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A Multicenter Study of Antifungal Use and Species Distribution and Antifungal Susceptibilities of *Candida* Isolates in South Korea

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Background: Candidiasis control should include monitoring the epidemiology and resistance to various antifungal agents. In this study, the researchers investigated the *Candida* species recovered from clinical specimens at particular geographic areas or hospitals.

Objective: The present study is geared toward the evaluation of antifungal drug usage at Korean hospitals in 2016. It is also essential that species distribution and antifungal susceptibilities of *Candida* isolates should be looked into to provide important data that can help devise therapeutic strategies to control the disease.

Methods: Systemic antifungal agent usage over a one-year period was investigated at 10 Korean hospitals. Identification and antifungal susceptibility tests were performed on clinical isolates of the *Candida* species, which were collected over a three-month period.

Results: The total antifungal usage in each hospital ranged from 7.7 to 158.9 defined daily doses (DDDs) per 1,000 patient days. Fluconazole was most commonly used (37.1%), followed by amphotericin B (30.6%), itraconazole (9.7%), echinocandins (8.8%), voriconazole (7.5%), and posaconazole (6.3%), respectively. Among 274 *Candida* isolates, *C. albicans* was the most frequently recovered (51.1%), followed by *C. glabrata* (15.7%), *C. tropicalis* (15.0%), and *C. parapsilosis* (13.5%), respectively. Through the application of either species-specific clinical

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breakpoints or epidemiological cutoff values to *Candida* isolates, the non-susceptibility rates to fluconazole, voriconazole, amphotericin B, and micafungin were found in 20.7%, 5.6%, 0%, and 0% of isolates, respectively.

Conclusion: This nationwide multicenter study showed that total antifungal use varied considerably according to each hospital. Non-susceptibility to fluconazole should be further monitored, considering the drug's frequent use in Korea.

Key Words: Antifungal susceptibility, Antifungal usage, *Candida*, Multicenter surveillance

INTRODUCTION

Candida species are commonly found as normal flora, but they can cause a variety of opportunistic infections such as oropharyngitis, esophagitis, vulvovaginitis, urinary tract infection, meningitis, endophthalmitis, endocarditis, peritonitis, osteomyelitis, and candidemia[1,2]. Candidiasis control should include monitoring the *Candida* species' epidemiology and resistance to various antifungal agents[1,2]. Antifungal usage is associated with the distribution and antifungal susceptibility of *Candida* species[3-5]. According to previous multicenter studies conducted in South Korea since 2004[3,5,6], the pattern of antifungal drug usage, the rank order of occurrence, and antifungal susceptibilities of disease-causing *Candida* species varied across hospitals and different time periods. This multicenter study was performed in order to investigate the antifungal drug usage in Korean hospitals and the antifungal susceptibility of *Candida* species recovered from clinically relevant specimens.

MATERIALS AND METHODS

A prospective surveillance study was conducted in 10 Korean hospitals (A-J). Data was collected over a one-year period regarding the usage of systemic antifungal agents by patients admitted in a particular hospital. This was then determined by calculating the number of defined daily doses per 1,000 patient days (DDD/1,000 PD), as specified by the WHO ATC/DDD system (www.whocc.no/atcddd/) and DDD measurement methodology[7]. The non-duplicated clinical isolates of the *Candida* species, which were collected from clinically relevant specimens over a three-month period (from October 2016 to December 2016), were sent to Chonnam National University Hospital for species identification and antifungal susceptibility testing. The identification results of *Candida* species were obtained using the conventional laboratory tests of each hospital. After gathering the results, the final identification was performed using CHROMagar *Candida* (BBL; Becton Dickinson, Sparks, MD, USA), VITEK 2 system with a VITEK 2 YST card (bioMérieux, Marcy l'Étoile, France), MALDI Biotyper (software version 3.1, reference database version 4.0.0.1, Bruker Daltonics, Billerica, MA, USA), or sequencing, if further confirmation is needed[8]. The antifungal susceptibility test was performed to measure their resistance to fluconazole, voriconazole, amphotericin B, and micafungin, and it was done using the Clinical and Laboratory Standards Institute (CLSI) broth microdilution method M60[9]. The minimum inhibitory concentration (MIC) of each isolate was categorized by applying CLSI clinical breakpoints (CBPs) or epidemiological cutoff values (ECVs)[10,11].

RESULTS

The data in Table 1 suggest that the total antifungal use varied considerably in 10 selected hospitals, ranging from 7.7 to 158.9 DDD/1,000 PD, with fluconazole being the most commonly used agent (with an average of 18.6 DDD/1,000 PD, or 37.1% of total DDD/1,000 PD), followed by amphotericin B (15.3, 30.6%), itraconazole (4.9, 9.7%), voriconazole (3.8, 7.5%), posaconazole (3.1, 6.3%), caspofungin (2.6, 5.2%), micafungin (1.6, 3.2%), and anidulafungin (0.2, 0.4%), respectively. Table 2 summarizes the species identification and *in vitro* susceptibility to fluconazole, voriconazole, amphotericin B, and micafungin for 274 *Candida* isolates obtained from clinically relevant specimens over a three-month period. *C. albicans* (n=140, 51.1%) was the most commonly recovered, followed by 43 *C. glabrata* (15.7%), 41 *C. tropicalis* (15.0%), 37 *C. parapsilosis* (13.5%), 4 *C. krusei* (1.5%), 3 *C. guilliermondii* (1.1%), 2 *C. auris* (0.7%), 1 *C. fabianii* (0.4%), 1 *C. lusitaniae* (0.4%), 1 *C. magnoliae* (0.4%), and 1 *C. pelliculosa* (0.4%) isolates. All the isolates showed an MIC range of 0.125 to > 64 μg/mL for fluconazole, 0.03~8 μg/mL for voriconazole, 0.25~2 μg/mL for amphotericin B, and 0.015~

Table 1. Antifungal drug use in 10 Korean hospitals over a one-year period (2016)

Antifungals	Antifungal drug use by hospitals (defined daily dose/1,000 patient days)										Average	Total (%)
	A	B	C	D	E	F	G	H	I	J		
Azoles	115.3	37.1	46.5	27.1	11.2	21.8	13.3	12.4	12.9	6.2	30.4	303.9 (60.6)
Fluconazole	45.0	30.1	27.9	25.3	8.5	8.7	11.5	11.1	12.6	5.4	18.6	186.0 (37.1)
Itraconazole	40.8	0.6	2.4	0.6	1.1	2.5	0.0	0.4	0.0	0.4	4.9	48.6 (9.7)
Voriconazole	15.0	2.4	12.2	1.3	1.7	3.1	0.4	0.9	0.4	0.5	3.8	37.8 (7.5)
Posaconazole	14.5	4.1	4.1	0.0	0.0	7.5	1.3	0.0	0.0	0.0	3.1	31.4 (6.3)
Amphotericin B*	34.3	50.2	35.4	4.0	16.9	4.7	3.3	2.5	2.2	0.0	15.3	153.3 (30.6)
Echinocandins	9.3	13.1	9.1	1.4	2.6	2.6	3.1	0.9	0.4	1.5	4.4	44.0 (8.8)
Caspofungin	3.0	12.4	2.9	1.4	1.4	1.8	1.8	0.1	0.3	0.9	2.6	26.1 (5.2)
Micafungin	6.3	0.7	5.3	0.0	0.6	0.7	0.9	0.8	0.0	0.5	1.6	15.8 (3.2)
Anidulafungin	0.0	0.0	0.9	0.0	0.5	0.1	0.5	0.0	0.0	0.0	0.2	2.0 (0.4)
Total	158.9	100.4	91.0	32.5	30.6	29.1	19.7	15.8	15.5	7.7	50.1	501.2 (100)

*This category included the lipid formulations of amphotericin B

2 μg/mL for micafungin. Upon applying CBPs or ECVs to available *Candida* species, the resistance and non-susceptible rates for these species to fluconazole, voriconazole, amphotericin B, and micafungin were found to be 5.2% and 20.7%; 3.3% and 5.6%; 0.0% and 0.0%; and 0.0% and 0.0%, respectively.

DISCUSSION

This nationwide multicenter study aims to investigate the antifungal drug usage and the antifungal susceptibility of *Candida* species recovered from clinically relevant specimens. We found that the total antifungal use varied considerably according to each hospital. The average total usage of antifungals was 50.1 DDD/1,000 PD, which was similar to reports shown by previous Korean multicenter studies (63.2 DDD/1,000 PD in 2011 and 39.2 in 2005)[3,5]; however, the pattern of antifungal agent usage was different in each hospital. Fluconazole was the most commonly used agent in seven hospitals, while amphotericin B was the most commonly used in three hospitals, specifically hospitals B, C, and E. The average usages of fluconazole, itraconazole, voriconazole, amphotericin B, caspofungin, and micafungin showed a 0.6-, 0.4-, 0.8-, 1.2-, 1.6-, and 1.1-fold increase, respectively, compared with those found in the 2011 study[10]. Consistent with previous studies, fluconazole was still the most commonly used antifungal agent[5], but the overall echinocandin use has increased (average of 4.4 DDD/1,000 PD and 8.8% of total DDD/1,000 PD in this study; 3.1 DDD/1,000 PD and 4.9% in 2011; 0.5 DDD/1,000 PD and 1.2% in 2005)[3,5] as well. The use of fluconazole has been associated with an increased risk of candidemia caused by non-albicans *Candida* species due to their inherently higher level of resistance to certain anti-

Table 2. Antifungal susceptibilities of 274 clinical isolates of *Candida* species obtained from 10 Korean hospitals over three months

Antifungal agents and *Candida* species	Number of isolates	MIC (μg/mL)			Number (%) of isolates classified by CBPs or ECVs		
		Range	50%	90%	R	SDD or I	Total
Fluconazole							
C. albicans	140	0.125~2	0.25	0.5	0 (0.0)	0 (0.0)	0 (0.0)
C. glabrata	43	4~>64	8	32	3 (7.0)	40 (93.0)	43 (100)
C. tropicalis	41	0.125~8	0.25	1	1. (2.4)	0 (0.0)	1 (2.4)
C. parapsilosis	37	0.25~16	1	4	3 (8.1)	2 (5.4)	5 (13.5)
C. krusei	4	32~>64			4 (100)†	NA	4 (100)
C. guilliermondii	3	0.25~>64			1 (33.3)‡	NA	1 (33.3)
C. auris	2	>64			NA	NA	NA
C. lusitaniae	1	>64			1 (100)‡	NA	1 (100)
C. pelliculosa	1	>64			1 (100)‡	NA	1 (100)
Other *Candida* species*	2	2-64			NA	NA	NA
Total	274	0.125~>64			14 (5.2)	42 (15.6)	56 (20.7)
Voriconazole							
C. albicans	140	0.03~0.5	0.03	0.03	0 (0.0)	1 (0.7)	1 (0.7)
C. glabrata	43	0.03~4	0.5	0.5	3 (7.0)‡	NA	3 (7.0)
C. tropicalis	41	0.03~1	0.06	0.06	1 (2.4)	0 (0.0)	1 (2.4)
C. parapsilosis	37	0.03~0.5	0.06	0.125	0 (0.0)	3 (8.1)	3 (8.1)
C. krusei	4	0.5~4			1 (25.0)	2 (50.0)	3 (75.0)
C. guilliermondii	3	0.25~8			2 (66.7)‡	NA	2 (66.7)
C. auris	2	2			NA	NA	NA
C. lusitaniae	1	2			1 (100)‡	NA	1 (100)
C. pelliculosa	1	4			1 (100)‡	NA	1 (100)
Other *Candida* species	2	0.06~0.125			NA	NA	NA
Total	274	0.03~8			9 (3.3)	6 (2.2)	15 (5.6)
Amphotericin B							
C. albicans	140	0.25~2	0.5	0.5	0 (0.0)‡	NA	0 (0.0)
C. glabrata	43	0.25~1	1	1	0 (0.0)‡	NA	0 (0.0)
C. tropicalis	41	0.5~1	1	1	0 (0.0)‡	NA	0 (0.0)
C. parapsilosis	37	0.25-1	0.5	1	0 (0.0)‡	NA	0 (0.0)
C. krusei	4	1			0 (0.0)‡	NA	0 (0.0)
C. guilliermondii	3	0.25~0.5			0 (0.0)‡	NA	0 (0.0)
C. auris	2	0.5~1			NA	NA	NA
C. lusitaniae	1	0.5			0 (0.0)‡	NA	0 (0.0)

Table 2. Antifungal susceptibilities of 274 clinical isolates of *Candida* species obtained from 10 Korean hospitals over three months (Continued)

Antifungal agents and *Candida* species	Number of isolates	MIC (μg/mL)			Number (%) of isolates classified by CBPs or ECVs		
		Range	50%	90%	R	SDD or I	Total
C. pelliculosa	1	1			NA	NA	NA
Other *Candida* species*	2	0.25			NA	NA	NA
Total	274	0.25~2			0 (0.0)	NA	0 (0.0)
Micafungin							
C. albicans	140	0.015~0.06	0.015	0.015	0 (0.0)	0 (0.0)	0 (0.0)
C. glabrata	43	0.015~0.03	0.015	0.03	0 (0.0)	0 (0.0)	0 (0.0)
C. tropicalis	41	0.015~0.06	0.015	0.03	0 (0.0)	0 (0.0)	0 (0.0)
C. parapsilosis	37	0.015~2	1	1	0 (0.0)	0 (0.0)	0 (0.0)
C. krusei	4	0.015~0.25			0 (0.0)	0 (0.0)	0 (0.0)
C. guilliermondii	3	0.5~1			0 (0.0)	0 (0.0)	0 (0.0)
C. auris	2	0.015			NA	NA	NA
C. lusitaniae	1	0.06			0 (0.0)‡	NA	0 (0.0)
C. pelliculosa	1	0.015			NA	NA	NA
Other *Candida* species*	2	0.06~0.125			NA	NA	NA
Total	274	0.015-2			0 (0.0)	0 (0.0)	0 (0.0)

*Other *Candida* species included *C. fabianii* (one isolate) and *C. magnoliae* (one isolate)
†All *C. krusei* isolates are considered as resistant to fluconazole, irrespective of the MIC
‡The results were analyzed by ECVs[11]
Abbreviations: MIC, minimum inhibitory concentration; CBP, clinical breakpoint; ECV, epidemiological cutoff value; SDD, susceptible dose-dependent; I, intermediate; R, resistant; NA, not available

fungal agents[5]. In addition, the increasing trend in echinocandin use is important because it could pose a risk of emerging echinocandin-resistant *Candida* species[4].

The overall epidemiology of the *Candida* species agreed with the data of previous studies conducted in Korea[3,6]. More importantly, in the current study, *C. glabrata* was the most frequently found non-albicans *Candida* species, followed by *C. tropicalis* and *C. parapsilosis*, while the 2011 study found slightly different rates, where *C. parapsilosis* (17.8%) was the most common non-albicans *Candida* species, followed by *C. glabrata* (14.4%) and *C. tropicalis* (12.7%)[6]. When applying CBPs, 11 of 265 *Candida* isolates were found to be resistant to fluconazole, specifically three *C. glabrata*, one *C. tropicalis*, three *C. parapsilosis*, and four *C. krusei* isolates. The overall resistance rate was not much higher than that of other geographic regions[12,13]. The fluconazole resistance and non-susceptibility rates, however, showed a 2.0- and 1.3-fold increase, respectively, compared with the data provided by previous Korean studies (2.6% and 16.4% in 2011, respectively)[5]. Moreover, among nine isolates of uncommon *Candida* species, five showed fluconazole MICs of > 64 μg/mL (two *C. auris*, one *C. guilliermondii*, one *C. lusitaniae*, and one *C. pelliculosa* isolates). Recently, *C. auris* has been highlighted to be less susceptible to fluconazole compared with the other *Candida* species, and it was reported to have developed a high-level resistance to fluconazole like that of *C. glabrata*[14]. In addition, *C. auris* was reported to be misidentified as *C. haemulonii* by the conventional identification method[8,14]. In the present study, two *C. auris* isolates were identified as *C. haemulonii* using conventional laboratory tests conducted in each particular hospital, which required sequencing to confirm the identification. Two candidemia cases with echinocandin-resistant *Candida* isolates (*C. albicans* and *C. glabrata*) were reported in South Korea[15]; however, the researchers found no *Candida* isolate that was resistant to micafungin, confirming the low resistance rates of echinocandins in South Korea when compared with those of the traits of micafungin that were documented in the previous study of other geographic regions, such as 0.4% in North America, 0.5% in Europe, 0% in Latin America, and 0.3% in the Asia Pacific region[12].

CONCLUSION

We investigated the nationwide antifungal use, species distribution, and antifungal susceptibility in South Korea. The overall antifungal usage was similar with the established previous usage, but the use of echinocandins has increased, which might affect echinocandin non-susceptibility rates in *Candida* species. The *Candida* species distribution was similar as it was in the past, but the rank order and frequency of non-albicans *Candida* species was changing. The overall resistance rates of *Candida* species were still low, but fluconazole resistance and non-susceptibility rates have increased in South Korea. Although echinocandin resistance is still uncommon in Korea, we should be aware of its emergence in the near future, considering the increment of echinocandin use.

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CONFLICT OF INTEREST

In relation to this article, we declare that there is no conflict of interest.

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REFERENCES

1. Pfaller MA, Diekema DJ. Epidemiology of invasive candidiasis: a persistent public health problem. Clin Microbiol Rev 2007;20:133-163
2. Pappas PG, Kauffman CA, Andes DR, Clancy CJ, Marr KA, Ostrosky-Zeichner L, et al. Clinical practice guideline for the management of candidiasis: 2016 update by the Infectious Diseases Society of America. Clin Infect Dis 2016;62:e1-50
3. Kim SH, Shin JH, Kim EC, Lee K, Kim MN, Lee WG, et al. The relationship between antifungal usage and antifungal susceptibility in clinical isolates of *Candida*: a multicenter Korean study. Med Mycol 2009;47:296-304
4. Fournier P, Schwebel C, Maubon D, Vesin A, Lebeau B, Foroni L, et al. Antifungal use influences *Candida* species distribution and susceptibility in the intensive care unit. J Antimicrob Chemother 2011;66:2880-2886
5. Won EJ, Shin JH, Choi MJ, Lee WG, Park YJ, Uh Y, et al. Antifungal susceptibilities of bloodstream isolates of *Candida* species from nine hospitals in Korea: application of new antifungal breakpoints and relationship to antifungal usage. PLoS One 2015;10:e0118770
6. Won EJ, Shin JH, Lee WK, Koo SH, Kim SY, Park YJ, et al. Distribution of yeast and mold species isolated from clinical specimens at 12 hospitals in Korea during 2011. Ann Clin Microbiol 2013;16:92-100
7. Merlo J, Wessling A, Melander A. Comparison of dose standard units for drug utilisation studies. Eur J Clin Pharmacol 1996;50:27-30
8. Kim TH, Kweon OJ, Kim HR, Lee MK. Identification of uncommon *Candida* species using commercial identification systems. J Microbiol Biotechnol 2016;26:2206-2213
9. Clinical and Laboratory Standards Institute. Performance standards for antifungal susceptibility testing of yeasts. 1st ed. CLSI supplement M60. Wayne, PA: Clinical and Laboratory Standards Institute; 2017
10. Clinical and Laboratory Standards Institute. Reference

method for broth dilution antifungal susceptibility testing of yeasts. Fourth informational supplement, M27-S4. Wayne, PA: Clinical and Laboratory Standards Institute; 2012

11. Pfaller MA, Diekema DJ. Progress in antifungal susceptibility testing of *Candida* spp. by use of Clinical and Laboratory Standards Institute broth microdilution methods, 2010 to 2012. J Clin Microbiol 2012;50:2846-2856
12. Pfaller MA, Messer SA, Woosley LN, Jones RN, Castanheira M. Echinocandin and triazole antifungal susceptibility profiles for clinical opportunistic yeast and mold isolates collected from 2010 to 2011: application of new CLSI clinical breakpoints and epidemiological cutoff values for characterization of geographic and temporal trends of antifungal resistance. J Clin Microbiol 2013;51:2571-2581
13. Tan TY, Hsu LY, Alejandria MM, Chaiwarith R, Chinniah T, Chayakulkeeree M, et al. Antifungal susceptibility of invasive *Candida* bloodstream isolates from the Asia-Pacific region. Med Mycol 2016;54:471-477
14. Lee WG, Shin JH, Uh Y, Kang MG, Kim SH, Park KH, et al. First three reported cases of nosocomial fungemia caused by *Candida auris*. J Clin Microbiol 2011;49: 3139-3142
15. Park MS, Park JE, Song DJ, Huh HJ, Park S, Kang CI, et al. First case of echinocandin-resistant *Candida albicans* in Korea. Ann Lab Med 2017;37:556-558