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Variation in Susceptibility of Bloodstream Isolates of *Candida glabrata* to Fluconazole According to Patient Age and Geographic Location

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We examined the susceptibilities to fluconazole of 559 bloodstream infection isolates of *Candida glabrata* and grouped the isolates by patient age and geographic location within the United States. Susceptibility of *C. glabrata* to fluconazole was lowest in the Pacific (44%) and East South Central (47%) regions and was highest in the West South Central region (82%) (regions are as designated by the U.S. Bureau of the Census). Isolates from pediatric patients were virtually all susceptible to fluconazole, whereas the highest frequency of resistance was observed in isolates from patients 16 to 64 years of age.

Candida glabrata is an increasingly common cause of bloodstream infection in hospitalized patients (1, 3, 7, 12, 15, 16, 23). *C. glabrata* is innately less susceptible to fluconazole and amphotericin B than most other species of *Candida* (13, 17) and displays the capacity to rapidly develop resistance to all azoles via induction of the CDR efflux pumps (20–22, 25). Exposure of *C. glabrata* to subtherapeutic concentrations of fluconazole may result in resistance, and clinical studies have shown that the frequency of colonization and infection of patients with *C. glabrata* may be increased in populations subjected to fluconazole prophylaxis (1, 7, 10, 26). Additionally, the frequency of *C. glabrata* as a cause of bloodstream infection has been observed to increase with increasing patient age, although detailed studies of patients greater than 65 years of age are lacking (6, 8, 14–16). The emergence of *C. glabrata* as an important bloodstream infection pathogen may not be a simple matter of selective pressure by a drug (e.g., fluconazole) but may also be influenced by patient age, underlying diseases, geographic location, or other unknown factors (4, 8, 16, 18, 22).

The purpose of this study was to describe the variation in fluconazole susceptibility of *C. glabrata* bloodstream isolates stratified by patient age and geographic location in the United States.

Between 1992 and 2001, a total of 3,683 bloodstream isolates of *Candida* spp. from 167 medical centers in the United States were submitted to the University of Iowa College of Medicine (Iowa City) for identification and antifungal susceptibility testing with fluconazole. The isolates represented consecutive-incident isolates from patients with candidemia treated at hospitals within all nine U.S. Bureau of the Census regions (Table 1) (16). Patient ages were provided for 559 (83%) of the 674 bloodstream isolates of *C. glabrata*. These 559 isolates constitute the study set described herein.

All *C. glabrata* isolates were identified using Vitek and API products (bioMerieux, Hazelwood, Mo.), the results with which were supplemented by conventional methods as required, and stored as water suspensions until they were used. Prior to testing, each isolate was passaged on potato dextrose agar (Remel, Lenexa, Kans.) and CHROMagar (Hardy Laboratories, Santa Monica, Calif.) to ensure purity and viability.

Fluconazole was obtained from the manufacturer (Pfizer, New York, N.Y.) as a standard reagent-grade powder. Broth microdilution testing was performed in accordance with the guidelines in NCCLS document M27-A (11). The interpretive criteria for fluconazole were those published by the NCCLS (11), as follows: an isolate for which the MIC is ≤8 μg/ml is susceptible, an isolate for which the MIC is 16 to 32 μg/ml is susceptible in a dose-dependent manner, and an isolate for which the MIC is ≥64 μg/ml is resistant.

Quality control was accomplished by testing the following strains: *C. parapsilosis* ATCC 22019 and *C. krusei* ATCC 6258 (5, 11).

Overall, *C. glabrata* accounted for 18.3% of all *Candida* sp. bloodstream isolates and was the second most common *Candida* sp. isolated. The frequency of *C. glabrata* as a cause of candidemia in the United States ranged from 11.7% in region 4 (West South Central) to 37.3% in region 7 (New England) (Table 1).

The fluconazole susceptibilities of *C. glabrata* bloodstream isolates varied by region (Table 2). Interestingly, there was no relationship between the prevalence of *C. glabrata* as a cause of bloodstream infection and the rate of resistance (MIC, ≥64 μg/ml) to fluconazole; the region with the highest prevalence of *C. glabrata* (New England; 37.3%) had the lowest frequency of resistance (0%). The three regions with the highest percentage of resistance (regions 6, 7, and 9) were contiguous in the eastern and southeastern United States (Tables 1 and 2).

As expected from previous studies (14, 15, 19), few bloodstream infections due to *C. glabrata* were reported from pediatric age groups (≤15 years) (Table 2). Only 13 *C. glabrata* bloodstream isolates were submitted from patients ≤15 years of age. Only 1 of the 13 was resistant to fluconazole. Fluconazole resistance was greatest among *C. glabrata* isolates from patients 16 to 64 years of age (12%).

Although much has been written regarding the importance of fluconazole resistance among clinical isolates of *C. glabrata* (3, 7, 12, 18, 22–24, 26), we found that resistance to fluconazole

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TABLE 1. Frequency of *C. glabrata* as a cause of bloodstream infection in each of the nine U.S. Bureau of the Census regions

Region	Total no. of participating centers	Total no. of *Candida* bloodstream isolates	No. (%) of *C. glabrata* isolates
1. Pacific	48	439	81 (18.5)
2. Mountain	11	261	40 (15.3)
3. West North Central	21	302	57 (18.9)
4. West South Central	4	162	19 (11.7)
5. East North Central	12	376	99 (26.3)
6. East South Central	3	94	17 (18.0)
7. New England	3	83	31 (37.3)
8. Mid-Atlantic	13	736	138 (18.8)
9. South Atlantic	52	1,230	191 (15.5)
Total	167	3,683	674 (18.3)

among bloodstream isolates of *C. glabrata* was much lower than 10% in seven of the nine U.S. census regions and in four of the six patient age groups (Table 2). Aside from the Mid-Atlantic and South Atlantic regions (regions 8 and 9, respectively), resistance was negligible among *C. glabrata* bloodstream isolates from pediatric patients and from individuals >65 years of age. Even in the region with the highest prevalence of *C. glabrata* (region 7; 37.3%), the resistance rate was nil (Table 2).

Because we could not obtain fluconazole usage data for these regions, our study was not designed to examine the relationship between the emergence of *C. glabrata* as a fluconazole-resistant bloodstream infection pathogen and the level of fluconazole use (23). Although some investigators have found a strong association between fluconazole use and colonization and infection with *C. glabrata* (1, 26), others have not been able to corroborate those findings (2–4, 12). Baddley et al. (3) reported a stable rate of resistance to fluconazole of 10% among *C. glabrata* isolates despite a fluctuating prevalence of 18 to 31% and a steadily increasing usage of fluconazole over a 6-year period. Ásmundsdóttir et al. (2) found that although the usage of fluconazole in Iceland increased fourfold between 1990 and 1999, there was no increase in the prevalence of *C. glabrata* or in fluconazole resistance. Likewise, Nguyen et al. (12) found that *C. glabrata* was a prominent cause of breakthrough fungemia in amphotericin B-treated patients but not in those patients treated with fluconazole. Finally, if simply the selective pressure of fluconazole were responsible for both the emergence of *C. glabrata* and fluconazole resistance, one would expect that our data would reveal more fluconazole-resistant *C. glabrata* isolates in those regions with the highest prevalence of *C. glabrata*. Instead, the region with the highest *C. glabrata* prevalence had no fluconazole-resistant *C. glabrata* isolates.

The emergence of *C. glabrata* as a cause of bloodstream infection is therefore likely to be due to a complex set of factors, including changing demographics, antifungal usage, and an increasingly immunocompromised patient population (4, 18, 22). As one example, changes in mucosal defenses with

TABLE 2. Fluconazole susceptibilities of *C. glabrata* bloodstream infection isolates by patient age group in each of the nine U.S. Bureau of the Census regions[a]

Patient age group (yr[s])	No. tested	Isolate susceptibility category	% of S, S-DD, and R isolates (no. tested) in U.S. Census region:									
			1 (52)	2 (28)	3 (51)	4 (17)	5 (80)	6 (17)	7 (27)	8 (117)	9 (170)	All (559)
≤1	4	S	100	100						100	100	100
		S-DD	0	0						0	0	0
		R	0	0						0	0	0
2–15	9	S		100			100			50	33	67
		S-DD		0			0			50	33	22
		R		0			0			0	33	11
16–64	306	S	45	75	77	86	58	50	50	58	47	56
		S-DD	41	25	23	7	30	25	50	28	41	32
		R	14	0	0	7	12	25	0	14	12	12
65–74	113	S	40	33	79	100	64	0	57	67	71	64
		S-DD	60	33	21	0	36	100	43	6	26	29
		R	0	33	0	0	0	0	0	27	3	7
75–80	69	S	100	100	50	50	88		83	71	62	68
		S-DD	0	0	42	50	12		17	23	28	26
		R	0	0	8	0	0		0	6	10	6
>80	58	S	30	100	33		38		100	60	63	57
		S-DD	70	0	67		50		0	20	26	34
		R	0	0	0		12		0	20	11	9
All	559	S	44	71	69	82	61	47	70	62	55	60
		S-DD	48	22	29	12	31	30	30	23	35	31
		R	8	7	2	6	8	23	0	15	10	9

[a] Fluconazole susceptibility categories are as described in NCCLS document M27-A (11). S, the isolates are susceptible (MIC, ≤8 μg/ml; S-DD, the isolates are susceptible in a dose-dependent manner (MIC, 16 to 32 μg/ml); R, the isolates are resistant (MIC, ≥64 μg/ml).

age may underlie the observation that healthy older individuals with no exposure to fluconazole become colonized with *C. glabrata* (9). Fluconazole-naïve strains of *C. glabrata* may not demonstrate resistance to fluconazole; thus, bloodstream infection, when it occurs in individuals older than 70 years of age, may be more a function of changes in the ecology of the candidal flora with age than selection of *C. glabrata* due to drug pressure. Considerably more investigation is necessary before these complex issues are understood.

The relatively small number of isolates from several regions and age groups limits this study. In addition, we could not obtain complete data on several important patient-related factors that might have influenced the risk for *C. glabrata* or the risk for fluconazole resistance (e.g., severity of illness, device use, underlying disease, and antifungal use). Nonetheless, the overall size of this collection of *C. glabrata* bloodstream isolates does provide important descriptive information. We have provided the first U.S. regional surveillance data on bloodstream infection due to *C. glabrata*. This information may serve as the baseline for future studies regarding the prevalence and antifungal susceptibility of *C. glabrata* as a bloodstream pathogen.

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