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Study on Changing Trend in the Clinical Distribution of Candida Species and Their Antifungal Resistance in a Tertiary Care Hospital of North India.

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Abstract

Introduction: *Candidiasis, a major fungal infection caused by Candida spp., ranges from superficial to invasive disease. While Candida albicans predominates globally, non-albicans Candida (NAC) species are increasingly reported, often exhibiting higher antifungal resistance. Species-level identification and susceptibility testing are essential for optimal management. This study aimed to determine the distribution of Candida spp. from various clinical specimens in a tertiary care hospital in North India and assess their antifungal susceptibility profiles.*

Aim and Objective: *Study on changing trend in the clinical distribution of candida species and their antifungal resistance in a tertiary care hospital of north india.*

Materials and Methods: *Over one year, 2190 clinical specimens were processed in the Microbiology Department. Candida identification was based on direct microscopy, culture on Sabouraud's dextrose agar, and speciation using germ tube test, cornmeal agar, sugar fermentation/assimilation, and CHROMagar. Antifungal susceptibility was performed by CLSI agar diffusion method for ketoconazole, itraconazole, clotrimazole, fluconazole, amphotericin B, and nystatin.*

Results: *Out of 2190 samples, 153 (6.9%) yielded Candida spp., most frequently from urine (32.1%), high vaginal swabs (28.1%), and sputum (13.7%). Species distribution was: C. albicans (45.1%), C. tropicalis (26.7%), C. glabrata (13.7%), C. parapsilosis (8.4%), and C. krusei (5.8%). Diabetes mellitus (24.9%) and prolonged medication (19.6%) were leading predisposing factors. All isolates were sensitive to amphotericin B and nystatin. Resistance was highest to fluconazole (21.5%), particularly in NAC species (C. krusei 100%, C. parapsilosis 38.5%, C. glabrata 33.3%).*

Conclusion: *NAC species now surpass C. albicans in prevalence and exhibit greater azole resistance, notably to fluconazole. Routine speciation and antifungal susceptibility testing are crucial for guiding therapy and curbing resistance, especially in high-risk hospitalized patients*

Key words: *Non-albicans Candida, Candidiasis, Speciation,Antifungalresistanc*

INTRODUCTION:

Candidiasis, which accounts for 66-80% of fungal infections, is a superficial or a deep- Seated infection caused by opportunistic yeasts belong to the genus Candida. Among all Candida species, Candida albicans is by far the most common in all clinical forms of candidiasis, representing 70-80% of all yeast isolate(1). Candida can cause a great variety of infections, including simple mucocutaneous to severe invasive infections that can involve virtually any organ.(2) Candida spp. is one of the most frequent pathogens isolated in bloodstream infections (BSI), and is associated with significant morbidity and mortality in hospitalized patients (3).

In intensive care units, infection with Candida species is the 3rd most frequent cause of nosocomial bloodstream infection and is associated with a crude mortality rate of 47% (4). The overall increase in Candidemia in recent years is complicated by the emergence of non-albicans Candida species as both colonizers and pathogens causing nosocomial fungal blood stream infection (BSI) C. glabrata becomes an increasingly important pathogen with increase in age. The contribution of individual Non albicans

Candida (NAC) species also varies with patients and diagnostic groups. Candidemia due to C. parapsilosis is generally associated with catheters, hyper-alimentation, or prosthetic devices. Also, C. parapsilosis is the most common species of Candida to be isolated from the hands of health care workers in ICU, especially those who wear gloves (5)

Similar to the western world, the rise in frequency of NAC species has been observed in tertiary care centers in India as well with isolation rates ranging from 52 to 96% (5).Accurate species identification is important for the treatment of the Candida infections, as the non-albicans species of Candida continue to be increasingly documented and as not all the species respond to the same treatment.

The inappropriate use of antifungal drugs and introduction of over-the counter antimycotics in countries worldwide predispose development of antifungal resistance. This study is undertaken to Speciate Candida from the clinical cases of candidiasis and to determine the susceptibility of the Candida species to various antifungals

Materials and Methods:

All the Candida isolates which were isolated from various clinical samples submitted to the Central laboratory of Department of Microbiology during the study period of one year, were processed for speciation and antifungal susceptibility pattern.(6,7,8)

Defining criteria of candidiasis- Smear showing budding yeast cells, pseudohyphae and pus cells denoted infection by Candida species. The samples were subjected to various mycological tests as detailed below:

1. Direct examination: KOH Wet Mount &Gram's stain
2. Isolation of Candida: For Culture, Sabouraud's Dextrose Agar (SDA) was used. Colonies were identified by the colony characters and by examining a stained smear of the culture by gram's stain.

Once the colonies were confirmed, speciation was done by the following methods.

a. Germ tube
b. Corn meal agar inoculation
c. Sugar Fermentation
d. Sugar Assimilation
e. CHROMagar Candida

Antifungal susceptibility testing of the yeast isolates was assessed using the agar diffusion method according to CLSI guidelines. The discs were supplied by Hi-Media, Mumbai. Muller-Hinton agar supplemented with 2% glucose and 0.5µg/ml methylene blue was used for the sensitivity testing.

Observations and results:

A total of 2190 positive clinical culture samples were screened, out of which 153 Candida species were isolated from various clinical specimens which were further subjected to speciation and antifungal susceptibility testing. The prevalence of Candida species are studied in relation to age, sex, site of isolation, underlying conditions and predisposing factors

Hundred and fifty three (153) samples out of the 2190 specimens, tested positive for Candida species giving a prevalence rate of 6.9%. The highest number of specimen was urine culture which yielded 49 positive candida isolates, giving an incidence of 9.1% Candiduria.

Table-1: Prevalence of Candida Isolates in Clinical specimens

Clinical specimens	Total no. of screened	Total no. of isolates	Percentage
Urine	541	49	9.1%
High vaginal swab	408	43	10.5%
Sputum	482	21	4.3%
Nail	63	5	7.9%
Skin scrapping	93	9	9.6%
Oral swabs	87	11	12.6%
Pus	362	6	1.6%
Ear swabs	57	4	7%
Stool	59	3	5.1%
Blood	38	2	5.2%
Total	**2190**	**153**	**6.9%**

Table-2: Distribution of Candida isolates in Clinical Specimens

Clinical specimens	Total no. of isolates	Percentage
Urine	49	32.1%
High vaginal swabs	43	28.1%
Sputum	21	13.7%
Nail	5	3.2%
Skin scrapping	9	5.8%
Oral swab	11	7.1%
Pus	6	3.8%
Ear swabs	4	2.6%
Stool	3	1.9%
Blood	2	1.3%
Total	153	100%

The highest number of isolates were from urine samples (32.1%) followed by High vaginal swab (28.1%), sputum (13.7%), Oral swabs (7.1%), skin scrapping (5.8%), Pus (3.8%), Nail (3.2%), Ear swabs (2.6%), Stool (1.9%) and the least number from Blood (1.3%)

Table-3: Different Species of Candida isolate

Organism	Total Isolates	Percentage
Candida albicans	69	45.1%
Candida tropicalis	41	26.7%
Candida glabrata	21	13.7%
Candida parapsilosis	13	8.4%
Candida krusei	9	5.8%
Total	153	100%

Candida albicans was the major species accounting for 69 (45.1%) of the total isolates. Non albicans Candida constituted 41 (26.7%) of Candida tropicalis, followed by Candida glabrata 21 (13.7%), Candida parapsilosis13 (8.4%) and C.krusei9(5.8%)

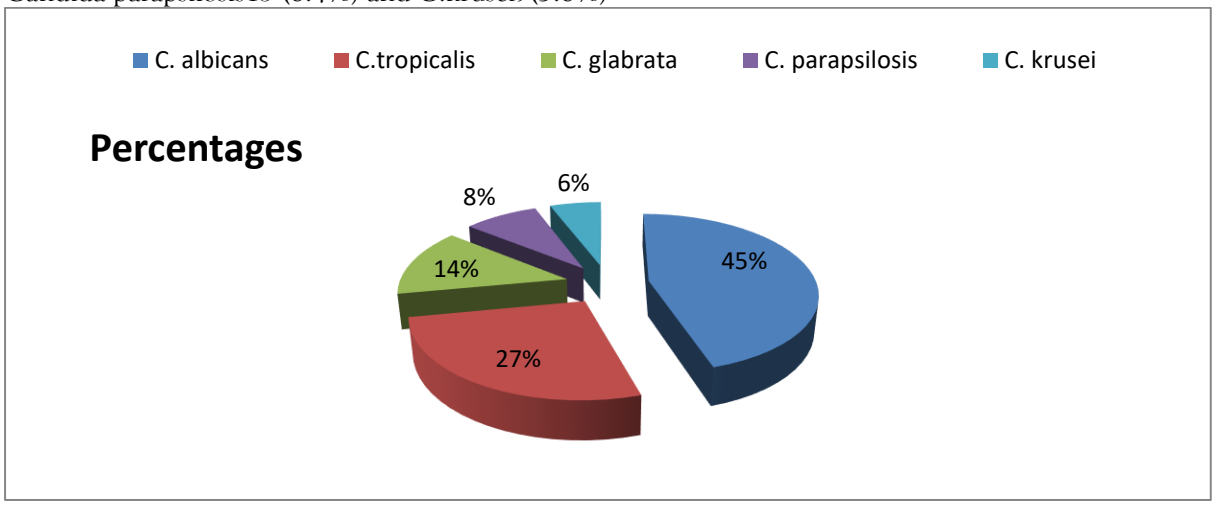


Table-4 Distribution of different species of Candida among various clinical specimen

Clinical specimens	C.albicans	C. tropicalis	C.glabrata	C.parapsilosis	C.krusei	Total (%)
Urine	6 (12.2%)	27 (55.1%)	11(22.4)	3 (6.1%)	2 (4.1%)	49 (32.1%)
High vaginal swab	32 (74.4%)	6 (13.9)	0	4 (9.3%)	1 (2.3%)	43 (28.1%)

Sputum	7 (33.4%)	5 (23.8%)	5 (23.8)	4 (19.1%)	0	21 (13.7%)
Nail	3 (60%)	0	0	2 (40%)	0	5 (3.3%)
Skin scrapping	5 (55.6%)	0	0	0	4 (44.5%)	9 (5.9%)
Oral swab	8 (72.7%)	0	2 (18.2%)	0	1 (9.1%)	11 (7.9%)
Pus	2 (33.4%)	1 (16.7%)	2 (33.4%)	0	1 (16.7%)	6 (3.9%)
Ear swab	4 (100%)		0	0	0	4 (2.6%)
Stool	2 (66.7%)	0	1 (33.4%)	0	0	3 (1.9%)
Blood	0	2 (100%)	0	0	0	2 (1.3%)
Total	69	41	21	13	9	153

The highest number of isolates was from urine samples isolated from Candiduria, constituting 49 (32.1%). The other major samples were High vaginal swab 43 (28.1%), sputum 21 (13.7%), oral swabs 11(7.9%), skin scrappings 9 (5.9%), pus 6 (3.9%), nail clippings 5 (3.3%), ear swabs 4(2.6%), followed by stool 3 (1.9%) and blood 2 (1.31%). Among the 49 urine specimens, 27(55.1%) isolates were C. tropicalis, 6 (12.2%) were C.albicans, 11(22.4%) were C. glabrata, 3 (6.1%) were C. parapsilosis and 2 (4.1%) were C. krusei. Among the 2nd highest specimen i.e 43 high vaginal swabs, 32 (74.4%) were C.albicans, 6(13.9%) were C.tropicalis, 4 (9.3%) were C.glabrata and 1(2.3%) were C. krusei.

Table-5: Gender wise distribution of isolates with various predisposing factor

Predisposing factor	**Male**	**Female**	**Total** (%)
Diabetes mellitus	21	17	38 (24.9%)
Biomedical devices	17	8	25 (16.3%)
Prolonged medication and Secondary to other disease	9	21	30 (19.6%)
Trauma	11	6	17 (11.1%)
Surgical cases	5	2	7 (4.6%)
Pregnancy	0	26	26 (17%)
Others	7	3	10 (6.5%)
Total	70	83	153 (100%)

In the study, Diabetes mellitus was the major predisposing factor constituting 38 (24.9%) cases followed by prolonged drug intake / secondary infection 30 (19.6%), pregnancy 26 (17%), Biomedical devices 25 (16.3%), Trauma 17 (11.1%) and Others constituting 10 (6.5%) cases among predisposing factors and included catheterization, stay in ICU, parenteral nutrition, prematurity and low socio-economic status, poor personal hygiene. Surgical cases were in least number constituting only 7 cases (4.6%).

Table-6: Distribution of Candida species among Various predisposing factors

Predisposing factors	**C. albicans** (%)	**Non-albicans Candida** (%)	**Total** (%)
Diabetes mellitus	29 (76.3)	9 (23.7)	38 (24.9%)
Biomedical devices	5 (20)	20 (80)	25 (16.3%)
Prolonged medication	7 (23.3)	23 (76.7)	30 (19.6%)
Trauma	3 (17.6)	14 (82.3)	17 (11.1%)
Surgical cases	1 (14.3)	6 (85.7)	7 (4.6%)
Pregnancy	24 (92.3)	2 (7.6)	26 (17%)
Others	0	10(100)	10 (6.5%)
Total	**69**	**84**	**153 (100%)**

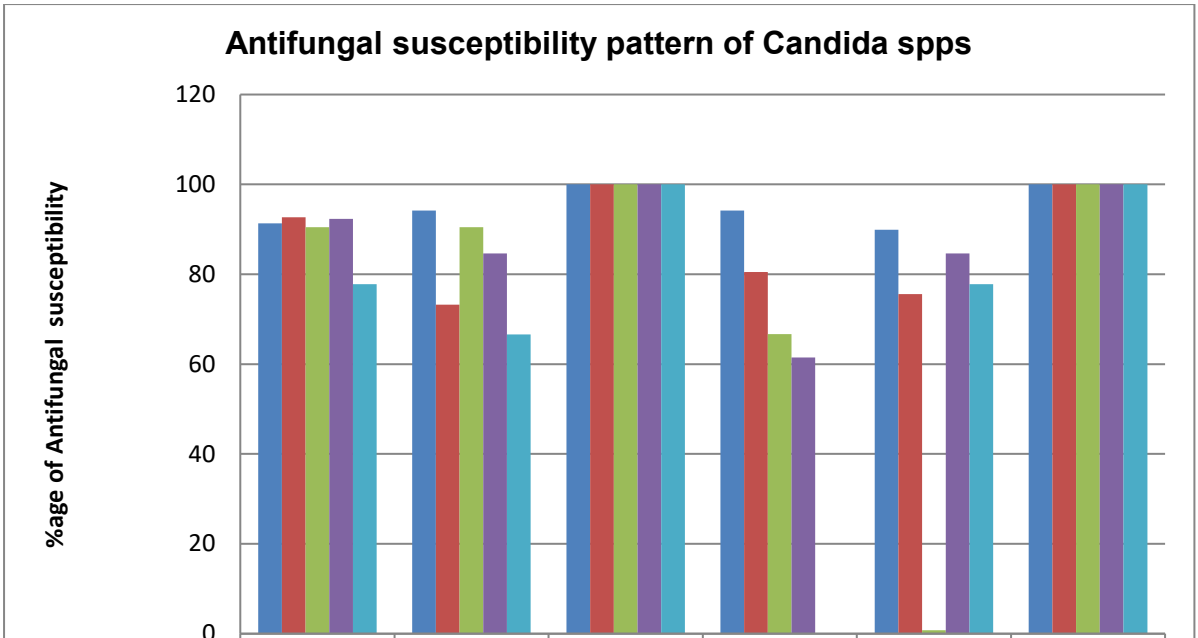
C. albicans 29 (76.3) was the major isolate in diabetes mellitus, followed by Various Non-albicans Candida 9 (23.7%) species. Among 26 Candida isolates having pregnancy as predisposing factors, 24

(92.3%) were C. albicans, 2 (7.2%) were Non-albicans Candida spp. The 30 Candida isolated from patients on prolonged broad spectrum antibacterial agents, corticosteroid and secondary to other infection, were C.albicans in 7 (23.3), and Non-albicans Candida in majority i. e 23 (76.7). Among 17 Candida isolates in patients with trauma 14 (82.3) isolates were Non-albicans Candida and 3 (17.6) isolates were C. albicans. All the patients with others as their predisposing factor showed Non-albicans candida isolates in their specimens. Among the 25 isolates of biomedical devices as their predisposing factor, 5 (20%) were C.albicans and 20 (80%) Non-albicans Candida 20 (80%).

Table-7 Antifungal susceptibility pattern of various Candida species

Species	KETO (1µg)		ITRA (10µg)		Nystatin (100U)		FLUC (10µg)		CLOT (10µg)		AMP (100U)	
	S	R	S	R	S	R	S	R	S	R	S	R
C. albicans (n=69)	63 91.3%	6 8.7%	65 94.2%	4 5.8%	69 100%	0 0%	65 94.2%	4 5.8%	62 89.9%	7 10.2%	69 100%	0 0%
C.tropicalis (n=41)	38 92.7%	3 7.3%	30 73.2%	11 26.8%%	41 100%	0 0%	33 80.5%	8 19.5%	31 75.6%	10 24.4%	41 100%	0 0%
C. glabrata (n=21)	19 90.5%	2 9.5%	19 90.5%	2 9.5%	21 100%	0 0%	14 66.7%	7 33.3%	16 76.1%	5 23.8%	21 100%	0 0%
C. parapsilosis (n=13)	12 92.3%	1 7.6%	11 84.6%	2 15.3%	13 100%	0 0%	8 61.5%	5 38.5%	11 84.6%	2 15.4%	13 100%	0 0%
C. krusei (n=9)	7 77.8%	2 22.2%	6 66.6%	3 33.3%	9 100%	0 0%	0 0%	9 100%	7 77.8%	2 22.2%	9 100%	0 0%
Total	139 90.9%	14 9.1%	131 85.6%	22 14.4%	153 100%	0 0%	120 78.4%	33 21.5%	127 83%	26 16.9%	153 100%	0 0%

In the present study, among 153 Candida isolates tested, no resistance was detected in the isolates to amphotericin-B and Nystatin For Itraconazole, 85.6% strains showed sensitiveness, 14.4% strains were resistant and among them 33.3% were C.krusei , 26.8% for C.tropicalis, 15.3% for C. parapsilosis, 5.8% for C. albicansand 9.5% for C. glabrata. For Fluconazole, maximum resistance of 21.5% was noted with only 78.4% sensitivity. C. krusei showed 100% resistance to fluconazole followed by C. parapsilosis (38.5%), C. glabrata (33.3%), C.tropicalis (19.5%). C. albicans showed least resistant to fluconazole (5.8%). Ketoconazole and Itraconazole were more sensitive followed by Clotrimazole and Fluconazole. All the isolates showed 100% sensitivity to Amphotericin B and Nystatin. Resistance was seen significantly among non-albicans Candida for fluconazole as compared to C. albicans.



	Ketoconazole	Itraconazole	Nystatin	Fluconazole	Clotrimazole	Amphotericin B
C. albicans	91.3	94.2	100	94.2	89.9	100
C. tropicalis	92.7	73.2	100	80.5	75.6	100
C. glabrata	90.5	90.5	100	66.7	76.10%	100
C. parapsilosis	92.3	84.6	100	61.5	84.6	100
C. krusei	77.8	66.6	100	0	77.8	100

DISCUSSION

Fungal infections have always been a problem for hospitalized patients but during the last thirty years fungal infections due to Candida spp., especially those that are caused by non-albicans species, increased enormously. Mostly candidiasis is caused by five common species, C. albicans, C. glabrata, C.parapsilosis, C. tropicalis and C. krusei(9). Common drugs used against candidiasis are amphotericin B and fluconazole belonging to polyenes and azoles groups of antifungal agents, respectively. Candida species show different antifungal susceptibility pattern against the same drug. Some non-albicans species of Candida, i.e., C. krusei and C. glabrata show complete resistance and reduced susceptibility to fluconazole respectively. This emphasizes that the correct identification and antifungal susceptibility of Candida species is essential for the selection of appropriate antifungal agent for successful therapy (6).

Among 2190 suspected clinical samples received in the Department of Microbiology laboratory, 153 samples were positive for Candida species, giving a prevalence rate was 6.9%. The prevalence rate by Rizvi et al, Amar CS et al and Feglo et al study was 10.6%, 6.1%, and 12.7% respectively. The prevalence rate was lower in present study (6.9%) in comparison to Rizvi et al and Feglo et al (10, 11). However it was comparable to Amar CS et al 6.1% (12).

Our study elucidated the distribution of various Candida spp. among Clinical isolates so that maximum numbers of Candida species were isolated from urine samples (32.1%). Roy R.C.et al., 2013, Lata R Patel et al., 2012, and Almeida, A.A et al., 2013 had also reported most of Candida spp. from urine samples in their study (62%) (13,14, 15). Out of 49 (32.1%) isolates in urine, 6 Candida isolates were Candida albicans and 43 were non- albicans Candida species so that among non albicans Candida species, Candida tropicalis (55.1%) was the most predominant species isolated in urine. This was in agreement with Kashid et al 2011, Patel et al 2012, Kumar et al 2013, as they found highest number of isolates from urine and among which Candida tropicalis was the most predominant species (16, 9,17).

The isolation of non-albicans Candida has been frequently encountered from Candidemic patients in the past few decades as shown in the study conducted by Kothari et al. (18). Nevertheless in our study, only two Candida isolates were obtained from blood specimen and both of them were C. tropicalis. This could be due to the limited number of blood specimens included in this study. This study reported the prevalence of five species i.e., C. albicans, C. tropicalis, Cglabrata, C. krusei and C. parapsilosis. Candida

albicans (45.1%) was dominant over the individual nonalbicans species of Candida. Results of our study are consistent with that of Ariff et al. 2011 reporting that C. albicans was the most prevalent among all the species, followed by C. tropicalis and C. glabrata(19).

we observed that when put together, Non albicans candida (55.9%) are more frequently encountered than C. albicans (45.1%). This is in agreement with the studies conducted by Chakrabart et al. and Mokaddas et al who found that Non albicans Candida had a higher isolation rate than C. albicans from different clinical samples (20, 21).

We have studied the association of the risk factors in all the 153 patients from whom the Candida species were isolated. As can be seen in [Table 6 of results], diabetes mellitus was the leading most frequently associated risk factor. A comparison of the incidence of diabetes among the cases of candidiasis in various studies is shown below in a table:

Comparison of incidence of diabetes mellitus in various studies

Workers' study	Total no. of cases	Diabetic cases	Percentage
Kandhari KC et al[22]	54	11	20.4%
Shroff PS et al[23]	150	22	14.66%
Present study	153	38	24.9%

In the present study, a history of multiple and prolonged drug usage was the second most frequently associated risk factor (19.6%) and is in agreement with the studies done by Guru P et al 2016 and Dharwad et al 2011 with 16% and 22% respectively (24, 25). Pregnancy was the third most common predisposing factor. In the present study Candida species was isolated from 17% cases of Pregnancy with C.albicans as the most common species isolated (92.3%). Praveen et al., 2008 found that frequency of vaginal candidiasis during pregnancy was 38% (26).

AFST was done for 153 isolates with Amphotericin, Fluconazole, Nystatin, Itraconazole, Clotrimazole and Ketaconazole. In our study, 14.4% showed resistance to Itraconazole, 16.9% showed resistance to Clotrimazole, 21.5% showed resistance to Fluconazole, 9.1% showed resistance to ketoconazole. No resistance detected in the isolates to amphotericin-B and nystatin in the present study. Rizvi et al also reported same finding, no resistance to amphotericin-B, nystatin and Voriconazole(10). Our findings correlated well with those in the studies by Kashid et a, Vijaya D et al and Roy et Al (16, 27, 28).

Table 5.3: Resistance to Fluconazole in different studies

Species	**C.albicans** (%)	**Non albicans Candida spp** (%)
Present study	2.6%	18.95%
Patel et al Gujrat 2012 [9]	25.5%	57.6%
Pahava et al indore 2014 [29]	2%	3,9%
Roy et al Northeast study Chhattisgarh 2013 [13]	12%	24%
Kashid et al Banglore 2011 [16]	18.60%	35.6%
Shivanand et al Karnataka 2011 [30]	22%	25%

In the present study the Non albicans Candida spp showed 18.95% resistance to azole group [Fluconazole] of drugs and C. albicans isolates showed 2.6% resistance to azole group of drugs. Also the finding was correlated with those of a study done by Shivanand et al in which NAC shows 25% resistance to fluconazole (30). In our study resistance for fluconazole by C. krusei was 100%, This was also confirmed b Amar C S et al and Roy et al who reported that all isolates of Candida krusei tested were resistance to fluconazole (12, 13). It is well established that Candida krusei is intrinsically resistant to fluconazole.

In recent years, the global epidemiology of candidiasis has shifted significantly, with non-albicans Candida (NAC) species surpassing C. albicans in many clinical settings. Our study corroborates this trend, with 55.9% of isolates belonging to NAC species, consistent with reports from other Indian tertiary care centers. Recent global surveillance data from 2024–2025 also highlight C. tropicalis and C. glabrata as leading causes of invasive candidiasis in Asia, whereas C. parapsilosis predominates in device-associated infections in Europe and Latin America. This species variability underscores the importance of regional surveillance in guiding empirical therapy.

Antifungal resistance continues to be a critical concern. In our study, fluconazole resistance was 21.5%, with the highest resistance in C. krusei (100%) and C. parapsilosis (38.5%). These findings align with recent multicenter surveillance studies in 2024, which report rising azole resistance across Asia, particularly in C. tropicalis and C. glabrata. Moreover, the emergence of multidrug-resistant C. auris,

although not isolated in our cohort, remains a growing public health challenge worldwide, as highlighted by WHO in 2025 as a "critical priority pathogen."

The clinical correlation between predisposing factors and species distribution in our study further reinforces established risk patterns. Diabetes mellitus and prolonged drug exposure remain the strongest predictors of candidiasis, while pregnancy-associated cases continue to be dominated by C. albicans. However, device-related and trauma-associated infections were significantly linked to NAC isolates, consistent with 2025 literature emphasizing biofilm-associated resistance as a major therapeutic hurdle.

Amphotericin B and nystatin retained 100% activity in our isolates, reflecting their continued reliability. Nevertheless, reliance on polyenes is limited by toxicity, and resistance surveillance for newer triazoles and echinocandins has become crucial. Reports in 2025 warn of increasing echinocandin resistance, particularly in C. glabrata, due to mutations in the FKS gene, raising concern over treatment failures. These developments emphasize the need for stewardship programs, routine antifungal susceptibility testing, and molecular resistance monitoring to mitigate therapeutic challenges.

Overall, our findings are consistent with the global 2025 consensus that candidiasis management requires a tailored approach—guided by local epidemiology, species distribution, and resistance patterns—to preserve the effectiveness of existing antifungal drugs and improve patient outcomes (31-36).

CONCLUSION

There are various reports from our country showing increasing trend towards fluconazole resistance especially among NAC Species which warrants its judicious use as a prophylactic agent in hospitals. It is important for both Clinicians and Microbiologists to identify the complete clinical response to the given treatment, for which adequate information regarding the change in distribution of Candida species and their antifungal susceptibility pattern will be beneficial.

DECLARATIONS

Conflicts of interest: There is no any conflict of interest associated with this study

Consent to participate: There is consent to participate.

Consent for publication: There is consent for the publication of this paper.

Authors' contributions: Author equally contributed the work.

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