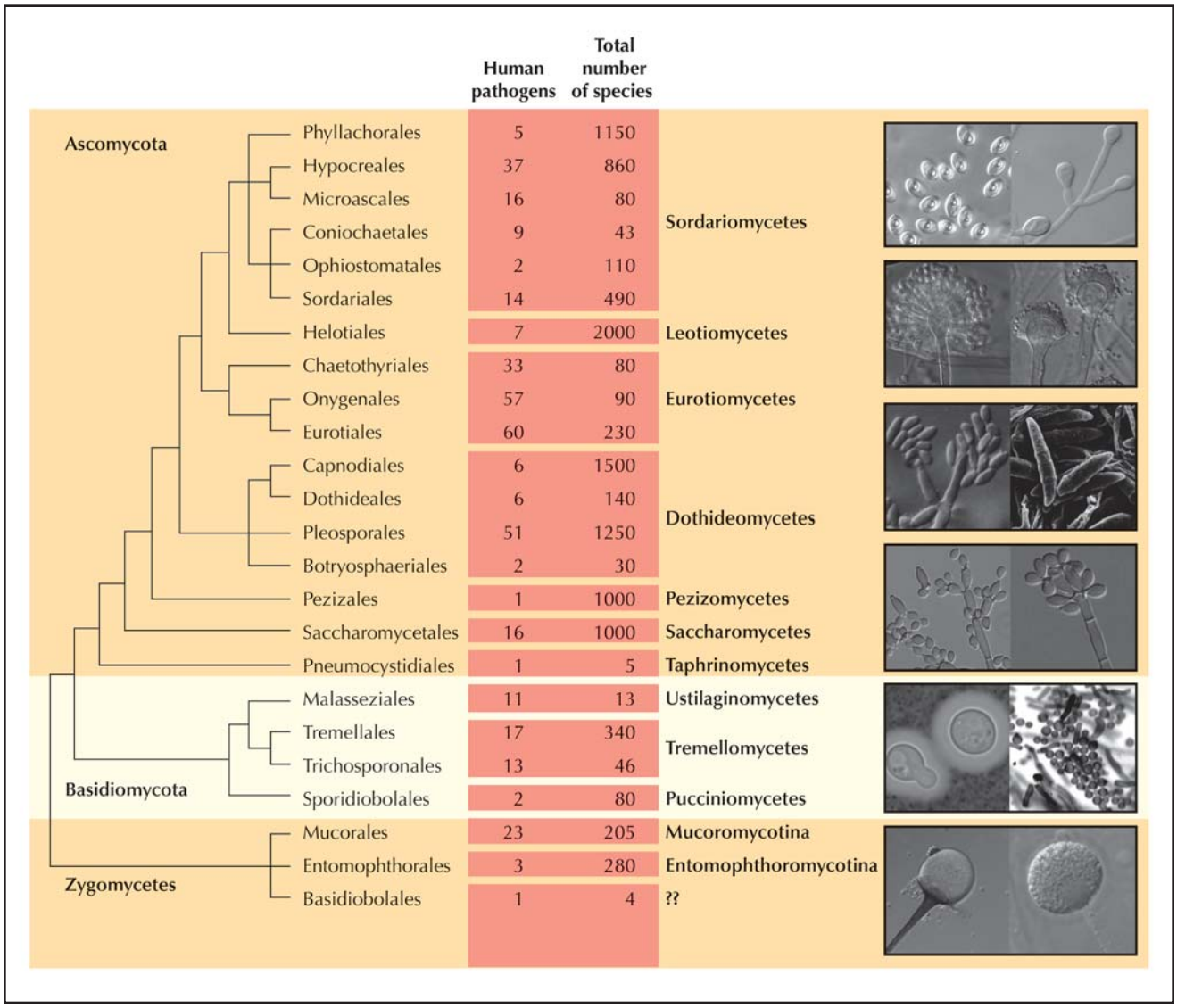

Figure 1. Simplified fungal tree of life showing the branches where human pathogens occur. Approximate numbers of pathogenic species and total number of species are indicated. Panel on the *right* shows some pathogenic fungi belonging to various parts of the tree of life as follows (*top to bottom*): Eurotiomycetes, *Pseudallescheria apiosperma*, ascospores (*left*), conidiophore and conidia (*right*); Eurotiomycetes, *Aspergillus calidoustus* (*left*), *A. fumigatiaffinis* (*right*), conidiophores and conidia; Sordariomycetes, *Fusarium oxysporum*, microconidia (*left*), *F. solani*, macroconidia (*right*); Eurotiomycetes, *Cladophialophora saturnica*, conidiophores and conidia (*left and right*); Tremellomycetes, *Cryptococcus gattii*, capsulated yeast cells (*left*), Ustilaginomycetes, *Malassezia globosa*, yeast cells and filaments in skin scrapings (*right*); Mucoromycotina, *Rhizopus arrhizus*, sporangiophore with columella (*left*), *Mucor hiemalis*, sporangium (*right*).

Mucor species are the second-most common agent of zygomycosis [3]. The most frequently occurring opportunistic species is *M. circinelloides* with forma *circinelloides* and forma *janssenii* (Walther et al., unpublished data). Other pathogens include *M. amphibiorum*, *M. hiemalis*, *M. indicus*, *M. plumbeus*, *M. racemosus*, and *M. ramosissimus*. Phylogenetic analysis using large subunit (LSU) rRNA gene and internal transcribed spacer (ITS) rDNA sequences revealed that the two varieties of *Rhizomucor variabilis*, both known as agents of zygomycosis, are related to *Mucor* (Walther et al., unpublished observation).

Absidia-like fungi represent three separate lineages: 1) *Absidia s.str.* growing at temperatures below 40° C (mesophilic); 2) *Leptomyces* (mesophilic); and 3) *Lichtheimia*, consisting of thermotolerant species that show good growth at body temperature [5]. *Lichtheimia* was initially named *Mycocladus*, but was recently renamed [6]. Sequence analysis using three different regions (ITS-region, D1/D2 domains of LSU, and translation elongation factor 1-α) revealed an apparently undescribed "*Mycocladus*" species associated with human zygomycosis that was named *Mycocladus lutetiensis* [7]. However, *M. lutetiensis* was recognized as a synonym of *Lichtheimia ramosa* (*Absidia ramosa*, *M. ramosus*). Besides *L. ramosa*, *L. corymbifera* (*A. corymbifera*, *M. corymbiferus*) and *L. ornata* (*A. ornata*) are clinically relevant (Alastruey-Izquierdo et al., unpublished data) (Table 1).

Table 1. Name changes of some clinically relevant fungi as a consequence of an improved taxonomy

Old name	New name
Zygomycetes	
Absidia ornata	*Lichtheimia ornata*
Mycocladus lutetiensis	*Lichtheimia ramosa*
Mycocladus (Absidia) corymbifera	*Lichtheimia corymbifera*
Rhizopus oryzae	*Rhizopus arrhizus*
Ascomycota	
Aspergillus fumigatus	*Neosartorya fumigata* (teleomorph)
Aspergillus fumigatus sl	*Aspergillus fumigatus* ss
	Aspergillus fumigatiaffinis
	Aspergillus lentulus
	Neosartorya udagawae
Clinical isolates of *Aspergillus ustus*	*Aspergillus calidoustus*
Aspergillus terreus sl	*Aspergillus terreus* ss
	Aspergillus alabamensis
Candida albicans sl	*Candida albicans* ss (including *C. stellatoidea*, *C. africana*)
	Candida dubliniensis
Candida glabrata sl	*Candida glabrata* ss
	Candida bracarensis
	Candida nivariensis
Candida parapsilosis sl	*Candida parapsilosis* ss
	Candida metapsilosis
	Candida orthopsilosis
Coccidioides immitis sl	*Coccidioides immitis* ss
	Coccidioides posadasii
Fusarium oxysporum sl	*Fusarium oxysporum* ss and 257 sequence types
Fusarium solani sl	*Fusarium solani* ss and 44 phylogenetic species, including *Fusarium falciforme*, *Fusarium lichenicola*, and *Neocosmospora vasinfecta*
Exophiala jeanselmei sl	*Exophiala jeanselmei* ss
	Exophiala xenobiotica
	Exophiala oligosperma

sl—sensu lato; ss—sensu stricto.

Ascomycetes

Aspergillus

The genus *Aspergillus* classified in order Eurotiales (class Eurotiomycetes) includes more than 250 species, among which about 20 have been reported to cause opportunistic infections in humans (Fig. 1) [2]. The most important human pathogens in this genus are assigned to sections *Fumigati*, *Flavi*, *Nigri*, *Terrei*, *Nidulantes*, and *Usti*. Correct species demarcation is important as it may affect therapy choice, warrant interventions to prevent invasive fungal infection in immunocompromised persons, or identify sources of nosocomial spreads. In the past, identification of aspergilli primarily relied on morphologic criteria. Reliable identifications, however, use sequence analysis (in conjunction with traditional phenotype-based methods) of the ITS regions and the β-tubulin or calmodulin genes [8].

A. fumigatus (section *Fumigati*) is the most common cause of invasive aspergillosis, a condition associated with substantial severity and mortality rates. Section *Fumigati* includes 10 anamorphic species and 23 species that reproduce sexually. The teleomorphs of these species have been assigned to the *Neosartorya* genus. Recently, the sexual stage of *A. fumigatus* has been described as *Neosartorya fumigata* [9•]. The presence of a sexual cycle provides a valuable tool for genetic analyses and will facilitate research into the genetic basis of pathogenicity and drug resistance in *A. fumigatus*, with the aim of improving methods to control aspergillosis. Recent molecular studies revealed that several clini-

Table 1. Name changes of some clinically relevant fungi as a consequence of an improved taxonomy ***(Continued)***

Old name	New name
Ascomycota	
Fonsecaea pedrosoi (including *F. compacta*)	*Fonsecaea pedrosoi* ss
	Fonsecaea monophora
Ramichloridium mackenziei	*Rhinocladiella mackenziei*
Phialophora richardsiae	*Pleurostomophora richardsiae*
Phaeoacremonium 3 spp	*Phaeoacremonium* 9 spp
Pneumocystis carinii sl	*Pneumocystis jiroveci*
	Four species on animals (including *Pneumocystis carinii* ss)
Pseudallescheria boydii sl, including *Scedosporium apiospermum* (anamorph)	*Pseudallescheria boydii* ss
	Pseudallescheria fusoidea
	Scedosporium apiospermum (anamorph)
	Scedosporium aurantiacum (anamorph)
	Scedosporium dehoogii (anamorph)
Basidiomycota	
Cryptococcus neoformans sl	*Cryptococcus neoformans* ss (including variety *grubii*)
	Cryptococcus gattii
Malassezia furfur sl	*Malassezia furfur* ss
	Malassezia globosa
	Malassezia restricta
	Nine other lipid-dependent species
Trichosporon beigelii	*Trichosporon asahii*
	Trichosporon asteroides
	Trichosporon cutaneum
	Trichosporon inkin
	Trichosporon mucoides
	Trichosporon ovoides

sl—sensu lato; ss—sensu stricto.

cal isolates, previously identified as *A. fumigatus* and that exhibit altered antifungal susceptibility, belong to other species, including *A. lentulus*, *N. udagawae*, and *A. fumigatiaffinis* (Fig. 1) [10].

Among black aspergilli (section *Nigri*), recent molecular data indicate that, besides *A. niger*, *A. tubingensis*, *A. awamori*, and *A. acidus* may also occur as human pathogens. In section *Usti*, *A. ustus* was considered a relatively rare human pathogen. However, recent reexamination of clinical isolates revealed that they represent a new species, *A. calidoustus* with decreased susceptibilities to several antifungal drugs [11]. In vitro experiments demonstrated that the triazoles, including posaconazole, are not active against this species. Invasive infections caused by *Emericella nidulans*, also known by its anamorph name *A. nidulans*, are uncommon in animals and humans. In humans, they appear to occur predominantly in patients with chronic granulomatous disease (CGD). Recent sequence-based re-identification of a set of clinical *Emericella* isolates identified a role of *E. quadrilineata* as an opportunistic fungal pathogen, especially in patients with CGD and in those with hematologic malignancy [12]. *E. nidulans* was less susceptible than *E. quadrilineata* to amphotericin B (AmB), but more susceptible to caspofungin, indicating that correct species demarcation and in vitro susceptibility testing may contribute to improved patient management. *Aspergillus terreus* (section *Terrei*), another important human pathogen that caused invasive aspergillosis in medical centers, has a decreased susceptibility to AmB. Multigene sequence analysis revealed that a new species, *Aspergillus alabamensis*, which is morphologically similar to *A. terreus*, may colonize immunocompetent humans and has a decreased in vitro susceptibility to AmB [13]. Several species of section *Flavi*, including *A. flavus*, *A. tamarii*, *A. nomius*, and *A. pseudonomius*, have also been identified as causative agents of fungal keratitis cases in India. *A. alliaceus*, which has reduced susceptibilities to AmB and caspofungin, can also cause aspergillosis.

Fusarium

Fusarium is a clinically important ascomycetous genus, classified in the order Hypocreales (class Sordariomycetes) (Fig. 1), which contains more than 30 species that may be implicated in outbreaks of diseases in humans ranging from benign to life-threatening (Fig. 1) [2]. In 1996, multiple *Fusarium* species were involved in nosocomial infections in a hospital in the United States [14,15]. In the Netherlands, the water supply system served as the environmental reservoir for such an outbreak. Other symptoms caused by these emergent opportunists include skin infections and disseminated diseases with a high rate of mortality. In the environment, *Fusarium* species are commonly found as saprophytes in soil or as plant pathogens worldwide.

Among species most frequently encountered in clinical settings are members of the *F. solani* and *F. oxysporum* species complexes. Traditionally, members of these species complexes have been referred to as single morphospecies (ie, *F. solani* and *F. oxysporum*). Because of their importance in agriculture, the use of a naming system based on the plant host has commonly been adopted. Recent molecular analyses have shown that these so-called *formae speciales* do not always correspond to natural groups and that each species complex may include a large number of phylogenetically distinct species (Table 1) [15–17]. Most newly discovered phylogenetic species have not yet been named formally. A system of multilocus haplotype nomenclature has been adopted to facilitate the use of these phylogenetic species [17]. It is important for clinicians to use molecular tools because the identification of susceptibility to antifungals differs among phylogenetic species.

Members of the *F. solani* complex were responsible for major contact lens–associated outbreaks of keratitis in the United States during 2005 and 2006 [16,18]. Recent studies showed that *F. solani* contains at least 45 phylogenetically distinct species distributed among three major clades [17]. Importantly, all clinically relevant strains cluster in a single group (clade 3) that contains at least 34 phylogenetic species, among which 20 are clinically relevant and only three have a formal name: *Fusarium falciforme*, an agent of white-grain mycetoma and opportunistic infections in patients with transplants [19]; *Fusarium lichenicola*, an agent of keratitis and cutaneous to disseminated infections [19]; and *Neocosmospora vasinfecta*, an agent of systemic infections in patients with transplants.

Similar to *F. solani*, *F. oxysporum* has been increasingly involved in disseminated infections of immunocompromised patients. Early molecular studies demonstrated that this morphospecies includes several phylogenetically distinct species. One recently dispersed and geographically widespread clonal lineage was the agent of many cases of nosocomial infections [15]. A recent study including 850 isolates of the *F. oxysporum* species complex showed that these isolates could be grouped into 257 sequence types using intergenic spacer (IGS) and elongation factor 1-α (EF1-α) gene sequences (O'Donnell et al., unpublished observation). Clinically relevant strains occur in 25 of these sequence types, some of them with apparent strict clinical origin, others also shared by plant pathogens.

A multistate haplotype nomenclature system based on molecular data (EF1-α gene and IGS sequences), called *Fusarium-ID*, is accessible online at http://isolate.fusariumdb.org/index.php [20]. This database, applicable in the fields of agriculture and medicine, will be a very powerful identification tool for clinically relevant fusaria.

Melanized fungi and *Scedosporium*

Melanized fungi belong to the order Chaetothyriales (class Eurotiomycetes) and typically cause infection in otherwise healthy individuals (Fig. 1) [2]. Chromoblastomycosis is known to be caused by a small number of interrelated species, and can be subdivided into a number of clinical types [21]. Phaeohyphomycosis is an umbrella term for a large diversity of infections in which brown hyphae are seen in tissue. The main category is brain infection. Chaetothyriales contain the largest number of agents infecting brain parenchyma. Most species characteristically cause primary encephalitis [22], characterized by neurologic symptoms that become apparent only in advanced disease stages. In disseminated infections, some species appear to be neurotropic, whereas others, such as *Exophiala spinifera*, are osteotropic. The fatality rate of all systemic invasive infections by melanized fungi remains high. Patients suffering from cystic fibrosis regularly show asymptomatic pulmonary colonization, whereas the same species is also found in the intestinal tract of asymptomatic carriers. A new, severely underreported category is mild cutaneous infection by black fungi caused, for example, by *Phialophora europaea* and *Coniosporium epidermidis*.

Multilocus gene sequence analyses of traditional pathogens, such as *Cladophialophora*, *Exophiala*, *Fonsecaea*, and *Phialophora* species, have shown that ITS data are sufficient for diagnostics [23]. *Exophiala jeanselmei*, an agent of human subcutaneous infections, proved to contain two cryptic species, *E. oligosperma* and *E. xenobiotica*, which are common agents of cutaneous infections (Table 1). In contrast, *E. asiatica* is known from a single, fatal disseminated case [24]. Four rare clinical *Cladophialophora* species cause different diseases, namely *C. saturnica* [25] and *C. immunda,* which are cutaneous; *C. mycetomatis*, which is subcutaneous; and *C. samoënsis*, an endemic agent of chromoblastomycosis [26]. *Fonsecaea compacta* has been reduced to synonymy with *F. pedrosoi*, but a sibling species *F. monophora* is repeatedly found in brain infections in addition to causing chromoblastomycosis [27]. The neurotropic fungus endemic to the Middle East is now referred to as *Rhinocladiella mackenziei.*

Some black fungi belong to other orders. *Phialophora richardsiae* has been renamed *Pleurostomophora richardsiae* (Calosphaeriales), and the genus *Phaeoacremonium (Diaporthales)* presently contains nine species that are clinically relevant [28].

Scedosporium and its teleomorph *Pseudallescheria* belong to the Microascales. The genus has been subdivided into a number of species that differ in virulence and predilection [29]. Most clinical strains belong to the *Scedosporium apiospermum* and *Pseudallescheria boydii*, which are now separate species, whereas *S. dehoogii* is mainly environmental [30].

Candida

Candida yeasts (asexual Saccharomycetales, Saccharomycotina) are a main source of infection (Fig. 1) [2]. *Candida* species are a common source of nosocomial bloodstream infections. The prevalence of *Candida* infections has increased considerably in the past decade, and has become a serious threat to high-risk patients (eg, HIV-infected patients, oncology patients, pediatric patients, transplant patients, neonates, newborns, women with recurrent vaginitis, patients with diabetes, and chronically bedridden patients) and often cause high rates of morbidity and mortality. The increase of *Candida* infections has seen an increase of non-*albicans Candida* species, mainly *C. glabrata*. Crude mortality is approximately 35%, with the highest values observed in the elderly, those who suffered from fungemia of *C. tropicalis*, and patients with solid tumors. Most isolates represent *Candida albicans*, followed by *C. glabrata*, *C. parapsilosis*, *C. tropicalis*, *C. krusei*, and other *Candida* species.

In recent years, a number of new clinically relevant *Candida* species has been recognized, namely *C. dubliniensis*, *C. nivariensis*, *C. bracarensis*, *C. orthopsilosis*, and *C. metapsilosis* (Table 1). *C. dubliniensis*, a sibling species of *C. albicans*, occurs worldwide and causes oral and oropharyngeal infections in HIV-infected AIDS patients, but also in those suffering from dental stomatitis, diabetes, cystic fibrosis, and neutropenia [31]. Most isolates are sensitive to commonly used antifungals, but resistance to fluconazole has been reported [31].

Recently, two sibling species of *C. glabrata*, *C. nivariensis* and *C. bracarensis*, have been described [32,33]. *C. nivariensis* was isolated from deep-seated infections, oropharynx of an HIV-infected patient, and a catheter-related infection from patients in Spain, Indonesia, Japan, and Great Britain. The species is susceptible to AmB, 5-flucytosine, the commonly used azoles, but also to voriconazole, posaconazole, caspofungin, and isavuconazole [34]. However, it has also been reported that the species is less susceptible to azoles [35]. *C. bracarensis* has been found in vulvovaginal candidiasis, blood, stool of an HIV-infected patient, abscess, and two oncology patients, and has been reported in Portugal, Great Britain, and the United States. Susceptibility to seven antifungals was similar to that of *C. glabrata*, except one isolate that was resistant to fluconazole, itraconazole, and posaconazole [36].

Two sibling species of *C. parapsilosis*, *C. orthopsilosis* and *C. metapsilosis,* were described to replace *C. parapsilosis* group II and III, respectively [37]. *C. orthopsilosis* has been reported to be involved in nosocomial outbreaks [38]. Differences in susceptibility to commonly used antifungals have been noted, but this does not seem to affect therapeutic choices [38].

Recently, multilocus sequencing–type (MLST) analysis has been proposed as the method of choice to genotype isolates of *C. albicans*, *C. dubliniensis*, *C. glabrata*, *C. krusei,* and *C. tropicalis* (as well as *Cryptococcus neoformans* and *Cr. gattii*) [39]. In an ongoing international collaboration, databases are rapidly filled with data from globally collected isolates, thus hinting at global patterns of dispersal, reproduction, and phylogeny within each species. The same data will also be most valuable to understand, for example, the origins of nosocomial outbreaks, relapse of infections, and microevolution as a consequence of antifungal treatment. Two available internet resources (http://pubmlst.org; http://www.mlst.net) provide data on the MLST genotypes of each species.

Basidiomycetes

Malassezia

Members of the genus *Malassezia*, classified in the order Malasseziales (class Ustilaginomycetes), are opportunistic yeasts of increasing importance. The genus had historically remained limited to *Malassezia furfur* and *M. pachydermatis*. *M. pachydermatis* is lipophilic but not lipid-dependent, and usually occurs in animals, where it may cause otitis externa, especially in dogs and cats. Until the recent taxonomic revision, pathologies caused by the lipid-dependent *M. furfur* sensu lato were ascribed to a single species [40]. Sequence analysis of the D1/D2 domains of the LSU rRNA gene, however, revealed six lipid-dependent species and, since then, seven more species have been reported (Table 1) [40].

Malassezia species are part of the normal skin microbiota, in particular of sebum-rich areas of the skin, such as the trunk, back, face, and scalp. Under certain conditions, *Malassezia* species may cause a superficial skin infection, and they have been associated with a number of diseases of the human skin, such as seborrheic dermatitis (SD) and dandruff, pityriasis versicolor (PV), *Malassezia (Pityrosporum)* folliculitis, atopic dermatitis (AD), and psoriasis [41]. Species other than *M. furfur* sensu stricto are implicated in most of these diseases, with *M. globosa* being the most common species cultured from PV lesions. *M. globosa* and *M. restricta* predominate lesions of SD and dandruff. In atopic dermatitis (AD), *Malassezia* species may act as allergens in susceptible patients. *M. pachydermatis* and *M. furfur* sensu stricto are occasionally involved in nosocomial outbreaks in neonates who receive intravenous lipid supplementation.

The recent elucidation of the genomes of *M. globosa* and *M. restricta* revealed important adaptations of the yeasts to the skin habitat [42•]. The absence of a fatty acid synthase gene explained the lipid dependence of both species, and multiple secreted lipases allow the fungus to scavenge lipids from the host. Secreted hydrolases

(eg, lipases, phospholipases, aspartyl proteases, and acid sphingomyelinases) are encoded by multiple genes. Interestingly, the arsenal of extracellular hydrolases encoded by the genome of *M. globosa* is similar to that of *C. albicans*, which phylogenetically is not closely related but occupies the same niche on skin. Finally, the genome sequence revealed the presence of mating-type genes, thus providing evidence that *Malassezia* may be capable of sexual reproduction.

Cryptococcus

Two species are recognized within the *C. neoformans–C. gattii* species complex, namely the asexual yeasts *C. neoformans* and *C. gattii* (Table 1). The species are classified in order Tremellales (ie, jelly fungi, class Tremellomycetes) (Fig. 1) [2]. Six haploid genotypic groups occur based on molecular fingerprinting and sequence analysis of coding and noncoding regions [43]. Two genotypic groups can be recognized within *C. neoformans* that are interpreted as varieties, namely var. *grubii* (serotype A) and var. *neoformans* (serotype D).

C. neoformans and *C. gattii* differ in epidemiology and ecologic niche. In immunocompetent individuals, *C. neoformans* infection is either cleared or remains dormant. However, in immunocompromised individuals, *C. neoformans* can disseminate to other organs, including the brain. Infection of the central nervous system may result in meningoencephalitis. *C. neoformans* var. *grubii* occurs throughout the world and causes by far the majority of cryptococcal infections in HIV-infected patients. In Europe, this variety occurred in about half of the isolates analyzed in a 30-month survey [44]. *C. neoformans* var. *neoformans* also occurs worldwide but seems more common in Europe.

C. gattii comprises four phylogenetic lineages that may represent species [43]. The species mainly infects immunocompetent individuals and occurs predominantly in tropical and subtropical areas. Serotype C isolates of *C. gattii*, however, were implicated in HIV-associated infections in California, Botswana, and Malawi [45]. The species has also been isolated in Europe in areas with a temperate or Mediterranean climate and has been reported in temperate climate zones in Colombia. One of the genotypes of *C. gattii* (AFLP6/VGII) is responsible for the ongoing outbreak of cryptococcosis on Vancouver Island, mainland Canada, and the United States [46,47]. Recently, some tourists who visited Vancouver Island also developed cryptococcal disease.

Diploid or aneuploid hybrids occur within the complex. The serotype AD hybrids (AFLP 3/VNIII) are well-studied and showed high incidences in Portugal, Greece, and Spain, for example [44]. Hybrids between *C. neoformans* and *C. gattii* were recently described based on molecular studies of clinical isolates and were designated as BD and AB hybrids, respectively.

Trichosporon

Trichosporon species are classified in order Trichosporonales (class Tremellomycetes) and cause infections in patients with hematologic malignancies that are associated with high mortality rates (Fig. 1). The taxonomy of the genus *Trichosporon* was revised based on sequence analysis of partial LSU rRNA sequences [48]. Six opportunistic human pathogens were recognized in the old species *T. beigelii* (Fig. 1): *T. asahii* and *T. mucoides* cause deep invasive infections; *T. asteroides* and *T. cutaneum* cause superficial skin infections; *T. ovoides* cause white piedra of the scalp and *T. inkin* is involved in white piedra of the pubic hair [48,49]. *T. asahii*, the clinically most important species, is isolated from sources such as blood, lung tissue, bones, and urine, but also skin, nails, and white piedra. New, clinically relevant species are emerging as discussed in Taj-Aldeen et al. [49]. *Guehomyces pullulans* (formerly *T. pullulans*) caused infection in patients with CGD and has been isolated from the oral cavity of patients with AIDS. *T. dermatis* caused fungemia in a 13-month-old male with a history of autoimmune enteropathy, and *T. asteroides*, *T. loubieri,* and *T. dermatis* were involved in disseminated trichosporonosis. *T. asahii* occurred as the most common species in a study on trichosporonosis in Qatar (63% of cases), followed by *T. inkin* and *T. dohaense* (11%), and *T. faecale* and *T. japonicum* (7.4%) [49]. The species may differ in antifungal susceptibilities [49,50]. *T. asahii*, *T. faecale,* and *T. coremiiforme* showed high minimal inhibitory concentrations (MICs) for AmB. Most isolates from the Qatar-based study showed high MICs for AmB and caspofungin, and anidulafungin demonstrated no in vitro activity against *Trichosporon* species. It was concluded that polyenes and echinocandins should not be used to treat *Trichosporon* infections, but itraconazole, voriconazole, posaconazole, and isavuconazole showed good in vitro activity against *Trichosporon* species, with the latter being the most potent agent [49].

Conclusions

It is clear that our understanding of fungal pathogens is changing considerably. In many cases, traditional taxa were found to represent species aggregates, and novel taxa from clinical sources are frequently discovered (Table 1). Such developments may not be appreciated by all professionals in the field, however, as one must keep track of all proposed modifications, even related to "textbook" pathogens. The refined taxonomy as a result of comparative molecular studies will contribute to a better understanding of the diseases they cause and result in appropriate treatment, thus contributing to improved patient care. Recent molecular phylogenetic studies have resulted in a considerable increase of newly recognized fungal pathogens. In most if not all cases, molecular identifications are needed to provide an accurate species assessment. In some cases, the newly discovered species were found to be important human pathogens, which need to be treated appropriately. Importantly, differences in susceptibility patterns have been noted to occur, thus indicating a need for proper identification of fungal pathogens.

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Disclosure

No potential conflicts of interest relevant to this article were reported.

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