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ORIGINAL ARTICLE

Changing epidemiology of candidemia in a medical center in middle Taiwan

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Background: Candidemia remains a major cause of morbidity and mortality in the health care setting, and the epidemiology of *Candida* infection is changing.
Methods: Clinical and laboratory data from patients with candidemia were collected retrospectively at a tertiary medical center in Taiwan from July 1, 2009 to June 30, 2012 (a 36-month period). Demographics, clinical characteristics, and drug susceptibility of the invading *Candida* species of patients at the onset of candidemia were analyzed and compared with previous study from January 1, 2001 to June 30, 2003 (a 30-month period).
Results: A total of 209 episodes of candidemia in 205 patients were identified in this study period. When compared with the previous study period, more patients were admitted for medical conditions at percentages ranging from 49.5% to 69.8%; the incidence rate of health care-associated candidemia increased from 0.76 to 1.14 per 1000 discharges; the proportion of *Candida albicans* in patients with candidemia decreased from 64.8% to 43.6% whereas the proportion of *Candida glabrata* increased greatly from 1.1% to 21.6% and the proportions of *Candida tropicalis* and *Candida parapsilosis* were slightly elevated (19.8–22.0% and 2.2–7.3%, respectively). All of the *C. albicans* isolates remained susceptible to fluconazole, whereas 66.7% of *C. glabrata* isolates were dose-dependent susceptible, and 4.4% of *C. glabrata* isolates and 11.6% *C. tropicalis* isolates were resistant. There was one *C. glabrata* and one *Candida guilliermondii* resistant to echinocandin. The predictors for 30-day mortality included the high Acute Physiology and Chronic Health Evaluation II (APACHE II) score, use of parenteral nutrition, underlying malignancy, liver cirrhosis, and neutropenia whereas candidemia by *C. parapsilosis* or *C. glabrata* is a favorable predictor when compared with *C. albicans*.

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Conclusion: The distribution of *Candida* species in candidemia was changed. Although *C. albicans* remained the major species, the isolation of non-*C. albicans* spp., especially *C. glabrata*, increased. Patients with candidemia still had high mortalities due to severity of illness and underlying conditions.

Introduction

Invasive candidiasis has been one of the important issues in clinical settings and has been a leading cause of health care-associated infections (HAIs), resulting in increased length and cost of hospitalizations as well as morbidities/mortalities of patients.[1] In Taiwan, candidemia is the second most common cause of health care-associated bloodstream infections (HA-BSIs), ranking only after bacteremia by *Staphylococcus* species and followed by *Acinetobacter baumannii* and *Escherichia coli* bacteremia. Furthermore, although health care-associated bloodstream infections by pathogens such as *S. aureus* and *Pseudomonas aeruginosa* are decreasing, the incidence of candidemia is increasing.[2–4] The reasons for increased candidal infections are thought to be due to the use of broad-spectrum antibiotics and immunosuppressants such as steroids, prolonged hospital and intensive care unit (ICU) stay, increased invasive procedures in medical practices, and more complicated underlying diseases such as malignancies, as well as prolonged survival of patients in the modern health care system.[5–7] The epidemiology of candidemia, however, varies among different geographic areas and time periods.[8] Therefore, the knowledge of the local epidemiology of candidal infections is important to offer the sound management of invasive candidiasis.[9] In this study, we retrospectively reviewed the cases of candidemia from July 1, 2009 to June 30, 2012 in Changhua Christian Hospital (CCH), performed the antifungal susceptibility testing for the available clinical isolates, and compared the results with that of our previous study[10] to clarify the changing epidemiology of candidemia.

Materials and methods

Patients and setting

All the patients were included if they were older than 16 years with at least one blood culture positive for *Candida* species from July 1, 2009 to June 30, 2012 in Changhua Christian Hospital, a 1700-bed tertiary hospital providing primary and tertiary patient care in middle Taiwan with 166 ICU beds for medical and surgical adult patients and pediatric patients.

Data collected from the medical records of the patients included demographic characteristics, underlying diseases (e.g., malignancy, diabetes mellitus, renal failure, liver cirrhosis), other associated conditions (e.g., concurrent bacteremia, *Candida* colonization, neutropenia, broad-spectrum antibiotics use, central venous catheterization, shock, parenteral nutrition, abdominal surgery, and steroid use), Acute Physiology and Chronic Health Evaluation II (APACHE II) scores, and outcome.

Definition

One episode of candidemia was defined by the isolation of *Candida* species from one or more blood cultures in a patient. If more than one blood culture was positive, a new episode was defined if more than 30 days had elapsed since the previous positive blood culture unless a persistent focus was identified. Episodes of candidemia were categorized as community acquired or healthcare associated following the definition of the Centers for Disease Control and Prevention.[11,12] Except for impaired renal function, which was determined by estimated glomerular filtration rate < 30 mL/min/1.73 m^2 or creatinine more than 2 mg/dL on presentation, the classification of patients with underlying conditions including malignancy, diabetes mellitus, and liver cirrhosis were determined by clear documentation of the diagnoses and/or associated treatment in medical records. Other definitions of terms used in this study are in concordance with our previous study[10] and are described briefly: *Candida* colonization is defined as the presence of the same *Candida* species in both non-sterile (obtained from urine, stool, and pus from a surgical wound) and sterile (such as a central venous catheter tip) specimens within the 14 days before and after the onset candidemia. Neutropenia is defined as absolute neutrophil count < 500 cells/mm^3. Central venous catheterization is the presence of a central venous catheter within 2 weeks of the onset of candidemia. Shock is defined as a decrease in systolic blood pressure to a level of less than 90 mmHg or a decrease of at least 40 mmHg from baseline blood pressure despite adequate fluid resuscitation or the use of vasopressors. Corticosteroid treatment is defined as receiving corticosteroids equivalent to prednisolone to at least 20 mg/day more than 7 days within 4 weeks of the onset of candidemia. Abdominal surgery is defined as an abdominal surgical procedure that was performed within 1 month of the onset of candidemia. Severity of illness was assessed by APACHE II scores. Receipt of parenteral nutrition is defined as the administration of lipid emulsion nutrition via peripheral or central veins for more than 3 days within 1 week prior to candidemia episode. Thirty-day crude mortality was used for primary outcome analysis.

Isolation, identification, and antifungal susceptibility testing of *Candida* isolates

There was no difference in the culture system and identification system between the two study periods. The blood

specimens were cultured in the BD BACTEC FX (Becton Dickinson, Sparks, MD, USA, www.bd.com) blood culture system, which is fully automated for microorganism growth detection. A yeast-like microorganism was detected by microscopic inspection after Gram staining and the growth characteristics were seen on agar plates. Further identification was performed using the API Candida Manufacturer: BioMerieux, Marcy L-Etoile, France yeast identification system. All blood isolates were stored at −80°C in the Thermo Scientific Forma (Thermo Fisher Scientific, Waltham, Massachusetts, USA, www.thermofisher.com) 900 series 955 model. For drug susceptibility testing, all available stored isolates were thawed and inoculated to Sabouraud dextrose agars (SDA) and incubated in an incubator at 35°C for 24−25 hours. The procedures were done according to manufacturer's protocol of Sensitire YeastOne (TREK Diagnostic Systems, Units 17-19 Birches Industrial Estate, East Grinstead,West Sussex. RH19 1XZ, UK).[13] Briefly, we placed the yeast colonies from the SDA plates into test tubes containing 10 μL normal saline, and vortexed and adjusted the turbidity of the suspensions to 0.5 Mcfarland. The suspensions were then placed into the panel wells and incubated in a 35°C incubator for 24−25 hours. A control well without an antifungal agent was used for ensuring the quality of growth and purity that the suspension from the well after incubation was subcultured onto another SDA plate to make sure the growth of 10−80 pure colonies for each corresponding candidal isolate. The panel wells were read and interpreted automatically using a Vizion instrument (Thermo Scientific Sensititre Vizion Digital MIC Viewing System, Manufacturer: Thermo Fisher Scientific, Waltham, Massachusetts, USA, www.thermofisher.com).

Statistical analysis

Univariate analyses were performed to examine the differences of characteristics of patients between two study periods and to identify possible risk factors associated with 30-day mortality of patients. For categorical measures, the Pearson chi-square or Fisher exact test was carried out as appropriate. For continuous measures, the Student *t* test was used. Results with $p < 0.05$ were considered statistically significant. Multivariate analyses using stepwise logistic regression were performed to identify independent factors associated with 30-day mortality in patients with candidemia. Predictors that improved the multiple logistic regression model with $p < 0.05$ were allowed to enter into the model.

Results

Characteristics of patients

During the study period, 217 cases of candidemia were identified from the database of the microbiological laboratory in our institute and 12 pediatric cases (age 16 years or younger) were excluded from the study. A total of 218 *Candida* isolates from 209 candidemia episodes in 205 patients were included for analysis. Among these patients, four patients had two candidemia episodes, including one having the same species (*C. glabrata*) in two episodes and three having different species for each episode (*C. albicans*/*C. tropicalis*, *C. glabrata*/*C. tropicalis* and *C. albicans*/*Candida* spp. in each patient) and 201 patients had one candidemia episode, including nine patients having two kinds of candidal species in the same episode (*C. albicans*/*C. glabrata* in four patients, *C. glabrata*/*C. tropicalis* in two, and *C. albicans*/*C. tropicalis*, *C. parapsilosis*/*C. tropicalis* and *C. famata*/*Candida* spp. both in one patient each). Among 209 candidemic episodes, 195 (93.3%) were healthcare associated. The incidence of healthcare-associated candidemia was estimated to be 1.14 episodes per 1000 discharges in the current study period, significantly higher than that in the previous study (0.76/1000 discharges, $p < 0.002$). The demographic and clinical characteristics of patients were summarized and compared with that of a previous study as shown in Table 1.

When compared with the previous study, more patients (69.8%) were admitted to the medical units while the proportion of admissions to the surgical intensive care units (SICU) was significantly decreased from 36.3% to 10.0% ($p < 0.001$). There was an increasing trend in the proportion of patients with malignancy although not statistically significant (38.5% vs. 46.9%, $p = 0.177$). For known factors predisposing to candidemia, the percentage of patients with recent abdominal surgery was significantly decreased from 39.6% to 21.5% ($p = 0.001$). A decreased proportion of patents with parenteral nutrition from 40.7% to 32.5% and increased proportion of patients with neutropenia from 6.6% to 11% were noted, but these were not statistically significant ($p = 0.175$ and 0.235, respectively). The distribution of *Candida* species in candidemia was different between the two study periods. Although *C. albicans* remained the most common isolated candidal species, the proportion declined from 64.8% to 43.6%. This was largely due to the significantly increased isolation of *C. glabrata* (1.1−21.6%), which had become the third most common isolated candidal species in the current study. The frequencies of isolation for *C. tropicalis* and *C. parapsilosis* were also slightly elevated (19.8% to 22.0% and 2.2% to 7.3%, respectively) and partially responsible for the increased proportion of non-*C. albicans* candidemia.

The characteristics of patients with pure candidemia of the four major *Candida* species, e.g., *C. albicans*, *C. tropicalis*, *C. glabrata*, *and C. parapsilosis* in their episodes were compared to identify host factors associated with specific *Candida* species in Table 2. We revealed that patients with *C. glabrata* candidemia were more likely to be female (58.5%) and older (mean age ± standard deviation [SD] = 70 ± 16); patients with *C. tropicalis* candidemia were less likely to have diabetes mellitus (20.5%) and more likely to be predisposed to neutropenia (27.3%); patients with *C. parapsilosis* candidemia were younger (mean age ± SD = 60 ± 15), and less likely to have colonization of the same species at sites other than bloodstream (13.3%); and patients with *C. albicans* or *C. glabrata* were likely to present with impaired renal function. Higher proportion of patients with *C. tropicalis* or *C. glabrata* were exposed to fluconazole before onset of candidemia (22.7% and 19.5%, respectively) but this is not statistically significant ($p = 0.210$). Patients with *C. tropicalis* had the worst outcomes with 65.9% of 30-day mortality whereas *C. parapsilosis* was associated with the lowest mortality rate (20.0% of 30-day mortality).

Table 1 Comparison of the characteristics of patients with candidemia in 2001–2003 versus 2009–2012

		Study period		*p*
		2001/1–2003/6 (*N* = 91)	2009/7–2012/6 (*N* = 209)	
Sex	M	51 (56.0)	130 (62.2)	0.316
	F	40 (44.0)	79 (37.8)	
Age, y		68 ± 11 (32–96)	66 ± 15 (20–95)	0.875
Admitting ward type	ER	0 (0)	1 (0.5)	< 0.001
	Medical ward	22 (24.2)	100 (47.8)	
	MICU	23 (25.3)	46 (22.0)	
	SICU	33 (36.3)	21 (10.0)	
	Surgical ward	13 (14.3)	41 (19.6)	
APACHE II score		18 ± 9 (0–38)	19 ± 8 (0–42)	0.616
Time from admission to candidemia, d		19 ± 16 (0–107)	16 ± 18 (0–118)	0.021
Time from candidemia to antifungal therapy, d		2 ± 3 (−19)	0 ± 3 (−23)	< 0.001
Length of hospital stay, d		39 ± 31 (0–176)	37 ± 37 (0–270)	0.153
Colonization with *Candida* species[a]		48/79 (60.8)	101/179 (56.4)	0.74
Concurrent bacteremia		21 (23.1)	71 (34.0)	0.06
30-d mortality		47 (51.6)	112 (53.6)	0.757
Underlying conditions				
Malignancy		35 (38.5)	98 (46.9)	0.177
Diabetes mellitus		33 (36.3)	71 (34.0)	0.701
Impaired renal function		44 (48.4)	97 (46.4)	0.757
Cirrhosis		9 (9.9)	18 (8.6)	0.722
Predisposing factors				
Broad spectrum antibiotic exposure		77 (84.6)	170 (81.3)	0.494
Central venous catheter		77 (84.6)	171 (81.8)	0.556
Parenteral nutrition		37 (40.7)	68 (32.5)	0.175
Recent abdominal surgery		36 (39.6)	45 (21.5)	0.001
Use of steroid		32 (35.2)	70 (33.5)	0.779
Neutropenia		6 (6.6)	23 (11.0)	0.235
Candida species				
C. albicans		59 (64.8)	95 (43.6)	< 0.001
C. tropicalis		18 (19.8)	48 (22.0)	
C. glabrata		1 (1.1)	47 (21.6)	
C. parapsilosis		2 (2.2)	16 (7.3)	
C. krusei		1 (1.1)	3 (1.4)	
C. guilliermondii		6 (6.6)	2 (0.9)	
C. famata		1 (1.1)	1 (0.5)	
C. membranifaciens		2 (2.2)	0 (0.0)	
C. pelliculosa		0 (0.0)	1 (0.5)	
C. sake		1 (1.1)	0 (0.0)	
Undetermined *Candida* species		0 (0.0)	5 (2.3)	

[a] *n*/*n*(%): no. of positivity for same *Candida* species/no. of microbiological investigation during candidemia episodes.
Data are *n* (%) or mean ± standard deviation (minimum–maximum) unless otherwise indicated.
APACHE II = Acute Physiology and Chronic Health Evaluation II; ER = emergency room; MICU = medical intensive care unit; SICU = surgical intensive care unit.

Antifungal susceptibility of clinical isolates

A total of 205 clinical isolates from 190 patients from the culture collections of the microbiological laboratory in our institute were available for susceptibility testing. Three isolates from three patients failed to produce results of minimum inhibitory concentration for the drugs tested drugs and finally a total of 202 candidal isolates from 187 patients were analyzed. The results are shown in Table 3. Notably, no resistance to fluconazole was detected in 90 *C. albicans* isolates. Two (4.4%) isolates of *C. glabrata* were resistant to fluconazole and 30 (66.7%) isolates were dose-dependent susceptible. There was no resistance to fluconazole in 18 *C. tropicalis* isolates in our previous study, whereas five (11.6%) of *C. tropicalis* isolates showed resistance to fluconazole in the current study. *C. parapsilosis* isolates were well susceptible to fluconazole. All candidal isolates except two (one *C. glabrata* and one *C. guilliermondii)* were susceptible to echinocandins. It was not feasible to compare the susceptibility of *Candida* spp. to echinocandins with that of our previous study in which the susceptibility for echinocandins was not performed.

Table 4 showed that in the current study, female sex and previous broad-spectrum antibiotic and fluconazole exposure were significant factors associated with candidemia by fluconazole reduced-susceptible *Candida* strains in

Table 2 Comparisons of characteristics among the four most isolated *Candida* species

Variables[b]	*C. albicans*	*C. tropicalis*	*C. glabrata*	*C. parapsilosis*	*p*
	$n = 90$	$n = 44$	$n = 41$	$n = 15$	
Sex, male	62 (68.9)	28 (63.6)	17 (41.5)	10 (66.7)	0.026[a]
Age, y	66 ± 14	62 ± 19	70 ± 16	60 ± 15	0.050
APACHE II score	19 ± 10	20 ± 8	20 ± 8	17 ± 9	0.698
Underlying disease					
Malignancy	40 (44.4)	22 (50)	19 (46.3)	10 (66.7)	0.446
DM	37 (41.1)	9 (20.5)	15 (36.6)	7 (46.7)	0.094
Impaired renal function	47 (52.2)	15 (34.1)	24 (58.5)	4 (26.7)	0.035[a]
Cirrhosis	11 (12.2)	4 (9.1)	1 (2.4)	0 (0)	0.172
Predisposing factor					
Broad spectrum antibiotic exposure	71 (78.9)	39 (88.6)	36 (87.8)	9 (60)	0.055
CVC	75 (83.3)	36 (81.8)	30 (73.2)	13 (86.7)	0.515
Parenteral nutrition	28 (31.1)	14 (31.8)	13 (31.7)	6 (40)	0.924
Recent abdominal surgery	20 (22.2)	6 (13.6)	11 (26.8)	4 (26.7)	0.465
Use of steroid	30 (33.3)	16 (36.4)	14 (34.1)	3 (20)	0.707
Neutropenia	4 (4.4)	12 (27.3)	2 (4.9)	2 (13.3)	< 0.001[a]
Associated conditions					
Fluconazole exposure	10 (11.1)	10 (22.7)	8 (19.5)	1 (6.7)	0.210
Colonization investigation	79 (87.8)	37 (84.1)	35 (85.4)	12 (80)	0.846
Colonization with *Candida* species	47 (52.2)	20 (45.5)	21 (51.2)	2 (13.3)	0.043[a]
Concurrent bacteremia	36 (40)	11 (25)	11 (26.8)	5 (33.3)	0.265
30-d mortality	51 (56.7)	29 (65.9)	17 (41.5)	3 (20)	0.007[a]

[a] $p < 0.05$.
[b] Data are *n* (%) or mean ± SD unless otherwise indicated.
APACHE II = Acute Physiology and Chronic Health Evaluation II; CVC = central venous catheter; ER = emergency room; MICU = medical intensive care unit; SICU = surgical intensive care unit.

univariate analysis. Female sex and fluconazole exposure are independent factors in multivariate analysis.

Treatment and outcome analysis

Of the 209 candidemia episodes, 39 (18.7%) were not treated with antifungal agents. Most (28 of 39, 71.8%) were due to deaths that occurred before the diagnoses of candidemia were made and effective treatment could be commenced. In 170 treated episodes, the mean time from candidemia episode to antifungal therapy was 0 ± 3 days and was significantly shorter than that of our previous study (2 ± 3 days, $p < 0.001$). However, the 30-day overall mortality rate did not change (51.6% vs. 53.6%, $p = 0.757$).

In identification of predictors for mortality in current study, we revealed that APACHE II score, malignancy, liver cirrhosis, use of parenteral nutrition, presence of central venous catheter, use of steroid, neutropenia, and *Candida* spp. were significantly associated with 30-day mortality in univariate analysis. The effect of APACHE II score, malignancy, liver cirrhosis, use of parenteral nutrition, neutropenia, and *Candida* species on mortality was still present in multivariate analysis. Patients with *C. glabrata* or *C. parapsilosis* candidemia had more favorable outcomes then those with *C. albicans* or *C. tropicalis* candidemia. The time from onset of candidemia to effective antifungal therapy and the susceptibility of *Candida* isolates did not show a significant difference between survived and fatal episodes. The results are shown in Table 5.

Discussion

In Taiwan, the incidence of candidiasis in the health care setting has been high and has increased in recent years.[4] Our study revealed an increased trend of incidence of health care-associated candidemia in our institute from 0.76/1000 discharges during 2001—2003 to 1.14/1000 discharges during 2009—2012. Our candidemia incidence was lower than that in the report by Chen et al,[14] which compared the characteristics of patients and the incidence of candidemia between 2002 and 2010 in their hospital, and although the incidence of candidemia did not increase significantly (2.78 per 1000 admissions in 2002 and 2.88 per 1000 admissions in 2010, $p = 0.71$), the incidence density increased from 0.34 per 1000 patient-days in 2002 to 0.41 per 1000 patient-days in 2010 ($p = 0.04$). The increased incidence density of candidemia is thought to be associated at least partly with the increase in the proportion of patients at a higher risk of candidemia, such as older age, higher comorbidity, and more underlying disease/status, including chronic pulmonary diseases, moderate to severe renal diseases, leukemia, lymphoma, and gastrointestinal malignancies. Although a hospital-wide analysis of patient papulation is not performed in the current study, the increased proportion of medical patients and decreased proportion of patients with abdominal surgery among candidemia cases in our two study periods still indicated the changing of the patient population and medical practices in our institute.

Table 3 *In vitro* susceptibility test of *Candida* species of antifungal agents

Species (No.)	Antifungal agent	Range		MIC (μg/mL)		%		(%) R or Non-S
				50%	90%	S	SDD	
C. albicans (90)	Amphotericin B	≤0.12	0.5	0.5	0.5	NA	NA	NA
	Fluconazole	≤0.12	8	0.25	0.5	100	0	0
	Voriconazole	≤0.008	0.12	≤0.008	≤0.008	100	0	0
	Itraconazole	≤0.015	0.5	0.03	0.06	98.9	1.1	0
	Posaconazole	≤0.008	2	0.015	0.03	NA	NA	NA
	Micafungin	≤0.008	≤0.008	≤0.008	≤0.008	100	NA	NA
	Caspofungin	≤0.008	0.06	0.06	0.06	100	NA	0
	Anidulafungin	≤0.015	0.12	0.06	0.12	100	NA	NA
	Flucytosine	≤0.06	0.5	≤0.06	0.12	100	NA	NA
C. glabrata (45)	Amphotericin B	≤0.12	1	0.5	1	NA	NA	NA
	Fluconazole	4	> 256	16	32	28.9	66.7	4.4
	Voriconazole	0.06	4	0.25	0.5	93.3	4.4	2.2
	Itraconazole	0.25	> 16	1	1	0	44.4	55.6
	Posaconazole	0.5	> 8	1	2	NA	NA	NA
	Micafungin	≤0.008	2	0.008	0.008	100	NA	NA
	Caspofungin	0.06	> 8	0.12	0.12	97.8	NA	2.2
	Anidulafungin	≤0.015	2	0.12	0.12	100	NA	NA
	Flucytosine	≤0.06	≤0.06	≤0.06	≤0.06	100	NA	NA
C. tropicalis (43)	Amphotericin B	0.25	1	0.5	1	NA	NA	NA
	Fluconazole	0.25	> 256	2	64	88.4	0	11.6
	Voriconazole	0.015	> 8	0.12	0.25	90.7	7	2.3
	Itraconazole	0.06	> 16	0.25	0.5	27.9	69.8	2.3
	Posaconazole	0.5	> 8	0.25	0.5	NA	NA	NA
	Micafungin	≤0.008	0.06	0.015	0.015	100	NA	NA
	Caspofungin	0.03	0.25	0.06	0.12	100	NA	0
	Anidulafungin	0.06	0.25	0.12	0.25	100	NA	NA
	Flucytosine	≤0.06	0.25	≤0.06	≤0.06	100	NA	NA
C. parapsilosis (18)	Amphotericin B	≤0.12	0.5	0.5	0.5	NA	NA	NA
	Fluconazole	0.5	16	0.5	4	94.4	5.6	0
	Voriconazole	≤0.008	0.12	≤0.008	0.12	100	0	0
	Itraconazole	≤0.015	0.25	0.06	0.12	94.4	5.6	0
	Posaconazole	0.015	0.25	0.03	0.12	NA	NA	NA
	Micafungin	0.25	1	0.5	1	100	NA	NA
	Caspofungin	0.25	1	0.5	1	100	NA	0
	Anidulafungin	0.5	2	1	2	100	NA	NA
	Flucytosine	≤0.06	0.5	0.12	0.275	100	NA	NA
C. krusei (2)	Amphotericin B	0.5	1			NA	NA	NA
	Fluconazole	32	64					50
	Voriconazole	0.25	0.25					0
	Itraconazole	0.25	0.5					0
	Posaconazole	0.25	0.25			NA	NA	NA
	Micafungin	0.06	0.06				NA	NA
	Caspofungin	0.25	0.25				NA	0
	Anidulafungin	0.12	0.12				NA	NA
	Flucytosine	16	16				NA	NA
C. famata (2)	Amphotericin B	0.5	8			NA	NA	NA
	Fluconazole	2	> 256					50
	Voriconazole	0.015	> 8					50
	Itraconazole	0.25	> 16					50
	Posaconazole	0.12	> 8			NA	NA	NA
	Micafungin	0.06	0.06				NA	NA
	Caspofungin	0.25	0.25				NA	0

(*continued on next page*)

Table 3 (*continued*)

Species (No.)	Antifungal agent	Range		MIC (μg/mL) 50%	90%	% S	SDD	(%) R or Non-S
	Anidulafungin	0.12	0.25				NA	NA
	Flucytosine	≤0.06	≤0.06				NA	NA
C. guilliermondii (2)	Amphotericin B	0.25	0.5			NA	NA	NA
	Fluconazole	4	8					0
	Voriconazole	0.06	0.12					0
	Itraconazole	0.5	0.5					0
	Posaconazole	0.25	0.5			NA	NA	NA
	Micafungin	0.05	> 8				NA	NA
	Caspofungin	0.25	> 8				NA	50
	Anidulafungin	1	> 8				NA	NA
	Flucytosine	≤0.06	0.25				NA	NA

I = intermediate; NA = not applicable; Non-S = nonsusceptible; R = resistant; S = susceptible; SDD = dose-dependent susceptible. Breakpoint (μg/mL) for antifungal agents. Fluconazole: Susc: ≤ 8; SDD 16–32; R ≥ 64 Voriconazole: Susc: ≤ 1; SDD 2; R ≥ 4 Itraconazole: Susc: ≤ 0.125; SDD 0.25–0.5; R ≥ 1 Posaconazole: Susc: None Micafungin: Susc: ≤ 2; NS: >2 Caspofungin: Susc: ≤ 2; NS: >2 Anidulafugin: Susc: ≤ 2; NS: >2 Flucytosine: Susc: ≤ 4; I: 8–16; R ≥ 32.

A major finding in the current study is the changing distribution of *Candida* species among candidemia. In our institute, although *C. albicans* is still the most common pathogen, the proportion declined from 64.8% to 43.6%. This is mainly due to the increase in cases of *C. glabrata* candidemia from one (1.1%) during 2001–2003 to 47 (21.6%) during 2009–2012 ($p < 0.001$). The proportions of other non-*C. albicans* species, e.g., *C. tropicalis* and *C. parapsilosis*, are also elevated slightly. The increasing trend of non-*C. albicans* candidemia is consistent with other studies.[4,14–20] The reasons for the trend, however, are less conclusive. Previous studies had identified that when compared with *C. albicans* infections, fluconazole use,[21] antibiotic use,[22–24] malignancy,[22,24] abdominal surgery,[21,24] and age > 60 years[24] were independent predictors for *C. glabrata* infections. In addition to older age, we had found that a higher proportion of female patients was associated with *C. glabrata* candidemia. Some other studies, such as Horn et al[5] ($n = 274$ of 525, 52.2%), Tapia et al[21] ($n = 37$ of 68, 54.4%), Lee et al[25] ($n = 77$ of 144, 54%), and Malani et al[26] ($n = 49$ of 94, 52%), had shown a higher proportion of female patients with *C. glabrata* candidemia. In another study,[27] the authors found that *C. glabrata* is the most isolated *Candida* species from the vagina in female patients with type 2 diabetes, and the study by Yang et al[18] study revealed that urinary tract infection was the most common source for *C. glabrata* candidemia (71.9%). In the current study, we found that women had a higher proportion of urinary tract infection as the primary focus in comparison with men (12 of 24, 50% vs. 2/17, 11.8%, $p = 0.018$). We assumed that the anatomy and microbiota of genitourinary tract of women led to the predominance of *C. glabrata* candidemia in female patients.

In addition, environmental factors may have contributed to the increasing trend of non-*C. albicans* candidemia. The association of fluconazole use and increased incidence of *C. glabrata* candidemia had been reported in several studies.[21,22,24] Although not significant, our current study showed that higher proportions of patients with *C. tropicalis* or *C. glabrata* candidemia were exposed to fluconazole before the onset of candidemia (22.7% and 19.5%, respectively). The interactions among host factors, environment factors, and invading *Candida* species may be complex and, as shown in our study, may evolve with time. More meticulous studies are necessary to clarify these interactions.

In the current study, the overall resistance rate of the clinical *Candida* isolates to fluconazole was 4.95% and all

Table 4 Comparison of characteristics of patients with candidemia of susceptible and reduced-susceptible strains

	Fluconazole reduced-susceptible strain		Univariate	Multivariate		
	No ($n = 137$)	Yes ($n = 32$)	*p*	*p*	Odds ratio	95% confidence interval
Sex (male)	94 (68.6)	14 (43.8)	0.008	0.007	3.084	1.367–6.959
Broad-spectrum antibiotic exposure	108 (78.8)	30 (93.8)	0.050	0.129	—	—
Fluconazole exposure before candidemia	18 (13.1)	10 (31.3)	0.013	0.010	3.403	1.334–8.682

Table 5 Factors associated with 30-day crude mortality during candidemia episodes

	30-d mortality		Univariate	Multivariate		
	Survived ($n = 97$)	Died ($n = 112$)	*p*	*p*	OR	95% CI
APACHE score	16 ± 7	22 ± 9	< 0.001	< 0.001	1.134	1.084–1.188
Candidemia to antifungal therapy (d) ($n = 170$)	0.87 ± 3.949 ($n = 86$)	0.08 ± 3.915 ($n = 84$)	0.193	—	—	—
Malignancy	39 (39.8)	59 (60.2)	0.072	0.010	2.496	1.240–5.024
Cirrhosis	4 (22.2)	14 (77.8)	0.031	0.005	7.598	1.858–31.066
CVC	74 (43.3)	97 (56.7)	0.054	—	—	—
Parenteral nutrition	24 (35.3)	44 (64.7)	0.025	0.001	3.515	1.664–7.427
Use of steroid	24 (34.3)	46 (65.7)	0.013	—	—	—
Neutropenia	5 (21.7)	18 (78.3)	0.012	0.034	4.203	1.115–15.842
Candida species			0.011	0.017	—	—
C. albicans	39 (43.3)	51 (56.7)	Reference			
C. tropicalis	15 (34.1)	29 (65.9)	0.065	0.945	0.968	0.382–2.453
C. glabrata	24 (58.5)	17 (41.5)	0.082	0.040	0.389	0.158–0.958
C. parapsilosis	12 (80)	3 (20)	0.007	0.005	0.112	0.024–0.522
Other[a]	7 (36.8)	12 (63.2)	0.380	0.678	1.278	0.402–4.069
Candidemia by reduced-fuconazole susceptiblity strain			0.398	—	—	—
No	68 (49.6)	69 (50.4)	Reference	—	—	—
Yes	12 (37.5)	20 (62.5)	0.272	—	—	—
Other[b]	17 (42.5)	23 (57.5)	0.581	—	—	—

[a] Including episodes with mixed candidemia or candidemia by *Candida* species other than *C. albicans*, *C. tropicalis*, *C. glabrata*, *C. parapsilosis*.
[b] Patients whose isolates were both susceptible and reduced-susceptible were not available or failed susceptibilty testing.
CI = confidence interval; CVC = central venous catheter; OR = odds ratio.

the *C. albicans* isolates were susceptible to fluconazole, supporting the primary use of fluconazole in our institute. There were several previous studies in Taiwan showing low fluconazole resistance among all *Candida* fungemic isolates, including 1.9% in the report by Yang et al,[18] 4.4% in the study by Hsueh et al,[19] 0.04% in the study by Tsai et al,[16] and 3.8% in the study by Chen et al.[20] However, the increasing trend of fluconazole resistant non-*C. albicans* candidal infection had been cause for concern. As shown in our study, 72.1% (30 of 45) of *C. glabrata* isolates and 11.6% of *C. tropicalis* had reduced fluconazole susceptibility (e.g., dose-dependent susceptible and resistant). The resistance rate of *C. glabrata* in our study is compatible with the finding in the study by Ruan et al,[28] which showed that the rate of fluconazole dose-dependent susceptibility of *C. glabrata* isolates was higher (73%) in the later year (2003 vs. 2007, $p = 0.003$) but no resistance to fluconazole among *C. tropicalis* isolates was detected in their study. The resistance of *C. tropicalis* in our study is higher than that in the study by Chen et al, which showed 5.9% (10 of 171) of fluconazole resistance in *C. tropicalis* from 1999 to 2006. However, the interpretation of differences of drug resistance in *Candida* isolates among different studies and study periods should be cautious because the drug resistance may be affected by the methods used for susceptibility testing. The study by Lombardi et al[29] showed that the agreement between National Committee for Clinical Laboratory Standards (NCCLS) and Yeast One ranged from 95% (ketoconazole and itraconazole) to 100% (amphotericin B and flucytosine), whereas the agreement between E-test and YeastOne ranged from 72.5% (fluconazole) to 100% (amphotericin B and flucytosine).

The risk factors for acquiring candidemia due to drug-resistant strains had been studied. In a report by Tumbarello et al,[30] they found that patients with prior fluconazole use, diabetes, and a central venous catheter were more likely to develop fungemia due to a less susceptible isolate. In our study, female sex and fluconazole exposure are the only two independent risks for acquiring candidemia due to fluconazole-resistant strains. The female sex as a risk factor is likely due to the predominance of female patients with *C. glabrata* candidemia in our study, which showed that *C. glabrata* is the major *Candida* species with reduced fluconazole susceptibility.

Several studies have described risk factors for mortality of candidemia,[16,26,31] including increasing age, greater APACHE II score, use of immunosuppressive therapy, longer duration of prior antibiotic use, retention of central venous catheter, stay in ICU, malignancy, neutropenia, renal insufficiency, and receipt of parenteral nutrition. We identified that APACHE II score, malignancy, liver cirrhosis, use of parenteral nutrition, neutropenia, and the invading *Candida* species were independent risk factors for 30-day crude mortality. In our study, patients with *C. glabrata* or *C. parapsilosis* candidemia had favorable outcomes, with 30-day crude mortality of 41.5% for *C. glabrata* and 20% for *C. parapsilosis* (odds ratio = 0.389, $p = 0.040$ and odds ratio = 0.112, $p = 0.005$, respectively, when compared with the mortality for *C. albicans*). Previous studies had reported various mortality rates among different *Candida* spp.[20,22,31,32] The mortality rates range from 41.1% to 52% for *C. albicans*, 41% to 58% for *C. tropicalis*, 31% to 60% for *C. glabrata*, and 22.9% to 26% for C. *parapsilosis*. The variation of mortality rates in different studies may be owing to heterogeneous underlying conditions that influence the prognosis of patients.

Some reported treatment failures associated with infections of fluconazole resistant candidal strains. The study by Chen et al[33] showed that candidemia attributable mortality was correlated with reduced fluconazole susceptibility. However, as in the report by Lee et al,[25] we also failed to demonstrate the association between drug susceptibility of *Candida* species and clinical outcome. The 30-day mortality rate of patients infected with reduced fluconazole susceptible strains was similar to that of patients infected with fluconazole-susceptible strains (50.4% vs. 62.5%; $p = 0.272$). Prompt antifungal therapy might influence the outcome as noted in the study by Chen et al.[14] However, the difference of time from onset of candidemia to effective antifungal therapy between surviving and fatal cases was not significant in our study. More studies with large sample size and adjusting all the factors influencing mortality are necessary to make a conclusion about the effects of drug susceptibility of *Candida* species and timely and adequate use of antifungal agents on the outcome of patients.

The major limitation of this study is its retrospective design with high heterogenicity in characteristics among patients and small sample sizes when attempting subgroup analysis. However, our findings still offer valuable points in the management of patients with candidemia. In conclusion, the results of this study showed that although *C. albicans* remained the most isolated species, the proportion of non-*C. albicans* candidemia, especially *C. glabrata*, was increasing. The drug resistance of clinical candidal isolates to the primary agents, e.g., fluconazole and echinocandins, remained low but candidemias due to reduced fluconazole susceptibility non-*C. albicans Candida* species, especially *C. glabrata*, were increasing. The prognosis of patients with candidemia was still poor. Further studies are necessary with focus on prompt identification of patients at risk for candidemia due to resistant strains and the effect of appropriate antifungal therapy on mortality.

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