Original Research Article

A study of Candidiasis in immunocompromised patients attending in A Tertiary Care Hospital, Darbhanga, Bihar

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Abstract

In this new scenario, fungal infections have emerged as a critical issue in the compromised host. Among these, candida spp. is the most common fungal pathogens. **Materials and Methods:** The present study was carried out in DMCH, Darbhanga. The cases for study were selected from the patients attending the medicine, ICTC, Endocrinology, Haematology, Nephrology and Intensive Care unit of DMCH, Darbhanga over a period of one year (March 2020 – February 2021). **Result:** The overall prevalence of Candidiasis among the study population was found to be 38.57% Candida spp. was isolated in 48.33% of diabetic population, 39.13% of individuals with HIV/AIDS, 18.18% in patients with haematological malignancy and 25% in patients undergoing renal transplant ($p<0.02$). **Conclusion:** It may be concluded from this study that people who are immunosuppressed are at higher risk for Candida infection. The study highlights the diverse manifestations caused by Candida species and throws light on the species prevalent locally.

Keywords: Candidiasis, Immunocompromised

Introduction

A dramatic change in the epidemiology of infectious diseases has taken place with the advent of new chemotherapeutic agents, new immunosuppressive agents, organ transplantation, parenteral alimentation, broad-spectrum antibiotics and advanced surgical techniques. In this new scenario, fungal infections have emerged as a critical issue in the compromised host. Among these, candida spp. is the most common fungal pathogens [1].Opportunistic infections are known to occur in people in whom the immune system is impaired and are caused by infectious agents that do not ordinarily produce disease in healthy individuals. Mims et al[2] refers to immunocompromised hosts as people who have one or more defects in their body's natural defenses against microbial invaders. A host may be compromised by factors affecting either the innate or the adaptive system of immunity. Some of the causes of secondary adaptive immunodeficiency include infections especially HIV infection, neoplasia, diabetes mellitus, chemotherapy and radiation therapy, patients receiving solid organ transplant.

Factors contributing to the development of fungal infections in immunocompromised hosts are as follows:[3]

- Exposure to the fungus
- Inoculum delivered during the exposure
- Virulence of the fungus
- Host's immune status
- Underlying disease
- Immunosuppressive therapy
- Prior exposure to the fungus

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Candidas are thin-walled,small yeasts (4 to 6 microns) that reproduce by budding. Even though there are more than 200 species of Candida, no more than 20 cause disease in humans with varying frequency [4]. Of these, *Candida albicans* causes almost 100% of cases of oropharyngeal candidiasis and at least 90% of cases of Candida vulvovaginitis. When Candida produces invasive candidiasis, the other species of Candida begin to be seen with increased frequency. Candida species are the most frequent cause of fungal infections in the immunocompromised host. Species most commonly recovered include *Candida albicans, Candida tropicalis,Candida* (Torulopsis) *glabrata* and *Candida parapsilosis.* Others such as *Candida lusitaniae*, *Candida rugosa* and *Candida pseudotropicalis* have more recently been reported to be associated with disseminated infection. Some are considered as a part of the usual flora, which makes the clinical significance of an isolate difficult to determine. Recovery of an organism from a normally sterile site or recovery of the same species from several different body sites is an indicator of disseminated infection with probable fungemia[5].Candida spp. is also competing with the bacteria as one of the leading causes of nosocomial infections [6]. During the 1990's Candida spp. was the fourth most common agent causing nosocomial bloodstream infection. In addition the ability of Candida spp. to produce oropharyngeal candidiasis in patients with HIV-AIDS has made candidiasis the leading fungal infection in this immunosuppressed population [7].Exposure to Candida species is universal since these organisms are a part of the human gastrointestinal (GI) tract flora. The amount of growth in the GI tract and hence, the potential inoculums, is increased by broad spectrum antibiotic therapy and the use of chemotherapeutic agents. In recent years, an important concern has been the prominence of non-albicans species in some medical centers. Several of these species, specifically C.*krusei* and C.*glabrata* are less susceptible to fluconazole, the primary azole antifungal agent used to treat Candida infections. Whether fluconazole therapy is instrumental in selecting these more resistant species has not yet been clearly defined but it is

likely that overuse of this azoles does play a role in some settings. Another concern is the emergence of fluconazole resistant C.*albicans* among AIDS patients who have frequent episodes of thrush that require treatment with multiple courses of fluconazole. The varies manifestations of candidiasis often create dilemma for the physician, both in diagnosis and treatment, and are frequently the major contributors to or are the actual cause of death in the patient. Several studies from different parts of the country have reported prevalence of Candidiasis in different immunocompromised patients [8]. However, there has been no record of such studies being conducted in the state of Bihar till date.

Aims and Objectives:

1. To study the prevalence of Candidiasis in immunocompromised patients attending DMCH, Darbhanga.
2. To identify the different species of Candida isolated in the study.

Materials and Methods

The present study was carried out in DMCH, Darbhanga. The cases for study were selected from the patients attending the medicine, ICTC, Endocrinology, Haematology, Nephrology and Intensive Care unit of DMCH, Darbhanga over a period of one year (March 2020 – February 2021). Immunocompromised patients of different categories like HIV seropositive individuals, diabetes mellitus, patients with haematological malignancies with total leukocyte count of less than 4000, transplant recipients and patients receiving immunosuppressive drugs were included in the study. Patients with history and clinical features suggestive of fungal infection were selected for the study. Informed consent was taken from the patient before conducting the study. The patients included both sexes and all age groups belonging to different socioeconomic status, religions and regions.

Laboratory Investigations:

Routine Blood Examination: Routine blood examination for haemoglobin, total and differential leukocyte count and erythrocyte sedimentation rate were done in all cases.

Serological Examination: Two rapid tests HIV COMB (Span Diagnostics Ltd. Surat, India) and Signal HIV (Span Diagnostics Ltd.) and one ELISA, MICROELISA (J.Mitra& Co. Pvt. Ltd.) for HIV antibodies was carried out after proper counselling and consent in selected cases attending the Integrated Counselling & testing Centre (ICTC) attached to the Department of Microbiol DMCH, Darbhanga. CD4/CD8 cell counts were also performed in the HIV seropositive cases using the fluorescent assisted cell sorter as per prescribed method.Organisms are identified on the Basis of Colony Character, Gram staning, KoH Mount, Biochemical Reaction and indin ink Prepration.

Result

A total of 100 immunocompromised individuals admitted in different departments were taken up for this study. The specimens were subjected to direct microscopy in 10% KOH mount, culture in Sabouraud's dextrose agar supplemented with Chloramphenicol (0.05 mg/ml). They were then incubated at 22^0C and 37^0C and observed for fungal growth. Special tests were done to isolated Candida species. They were Gram staining, Germ tube test, Hi Chrome Agar, Corn Meal agar morphology, sugar fermentation and sugar assimilation test. The results of all these tests were then tabulated and analyzed in relation to demographic factors like age, sex, socioeconomic status, clinical features, CD4 cell count and random blood sugar level.

The study population consisted of 49 cases of diabetes mellitus, 32 cases of HIV/AIDS, 12 patients with haematological malignancy, and 7 patients who had undergone renal transplant surgery.

The sample size comprised of a total of 140 samples, of which 50 were urine, 40 bloods, 20 oral swab, 16 sputum and 14 pus.

The overall prevalence of Candidiasis among the study population was found to be 38.57% Candida spp. was isolated in 48.33% of diabetic population, 39.13% of individuals with HIV/AIDS, 18.18% in patients with haematological malignancy and 25% in patients undergoing renal transplant ($p<0.02$).The maximal number of patients positive for Candida spp were in the age group of 31-40 years (34.1%) followed by 51-60 years (24.4%) age group. The sex distribution showed a male preponderance with 75.6% males and 24.4% females.The majority of the (63.41%) patients came from rural area ($p<0.01$).The majority of the patients were from a low socioeconomic status with 26.80% having completed education upto twelve standard and 53.65% belonging to low income group.Clinically the patients presented with fever in 73.17% of patients while the rest 26.83% manifested other clinical features ($p<0.01$).The CD4 cell counts were between 51-100 cells/mm^3 in 43% of HIV seropositive patients while 28.6% had counts between 101-150 cells/mm^3.The species profile of Candida reveals that non-albicans Candida (74.07%) outnumbered the *C. albicans* (25.92%) group. Among the non albicans group,*C. tropicalis* (n=22, 40.74%) was the most frequent isolate in all clinical specimen followed by *C. parapsilosis* (n=6,11.11%), *C. krusei* (n=5,9.25%), *C. guilliermondii* (n=2,3. 70%), *C.rugosa* (n=2,3.70%), *C.dubiliniensis*(n=1, 1.85%), *C. glabrata* (n=1,1.85%) and *C.zeylanoides* (n=1,1.85%).

Table 1: Clinical profile of study population

Study population	No (%) of patients		
	Male	Female	Total
Diabetes mellitus	32	17	49 (49%)
HIV/AIDS	27	5	32 (32%)
Malignancy	7	5	12 (12%)
Transplant	3	4	7 (7%)
Total	69(69%)	31(31%)	100(100%)

Table 2: Profile of Candida Species in Study Population

Species	Total	%
Candida albicans	14	25.92
C. tropicalis	22	40.74
C. parapsilosis	6	11.11
C. krusei	5	9.25
C.guilliermondii	2	3.70
C. rugosa	2	3.70
C. dubliniensis	1	1.85
C. glabrata	1	1.85
C. zeylanoides	1	1.85

Table 3: Profile of Different Species isolated in Various Specimens

	C. albicans	Non albicans candida spp.							
		C. tropicalis	*C. parapsilosis*	*C. krusei*	*C. rugosa*	*C. guilliermondii*	*C. glabrata*	*C. zeylanoides*	*C. dubliniensis*
Blood	3	7	1	2	1	0	0	0	0
Oral Swab	3	2	1	0	0	2	0	0	1
Sputum	0	3	0	1	0	0	1	0	0
Urine	7	10	3	1	1	0	0	1	0
Pus	1	0	1	1	0	0	0	0	0
Total	14	22	6	5	2	2	1	1	1

Table 4: Distribution of different species in 29 culture positive samples in diabetic population

Specimen	*C.albicans*	Non albicans candida spp.					
		C. tropicalis	*C. parapsilosis*	*C. krusei*	*C. glabrata*	*C. rugosa*	*C. zeylanoides*
Blood	2	3	1	1	0	1	0
Pus	1	0	1	1		0	0
Sputum	0	0	0	0	0	0	0
Urine	5	8	2	1	0	1	1
Total	8	11	4	3	0	2	1

Table 5: Distribution of different species in 18 culture positive samples in HIV/ AIDS group

Specimen	*C. albicnas*	Nonalbicans Candida					
		C. krusei	*C. tropicalis*	*C. guilliermondii*	*C. dubliniensis*	*C.glabrata*	*C.parapsilosis*
Blood	0	0	2	0	0	0	0
Oral Swab	2	0	2	2	1	0	1
Sputum	0	1	3	0	0	1	0
Urine	2	0	1	0	0	0	0
Total	4	1	8	2	1	1	1

Discussion

Candidiasis has emerged as a significant medical problem owing to indiscriminate long-term use of antibiotics, immunosuppressive agents and cytotoxic therapies, immune defects and more recently in AIDS and AIDS related complex.A number of studies have investigated the prevalence and epidemiology of candidiasis in immunocompetent as well as immunocompromised groups of people. From India, a few studies have been reported in respect of different etiological species, clinical presentations, risk factors and age and sex. The prevalence rates of candidiasis in immunocompromised individuals and species identification have been found to be varying depending on the study design, geographical location, population group, and the associated risk factors.

Age Distribution: The age distribution of the study population ranged from 7-75 years (not shown in table). In the diabetic group, majority of the patients belonged to the age group of 45-65 years (Table-3A). On the other hand, majority of patients belonged to 30-40 years in the HIV/AIDS group (Table 3B). In malignancy group most of the patients were seen to belong to extremes of age group (Table-3C). In case of transplant group majority of the patients were in the age group of 20-40 years.[8]

Prevalence Of Candidiasis: The overall prevalence of Candidiasis among the study population of 100 immunocompromised patients with varied clinical manifestation was found to be 38.57% (Table 11) (P<0.02). Among the total isolates, non albicans Candida comprised of 74% of the total isolates. Among the non albicans species, *C. tropicalis* (n=22, 40.74%) was the most common isolate in all the specimen followed by *C. Parapsilosis* (n=6, 11.11%), *C. Krusei* (n=5, 9.25%) *C. guilliermondii* (n=2, 3.70%), *C. rugosa*(n=2, 3.70%) *C. dubliniensis* (n=1, 1.85%), *C. glabrata* (n=1, 1.85%) and *C. zeylanoides* (n=1, 1.85%) [9].S .Giri et al (2013) reported *C.tropicalis*(74.35%) was the most common isolate followed by *C.albicans, C.parapsilosis, C.krusei, C.glabrata.*Manisha Jain et al (2012) reported non-albicans candida spp(71.4%) emerged as the predominant pathogen causing nosocomial UTI. The present study noted a prevalence of 25.92% in blood stream infections of which non albicans species constituted 74%. Few Indian studies have also reported on the prevalence of candidiasis in different study population.F. Alvarez Lerma et al (2003) reported 22% prevalence in urine of critically ill patients, of which *C. albicans* was the most frequent species.This is in contrast to the present study which reported prevalence of 42.59% in urine of the study group, of which non albicans Candida species outnumbered the albicans group[10].

Species Distribution : Kanitha Tritipwanit et al (2005) observed a 57.1% prevalence of non albicans species, whereas *C. albicans* constituted 42.9% of the isolates. Among the non albicans, *C. tropicalis* (9.9%) was the most frequent isolate followed by *C. parapsilosis* (1.52%) and *C. guilliermondii* (0.76%). This compares with the present study, which also noted a similar Chakrabarti et al (2005), Shivprakash et al (2007) reported *C.tropicalis (*38% & 35.6%, respectively) as the most frequent isolate followed by *C. parapsilosis, C. glabrata* and *C. pelliculosa*. On the other hand, Somansu et al(2003) reported *C. albicans* as the predominant isolate from various clinical specimen followed by *C. tropicalis, C. parapsilosis, C. krusei, C. kefyr, C. guilliermondii and C. glabrata*.The present study is similar to those of Chakrabarti et al (2005), Shivprakash et al[11].

Candidiasis and Diabetes: Candida spp. was isolated in 29 (48.33%) out of 60 varied clinical specimen from 49 diabetic patients Dorko et al (2005) reported *C. albicans* as the major species isolated from most of the specimen comprising 64.4% of the isolates followed by *C. parapsilosis* (19.62%), *C.tropicalis* (9.3%), *C.krusei* (3.73%), and *C.guilliermondii*(1.8%). But in the present study, *C. albicans* comprised only 27.58% of the total isolates. *C. tropicalis* (n=11/29, 37.93%) was the most frequent isolate in all the clinical specimen (followed by *C. parapsilosis* (n=4/29, 13.79%), *C. krusei* (n=3/29, 10.34%), *C. rugosa* (n=2/29, 6.89%) and*C. zeylanoides* (n=1/29, 3.44%).Thus, the present study highlights the correlation between candidiasis and diabetes mellitus.

Candidiasis and HIV: Candidiasis is known to be the most common opportunistic infection in persons infected with HIV. In the present study, 18 (39.13%) out of 46 samples from 32 HIV seropositive patients were found to be associated with candidiasis, the overall prevalence being 39.13%.Baradkar et al (1999) and Arora et al reported *C. albicans* as the most frequent species isolated. However, the present study noted an increased prevalence of non-albicans species with *C. tropicalis* (57.14%) being the most common isolate followed by *C. albicans* (22.22%), *C. guilliermondii* (14.2%) *C.*

dubliniensis (7.1%), *C. parapsilosis*(7.1%),*C.glabrata*(7.1%) and *C. krusei (*7.1%).The findings of our study corresponds to that of the above-mentioned studies and lends credence to the view suggested in many studies that HIV predisposes to candidiasis.

Candidiasis and Malignancy: The present study found candidiasis in 4 out of 22 samples from 12 patients with haematological malignancy, the overall prevalence being 18.1%.Several studies have reported on the species profile of Candida in malignancy patients.Kremery V Jr et al (1999) reported an increase in prevalence of non-albicans species. Of the cases of nosocomial fungemia due to species, other than *Candida albicans, C. parapsilosis* was the most frequent isolate.The present study however noted an equal isolation rate of *C.tropicalis* and *C. krusei* in blood, 1 isolate of *C. parapsilosis* in urine and 1 isolate of *C.albicans* in oral swab, constituting only 25% of the isolates .

Candidiasis and Transplantation: The present study isolated Candida species from 3 out of 12 clinical samples from 7 transplant patients, the overall prevalence being 25%.*C.tropicalis* was the most common species isolated in all clinical specimens whereas *C.albicans* comprised of 33.3% of the isolates .

Profile Of Candidiasis Patients:

Age and Sex Distribution: The maximal number of patients positive for Candida were in their fourth decade of life (34.1%), followed by patients in the age group 51-60 years (24.4%),age group 41-50(12.2) and 21-30 years (12.2%) each as illustrated in Table 13B. The age of the patients ranged from 7 years to 75 years; the sex distribution revealed a male preponderance with 75.61% males and 24.39% females.Aggarwal et al (2005) also reported a higher incidence of the disease in the reproductive age group (16-45 years) with a male preponderance.Thus, the present study corresponds to the previously mentioned observations in terms of age and sex.

Socioeconomic Status: It is evident from Table 14 that majority of the patients had completed education upto twelve standard (26.80%). On the other hand, most of the patients belonged to low income group (53.65%) followed by middle-income group (39.02%).Poor socioeconomic condition, inadequate nutrition and poor hygiene have been observed to have a bearing of the incidence of the disease (Daniela Moreira et al 2004, Chakrabarti et al 2008).

Clinical Presentation Of Fever:

It is evident from Table 16 that 73.17% of the Candida positive patients had manifested fever as a clinical feature. On the other hand, 26.83% had clinical features other than fever ($p<0.01$). Aggarwal et al (2005) in their study noted that 56% of the patients had presented with fever while the rest 44% had varied clinical manifestations. Ayyagari et al (1999) and Talib and Singh (1998) noted similar finding.The present study can be correlated with these studies as the patients here also presented with multiple clinical features other than fever alone .

Cd4 T Cell Counts in the Hiv Seropositive Candidiasis Patients:

The maximum number of HIV seropositive candidiasis patients in the present study had CD4 T cell counts between 51-100 cells/mm^3 (43.0%) while there were 28.6% patients with counts between 101-150 cells/mm^3.Caroline et al (1991) observed that the risk of Candidiasis infection increases in more profoundly immunosuppressed HIV seropositive persons.Recently, it has been suggested that HIV viral load may be as much or possibly more of a predictor for oropharyngeal candidiasis than CD4 cell counts demonstrating strong correlation of oropharyngeal candidiasis to high viral load (Rina et al 2003). However, this parameter was not studied in the present study.

Random Blood Glucose Level in Candidiasis Positive Diabetic Patients: Majority of the candidiasis positive diabetic patients had blood sugar above 250 mg/dl with majority (n=6, 27.3%) in the range of 401-450 mg/dl. Mazen S Bader et al (2004) in their retrospective study correlated on the impact of hyperglycemia on hospital mortality in diabetic patients with candidemia. The study reported severe hyperglycemia (>13.9 mmol/dL or >250 mg/dL) as a marker of increased mortality among hospitalized patients with candidemia. As stated above, the present study results parallels the above mentioned finding.[11]It may be concluded from this study that people who are immunosuppressed are at higher risk for Candida infection. The study highlights the diverse manifestations caused by Candida species and throws light on the species prevalent locally.The study also emphasizes on the need for introduction of mycological examination into the panel of methods evaluating the clinical condition of this category of patients and the need for formulating preventive and prophylactic measures.Despite the moderate (n=140) number of samples included in the study, the data here provide some background information that can form the basis of future, more elaborative and systematic studies. Apart from this, it can also potentially have an immediate impact on patient care by suggesting appropriate interventional measures based on the results.

Conflict of Interest: Nil Source of support:Nil

Conclusion

It may be concluded from this study that people who are immunosuppressed are at higher risk for Candida infection. The study highlights the diverse manifestations caused by Candida species and throws light on the species prevalent locally.The study also emphasizes on the need for introduction of mycological examination into the panel of methods evaluating the clinical condition of this category of patients and the need for formulating preventive and prophylactic measures.Despite the moderate (n=140) number of samples included in the study, the data here provide some background information that can form the basis of future, more elaborative and systematic studies. Apart from this, it can also potentially have an immediate impact on patient care by suggesting appropriate interventional measures based on the results.

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