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RESEARCH ARTICLE

CANDIDURIA: A DIAGNOSTIC AND THERAPEUTIC CHALLENGE

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ABSTRACT

Introduction: Inrecent years, Candida sp. has appeared as an emerging nosocomial pathogen causing Urinary Tract Infections. Candiduria is difficult to diagnose clinically as most often the patient is asymptomatic. Patients with candiduria often develop candidemia. Standard urine culture is less sensitive. Resistance to antifungal drugs have been more commonly encountered in *Candida* species isolated from urine. **Aim:** Hence, the present study was undertaken to detect the incidence of different Candida species isolated from urine and to study their antifungal drug susceptibility profile. **Material and Methods:** 630 urine samples were cultured on Sabouraud's Dextrose agar (SDA) with chloramphenicol. 55 Candida strains were detected by conventional tests. 05 Candida albicans and 07 Candida nonalbicans strains were further confirmed by Vitek2. Antifungal drug susceptibility profile was detected by disc diffusion method as per CLSI Guidelines. Minimum Inhibitory Concentration (MIC) of Fluconazole and Voriconazole was detected by E Test (bio Merieux). **Results:** *Candida tropicalis* was the commonest species (36.4%) followed by *Candida albicans* (29.1%), *Candida parapsilosis* (16.4%), *Candida glabrata* (7.3%) and *Candida pelliculosa* (7.3%). 22 (40%) Candida species were resistant to Fluconazole. Fluconazole MIC > 96 µg/ml was observed in 10 strains. **Conclusion:** Routinely all uine sample should be cultured on SDA for detection of Candiduria and antifungal drug sensitivity test should be done.

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INTRODUCTION

In recent years, Candida sp. has appeared as an emerging nosocomial pathogen causing Urinary Tract Infections (UTI). Risk factors for candiduria include indwelling catheters, prolonged ICU stay, female sex, anatomical abnormality of urinary tract, prolonged antibiotic therapy, Diabetes mellitus and other co-morbid conditions etc. Candiduria is difficult to diagnose clinically as most often the patients are asymptomatic. Even pus cells are not present in most of the cases (Achkar, 2010). Around 1-8% candiduric patients develop candidemia especially ICU patients. Standard routine urine culture is less sensitive and may lead to under estimation of Candiduria. Resistance to antifungal drugs have been more commonly encountered in *Candida albicans* and non-albicans species isolated from urine. Resistance may be intrinsic or acquired type. Hence, the present study was undertaken to detect the incidence of different Candida species isolated from urine and to study their antifungal drug susceptibility profile.

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MATERIALS AND METHODS

This short term cross-sectional experimental study was conducted in the department of Microbiology and was approved by Institutional Ethics Committee. Total 630 urine specimens, collected from Indoor Patient Departments (IPD) were cultured on Sabouraud's Dextrose agar (SDA) in the Department of Microbiology. Our Hospital is a tertiary care hospital in a rural set up. All urine samples were inoculated on Sabouraud's Dextrose agar (SDA) with chloramphenicol. The plates were incubated at 37 ^{o}C up to 48 hrs. Only the urine samples having pure growth of Candida species were included in the study. The colony morphology of each isolate was observed by Gram's staining, and if Gram positive budding yeast cells were observed then it was futher processed. 55 Candida strains were detected by conventional tests like microscopy, culture, Gram staining, Germ tube formation, colour on CHROM agar, chlamydospore formation on corn-meal agar, sugar assimilation test (Koneman *et al*, 2006). The nonalbicans species which were not identified by conventional tests were confirmed byVitek2 YST ID card. Hence, 5 *Candida albicans* and 7 Non-albicans species were further confirmed by Vitek 2 YST ID card.

Antifungal drug susceptibility was detected by disc diffusion method by using Mueller Hinton agar supplemented with 2% glucose and 0.5µg/ ml of methylene blue according to Clinical Laboratory Standard Institute (CLSI) Guidelines CLSI M44-A2 (CLSI, 2010). The reading was taken by measuring the growth inhibition zone. Results were expressed as susceptible (S), susceptible dose-dependant (SDD), and resistant (R) [3].

Minimum Inhibitory Concentration (MIC) of Fluconazole and Voriconazole were detected by E-test (bioMeriux) on SDA plate as per manufacturer's instruction. The Control strains used were C. albicans ATCC 90028 and C. tropicalis ATCC 750.

OBSERVATIONS AND RESULTS

A total 55 candida strains were isolated from 630 urine samples. Among them Non-albicans Candida species 39 (70.9%) were predominant compared to *Candida albicans* 16 (29.1%). The Non-albicans Candida species included *C. tropicalis* 20 (36.4%), *C. parapsilosis* 9 (16.4%), *C. glabrata* 4 (7.3%), *C. pelliculosa* 4 (7.23%), C. *krusei* 1 (1.3%) and *C. kefyr* 1 (1.3%).The most common species isolated was Candida tropicalis.

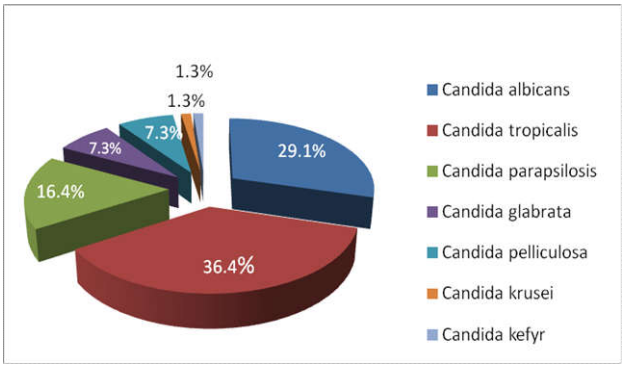


Fig 1. Incidence of different Candida sp. isolated (n=55)

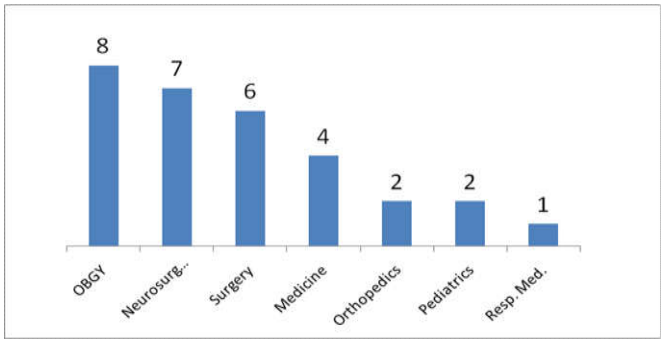


Fig. 2: Isolation of Candida strains from different hospital wards (n=30)

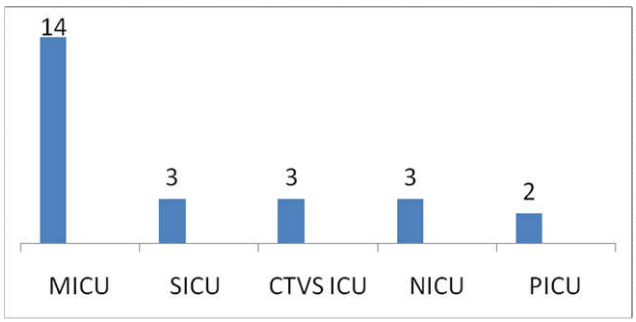


Fig. 3. Isolation of Candida strains from different ICUs (n=25)

Table 1. Isolation of Candida strains from male and female patients (n=55)

Male	Female	Ratio
24	31	1:1.3

Table 2. Isolation of Candida strains from catheterised and non catheterised patients (n=55)

Catheterized	Non-catheterized
46	09
83.7%	16.3%

Table 3. Antifungal sensitivity pattern to Voriconazole (VRC) & Fluconazole (FLC) by disc diffusion method

Sensitive to both VRC & FLC	24
Resistant to both VRC & FLC	16
Resistant to only VRC	0
Resistant to only FLC	4
SDD to FLC	6
Sensitive to only VRC	15

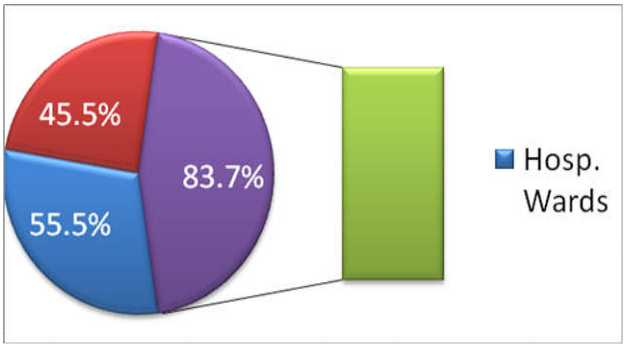
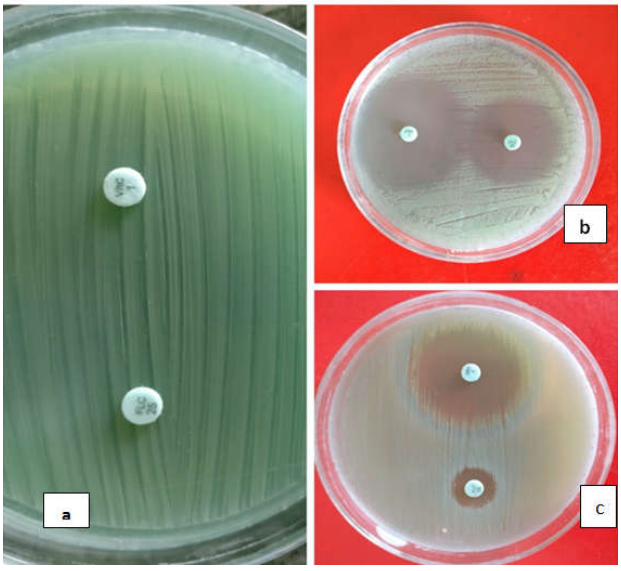


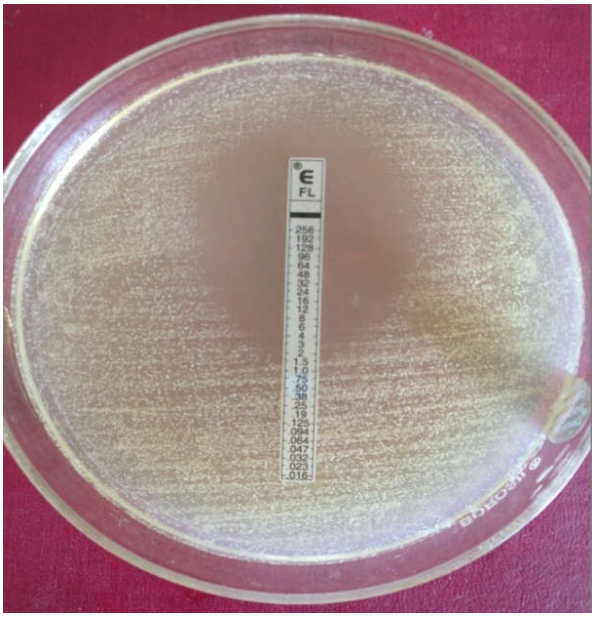
Fig. 4. Isolation of Candida species from patients with Health care Associated Infections (HAI)



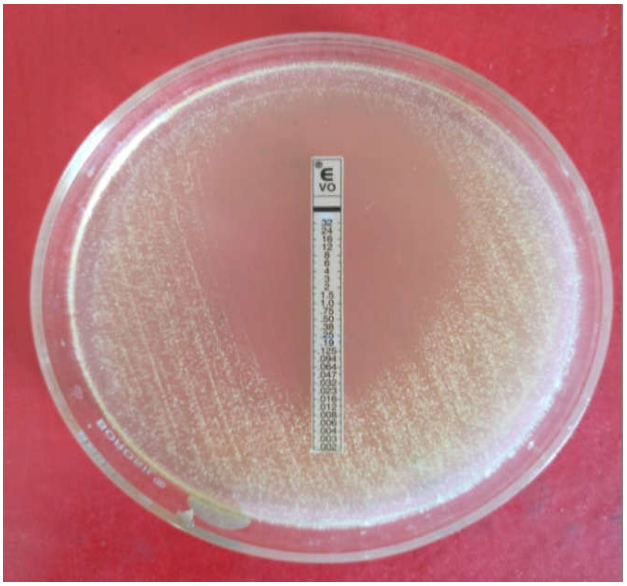
Photograph 1 Shows Candida strain

a-Resistant to both Fluconazole &Voriconazole,
b- Sensitive to both Fluconazole &Voriconazole,
c- Sensitive to only Voriconazole

Out of total 55 Candida isolates, 30 strains were isolated from hospital wards and 25 strains from ICUs. Maximum number of Candida strains i.e. 8 were isolated from Obstetrics and Gynaecology ward.



Fluconazole MIC: 1.5 µg/ml



Voriconazole MIC: .012 µg/ml

Photograph 2. MIC of Fluconazole and Voriconazole by E Test (bioMerieux)

Among 30 Candida strains isolated from different wards 10 (18.2%) were *C. tropicalis*, followed by 8 (14.5%) *C.albicans*, 6(10.9%) *C.parapsilosis*, 4 (7.3%) *C. pelliculosa* and 2(3.6%) *C. glabrata*. 25 Candida strains were isolated from different ICUs. Maximum number of Candida strains i.e.14 (56%) were isolated from Medicine ICU. Out of these 25 strains, 10 (40%) were *C. tropicalis* followed by 8 (32%) C.*albicans*, 3 (12%) were *C. parapsilosis*, 2 (8%) were *C. glabrata* and 1 (4%) each were *C. kefyr* and *C. krusei* respectively. Table 1shows the number of male patient were 24 and female patient were 31 . Male to female ratio was 1:1.3, showing female preponderance. Table 2 shows 46 (83.7 %) patients were catheterized for more than 5-7days, which was statistically significant where p value was <0.005; and 9(16.3 %) patients were non-catheterized. A total number of 48 (87.3%) patients were with Health care Associated infections (HAI). 55.5% patients were from different hospital wards and 45.5% patients were from different ICUs. 46 (83.7%) Candida strains were isolated from catheterised patients and 2 strains from non catheterised patients. 4 strains of *C. glabrata* and 1 strain of *C. krusei* were excluded from fluconazole susceptibility test as they are intrinsically resistant to fluconazole. Hence, only 50 strains were tested for fluconazole susceptibility, whereas all 55 Candida strains were tested for voriconazole susceptibility. Antifungal susceptibility pattern showed 24 strains sensitive to both fluconazole and voriconazole, 16 strains resistant to both fluconazole and voriconazole. There was no strain resistant to only voriconazole. 4 strains were resistant to only fluconazole and 6 strains were susceptibile-dose-dependent (SDD) to fluconazole. Total 20 (40%) strains were resistant to fluconazole.

DISCUSSION

In the present study, the incidence of candiduria by non-albicansspecies (70.9%) was high compared to *C. albicans* (29.1%) and this changing trends in etiopathogenesis of UTI is a matter of concern. The result was comparable to another study where ,*non albicans Candida* species were predominant (66.7%), compared to *Candida albicans*(33.3%) (Goyal *et al*., 2016). Candida tropicalis was the commonest species (36.4%) in our study followed by *Candida albicans* (29.1%) which was also reporterd by other workers that the most common species was C. tropicalis (68.2 %) followed by C. albicans (32 %) (Choudhary *et al*). In this study, female preponderance was observed with overall male: female ratio as 1:1.3 indicating that female gender is a risk factor which was also comparable with the study which has reported the ratio as 1:1.3 (Goyal *et al*., 2016). 25 candida isolates were from Intensive care units including 14 from medicine ICU which also correlated well with other study (Sharma *et al*, 2016). The most important predisposing factor for Candiduria in hospitalized patients is catheterization. In the present study, 83.7 % patients were catheterized for more than 7 days which was found to be statistically significant (p = <0.005) when compred with noncatherised patients having Candiduria. It has been reported that,the risk of non-albicanscandiduria increases 0.2-0.5 times higher with urinary catheterization and on admission to ICU or surgical units (Sharma *et al*, 2016).Species identification is important before starting the antifungal therapy as few species of Candida are intrinsically resistant to some antifungals. In present study, Antifungal susceptibility showed 40% resistance to Fluconazole which is comparable to other study i.e. 48.4% (Mohamoudabadi *et al*, 2013).In a tertiary care centre of South India, 30.8% resistance to fluconazole was noted (Giri *et al*., 2013).

Conclusion

We hereby conclude that there is need of considering candiduria as an emerging nosocomial infection, which represents diagnostic and therapeutic challenge for clinician. Routinely all urine samples should be cultured on SDA for detection of Candiduria and species identification and antifungal drug sensitivity test should be done for effective treatment of the patient.

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