RESEARCH

ASSESSMENT OF SPECIES DISTRIBUTION AND ANTIFUNGAL SUSCEPTIBILITY OF CANDIDA ISOLATES FROM CASES OF HEALTH-CARE ASSOCIATED INFECTIONS IN A TERTIARY CARE CENTRE IN CENTRAL KERALA

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Abstract

Background: To assess species distribution and antifungal susceptibility of Candida isolates from cases of health-care associated infections in a tertiary care centre in central Kerala. **Materials and Methods:** Candida isolates from cases of HCAI from medical intensive care unit were identified up to species level. Antifungal susceptibility testing of Candida isolate was done by Clinical and Laboratory Standard Institute (CLSI) broth microdilution (BMD) method. **Result:** Out of 160 patients, males comprised 90 and females 70. Out of 60 patients on central line, 12 developed catheter related blood stream infection (CR-BSI), out of 55 patients, 22 developed catheter associated urinary tract infection (CA- UTI) and out of 45 patients on central line, 18 developed ventilator associated pneumonia. In 12 cases of CR- BSI, *Klebsiella pneumoniae* and *C. krusei* was found in 4 cases each, *E. coli* in 2, *Pseudomonas aeruginosa* and *C. albicans* in 1 case each. In 22 cases of CA- UTI, *E. coli* was found in 10, *Klebsiella pneumoniae* in 6, *C. tropicalis* in 3, *Pseudomonas aeruginosa* in 2 and *C. krusei* in 1 case. In 18 cases of VAP, *Klebsiella pneumoniae* was seen in 10, *E. coli*, *Staphylococcus aureus* and *C. albicans* in 2 cases each and *Pseudomonas aeruginosa* and *C. krusei* in 1 case each. The difference was significant (P< 0.05). Fluconazole resistance was significantly high in NAC spp. compared to *C. albicans*. All *C. krusei* isolates were resistant to fluconazole. A total of 20% of *C. tropicalis* were resistant to fluconazole. Resistance to itraconazole, voriconazole and amphotericin B was not observed in *C. albicans*. Amphotericin B resistance was observed in 20% of *C. tropicalis*. **Conclusion:** Fluconazole resistance was significantly high in non-albicans *Candida* spp. compared to *C. albicans*. Predominance of non-albicans *Candida* spp. over *C. albicans* was noted.

INTRODUCTION

Candida species is a normal flora in the human oral cavity with high carriage rate of more than 70%.[1] Infections caused by *Candida* species are seen commonly in hospitalized patients. From the data obtained National Nosocomial Infections Surveillance (NNIS) system, U.S, out of the total fungal isolates associated with nosocomial infections, *Candida* species accounted for around 72%.[17] Factors predisposing to this high frequency of yeast infections include: immunosuppressive conditions associated with chemotherapy, and stem cell and solid organ transplantations; prolonged hospitalisation; vascular catheterizations; and prolonged administration of broad spectrum antibacterial agents. Cross contamination by hospital personnel may also account for increase in yeast infections in certain environments. Oral candidiasis is widely seen among hospitalised patients and is considered as a sign of impaired local or systemic defense mechanisms.[11,12]

Among various *Candida* species causing infections, *C. albicans* is regarded as the most prevalent one. But recently, non-albicans *Candida* (NAC) species are also emerging as pathogens.[2] But

there is difference in pathogenicity and antifungal susceptibility among different species.

In an ICU setting, *Candida* spp. accounts for nearly 10% of all bloodstream infections (BSI) and 25% of all urinary tract infections (UTIs).[3] In the United States, *Candida* spp. is 3rd or 4th common cause of health-care associated infections (HCAI), surpassing all Gram-negative bacilli. Nosocomial *Candida* infections are usually treatment resistant and are associated with prolonged hospitalization and increased healthcare-cost.[4]

The epidemiology of IC has significantly evolved in recent years. A progressive shift in the most common cause of infections from *Candida albicans* to non-albicans *Candida* spp. has been observed globally.[5] Antifungal resistance is also an increasingly difficult challenge for the implementation of effective empirical and prophylactic strategies, with emerging species exhibiting resistance to multiple classes of antifungal agents.[6,7] Considering this, we performed present study to assess species distribution and antifungal susceptibility of *Candida* isolates from cases of health-care associated infections.

MATERIALS AND METHODS

After considering the utility of the study and obtaining approval from ethical review committee of the institute, we performed the present study in the Department of Microbiology, P K Das Institute of Medical Sciences. Candida isolates from 52 cases of healthcare associated infections (HCAI) from in-patients of a tertiary care centre during January 2021 to December 2021 were enrolled. The demographical and clinical features of the patients suspected for HCAI were recorded.

Candida isolates were identified upto species by methods like KOH mount, Culture on Sabouraud's Dextrose Agar, Candida CHROM agar & Corn Meal Agar and by Germ Tube test. Antifungal susceptibility testing of *Candida* isolate was done by Clinical and Laboratory Standard Institute (CLSI) broth microdilution (BMD) method. The minimum inhibitory concentration (MIC) of *Candida* spp was detected for antifungal drugs like Amphotericin B, Fluconazole, Itraconazole and Voriconazole. MIC values were determined as the lowest concentration of antifungal drug that showed complete inhibition or a significant diminution (≥50% inhibition) of growth compared to the growth of control strain. *Candida krusei* ATCC 6258 and *Candida parapsilosis* ATCC 22019 were used as control strains. The results were compiled and subjected for statistical analysis using Mann Whitney U test. P value less than 0.05 was set significant.

RESULTS

Table 1: Patients distribution

Total- 160		
Gender	**Males**	**Females**
Number	90	70

Out of 160 patients, males comprised 90 and females 70 [Table 1].

Table 2: Health-care associated infections

Type of HCAI	**Total patients of medical device**	**Patients developing infection**	**P value**
CR- BSI	60	12	0.05
CA- UTI	55	22	
VAP	45	18	

Out of 60 patients on central line, 12 developed catheter related blood stream infection (CR- BSI), out of 55 patients, 22 developed catheter associated urinary tract infection (CA- UTI) and out of 45 patients on central line, 18 developed ventilator associated pneumonia. The difference was significant (P< 0.05) [Table 2].

Table 3: Pathogens isolated from health-care associated infection

Pathogens	**CR-BSI**	**CA-UTI**	**VAP**	**P value**
Staphylococcus aureus	0	0	2	0.05
Klebsiella pneumoniae	4	6	10	0.01
E. coli	2	10	2	0.02
Pseudomonas aeruginosa	1	2	1	0.05
C. albicans	1	0	2	0.12
C. krusei	4	1	1	1
C. tropicalis	0	3	0	0.01
Total	12	22	18	

In 12 cases of CR- BSI, *Klebsiella pneumoniae* and *C. krusei* in was found 4 cases each, *E. coli* in 2, *Pseudomonas aeruginosa* and *C. albicans* in 1 case each. In 22 cases of CA- UTI**,** *E. coli* was found in 10, *Klebsiella pneumoniae* in 6, *C. tropicalis* in 3, *Pseudomonas aeruginosa* in 2 and *C. krusei* in 1 case. In 18 cases of VAP, *Klebsiella pneumoniae* was seen in 10, *E. coli*, *Staphylococcus aureus* and *C. albicans* in 2 cases each and *Pseudomonas aeruginosa* and *C. krusei* in 1 case each. The difference was significant (P< 0.05) [Table 3, Figure 1].

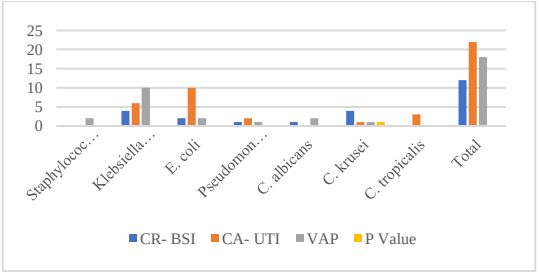

Figure 1: Pathogens isolated from health-care associated infection

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Table 4: Antifungal susceptibility profile of *Candida* spp

Candida spp	Antifungal agent	Range (µg/ml)	S (%)
C. albicans (3)	itraconazole	0.04-16	100%
	fluconazole	0.13-254	72%
	voriconazole	0.08-17	100%
	Amphotericin B	0.13-9	100%
C. krusei (6)	itraconazole	0.125-2	100%
	fluconazole	4-62	25%
	voriconazole	0.015-5	-
	Amphotericin B	0.25-4	-
C. tropicalis (3)	itraconazole	0.015-16	90%
	fluconazole	0.13-124	85%
	voriconazole	0.08-17	90%
	Amphotericin B	0.12-4	80%

Fluconazole resistance was significantly high in NAC spp. compared to *C. albicans*. All *C. krusei* isolates were resistant to fluconazole. A total of 20% of *C. tropicalis* were resistant to fluconazole. Resistance to itraconazole, voriconazole and amphotericin B was not observed in *C. albicans.* Amphotericin B resistance was observed in 20% of *C. tropicalis* [Table 4].

DISCUSSION

In the ECDC point prevalence survey of healthcare-associated infections and antimicrobial use in European acute care hospitals 2011–2012, *Candida* spp. was the fifth most common pathogen associated with bloodstream infections, isolated in 7.4% of all documented cases.[18] While *C. albicans* remains the predominant cause of invasive candidiasis, there has been a shift towards an increasing proportion of non-albicans Candida species such as *C. glabrata* in recent years.[19,20]The present study was conducted to assess the species distribution and antifungal susceptibility of *Candida* isolates from cases of health-care associated infections.

Out of 160 patients, males comprised 90 and females 70. Out of 60 patients on central line, 12 developed catheter related blood stream infection (CR- BSI), out of 55 patients, 22 developed catheter associated urinary tract infection (CA- UTI) and out of 45 patients on central line, 18 developed ventilator associated pneumonia. Among, the candida species isolated, majority was non albicans *Candida* followed by *Candida albicans.* Shafi *et al*,[13] evaluated pattern of various *Candida* species among hospitalised patients with oral candidiasis, to detect the antifungal resistance among *Candida* and to assess the possible risk factors associated with those patients. Out of 300 patients screened, oral thrush material was collected from 36 patients having oral candidiasis. *Candida* spp. were isolated and identified. Antifungal susceptibility test was performed by disk diffusion method. *Candida albicans* was the most frequently isolated species (64%). Highest resistance was seen with ketoconazole (18%). Except one *C. tropicalis*, all the isolates were sensitive to amphotericin B. All the patients were on broad spectrum antibiotic treatment. Diabetes mellitus was seen in 50 % of the patients. Other predisposing factors include tuberculosis, COPD, cancer and steroid treatment.

In 12 cases of CR- BSI, *Klebsiella pneumoniae* and *C.* krusei was found in 4 cases each, *E. coli* in 2, *Pseudomonas aeruginosa* and *C. albicans* in 1 case each. In 22 cases of CA- UTI, *E. coli* was found in 10, *Klebsiella pneumoniae* in 6, *C. tropicalis* in 3, *Pseudomonas aeruginosa* in 2 and *C. krusei* in 1 case. In 18 cases of VAP, *Klebsiella pneumoniae* was seen in 10, *E. coli*, *Staphylococcus aureus* and *C. albicans* in 2 cases each and *Pseudomonas aeruginosa* and *C. krusei* in 1 case each. Kamaljeet *et al*,[14] assessed species distribution and antifungal susceptibility pattern of *Candida* isolates from cases of HCAI. *Candida* isolates from cases of HCAI from medical intensive care unit were identified and antifungal susceptibility testing of *Candida* isolates were done. The rate of CA-UTI due *Candida* spp. was 0.3 per 1000 catheter associated days. The rate of catheter related *Candida* BSI was 0.9 per 1000 catheter associated days. Predominance of non albicans *Candida* spp. over *C. albicans* was noted. Fluconazole resistance was significantly high in NAC spp. compared to *C. albicans.* All *C. krusei* isolates were resistant to fluconazole. A total of 20% of *C. tropicalis* were resistant to fluconazole. Resistance to itraconazole, voriconazole and amphotericin B was not observed in *C. albicans*. Amphotericin B resistance was observed in 20% of *C. tropicalis*. In a study by Coco *et al*,[15] it was suggested that coexistence of mixed species could aggravate the clinical condition that further complicate the treatment. Babin *et al*,[16] also reported the higher rate of fluconazole resistance among oral *C. albicans* isolates.

CONCLUSION

Predominance of non albicans *Candida* spp. over *C. albicans* was noted and Fluconazole resistance was significantly high in NAC spp. compared to *C. albicans*. Therefore, more effective infection control measures and strict treatment protocols are required for patients admitted in ICUs, especially those with factors predisposing to invasive fungal infections and high mortality.

REFERENCES

1. Mohamed S. Biofilm formation and antifungal susceptibility of Candida isolates from various clinical specimens. Br Microbiol Res J. 2013;3:590-601.
2. Pathak AK, Jain NR, Joshi R. Antibiogram of Candida species isolated from mono and multi-species oral candidal carriage using disk diffusion method. Saudi J Health Sci. 2012;1:132-8.
3. Vijaya D, Harsha TR, Nagaratnamma T. Candida speciation using chrom agar. J Clin Diagn Res. 2011;5:755-7.
4. Golia S, Reddy KM, Karjigi KS, Hittinahalli V. Speciation of Candida using chromogenic and cornmeal agar with determination of fluconazole sensitivity. Al Ameen J Med Sci. 2013;6:163-6.

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5. Shaheen MA, Taha M. Species identification of Candida isolates obtained from oral lesions of hospitalized and non hospitalized patients with oral candidiasis. Egyptian Dermatol Online J. 2006;2:1-13.
6. Okonkwo EC, Alo MN, Nworie O, Orji JO, Agah MV. Prevalence of oral Candida albicans infection in HIV seropositive patients in Abakaliki. Am J Life Sci. 2013;1:72-6.
7. Campos de Pinho Resende J, Franco GR, Rosa CA, Hahn RC, Hamdam JS. Phenotypic and genotypic identification of Candida spp. isolated from hospitalized patients. Rev Iberoam Micol. 2004;21(1):24-8.
8. Gahlot R, Nigam C, Kumar V, Yadav G, Anupurba S. Catheter-related bloodstream infections. Int J Crit Illn Inj Sci. 2014;4(2):162–7.
9. Fisher J. Candida urinary tract infections-epidemiology, pathogenesis, diagnosis, and treatment: executive summary. Clin Infect Dis. 2011;52(6):429–32.
10. Lundstrom T, Sobel J. Nosocomial candiduria: A review. Clin Infect Dis. 2001;32(11):1602–7.
11. Jain M, Dogra V, Mishra B, Thakur A, Loomba S, Bhargava A, et al. Candiduria in catheterized intensive care unit patients: emerging microbiological trends. Indian J Pathol Microbiol. 2011;54(3):552–5.
12. Krcmery V, Barnes AJ. Non-albicans Candida spp. causing fungaemia: Pathogenicity and antifungal resistance. J Hosp Infect. 2002;50(4):243–60.
13. Shafi FT, Padmaraj SR, Mullessery NP. Species distribution and antifungal susceptibility pattern of Candida causing oral candidiasis among hospitalized patients. Arch Med Health Sci. 2015;3:247-51.
14. Kamaljeet, Saxena N, Thalquotra M. Species distribution and antifungal susceptibility of Candida isolates from cases of health-care associated infections. Indian J Microbiol Res. 2022;9(2):95-98.
15. Coco BJ, Bagg J, Cross LJ, Jose A, Cross J, Ramage G. Mixed Candida and Candida glabrata population associated with the pathogenesis of dentures stomatitis. Oral Microbiol Immunol. 2008;23:377-83.
16. Babin D, Kotigadde S, Rao PS, Rao TV. Clinico-mycological profile of vaginal candidiasis in a tertiary care hospital in Kerala. Int J Res Biol Sci. 2013;3:55-9.
17. Jarvis WR. Epidemiology of nosocomial fungal infections, with emphasis on candida species. Clin Infect Dis 1995;20:1526-1530.
18. European Centre for Disease Prevention and Control. Point prevalence survey of healthcare-associatedd infections and antimicrobial use in European acute care hospitals 2011-2012. Stockholm: ECDC; 2013. Available from: http://ecdc.europa.eu/en/publications/publications/healthcare-associated-infectionsantimicrobial-use-pps.pdf.
19. Kullberg BJ, Arendrup MC. Invasive Candidiasis. N Engl J Med. 2015 Oct 08;373(15):1445-56.
20. Pfaller MA, Diekema DJ. Epidemiology of invasive candidiasis: a persistent public health problem. Clin Microbiol Rev. 2007 Jan;20(1):133-63.

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