ISOLATION AND DISTRIBUTION OF CANDIDA SPECIES AMONG DIFFERENT CLINICAL SITUATIONS IN CRITICALLY ILL PATIENTS: PROSPECTIVE STUDY

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ABSTRACT

Bloodstream infections due to Candida species are important complications in severely ill hospitalized patients.A change in species distribution has been observed now a day with the emergence of many non-albicans Candida species. The aim of our study is to evaluate the incidence of distribution of different candida species in different clinical condition. Between 2009 and 2011 we encountered 106 episodes of candida species among 412 patients in ICU. Distribution of Candida species among culture positive in different clinical situation like 178 patient receiving TPN, 58 had candida species,124 patient of diabetes 20 had Candia species ,4 HIV patient 3 had Candida glabrata and 1 had C.tropicalis and in neutropenic patient 36 had Candia species out of 82 patient and all had statistically significant, p< 0.05.Among Candida isolates C.tropicalis was predominant species isolated in patient of diabetes, receiving TPN ,cancer and neutropenia.106 of blood Candida isolates were 52 (49.0%) Candida tropicalis, 28 (26.6%) C. albicans, 14(13.5%) Candida gullerimondi, 8 (7.8%) Candida glabrata and 4 (3.8%) were Candida krusei. Organisms were grown in Sabouraud dextrose broth.

Keywords: Candida isolates; risk factors; Candida culture; clinical situations

1. INTRODUCTION

Candida species are important nosocomial pathogens in critically ill patients and are associated with substantial mortality and prolonged hospitalization in the intensive care unit .[1,2] Candida albicans accounts for the majority of cases with candidemia, but an increasing number of infections due to non-albicans species. [3, 4]Two very large studies, The European Prevalence of Infection in Intensive Care Unit (EPIC) [5] and the Pfaller-Wenzel in the USA[6,] showed that Candida is the fifth most predominant nosocomial pathogen. Invasive Candidiasis causes a high crude mortalities usually between 50 and 60% in critically ill non-neutropenic patients, while related or attributable mortality varies from 21 to 38%[7,8]. Although Candida albicans remains the most prevalent species, there has been a clear shift towards non-albicans species[9,10] namely Candida tropicalis, Candida parapsilosis, Candida kruzei particularly found in the neutropenic patient and Torulopsis glabrata found especially in patients with solid tumor. Majority of studies were retrospective analyses but our study is prospective. The antimicrobial Resistance Surveillance program reported that the rank order of the various Candida non-albicans species differed among patients in various geographic locations, but the reason for such differences remains unclear[11]. The number of studies done in this subject is very little. Several retrospective studies have demonstrated that a number of predisposing factors for spread of candida infections in the ICU are total parenteral nutrition administration(TPN),use of multiple broad spectrum antibiotics, major surgeries, central venous catheter insertion , urinary catheter mechanical ventilation, persistent neutropenia, renal failure, glucocorticosteroid treatment ,burns, and hemodialysis.[12]

There are very few studies from India, especially Eastern India, on the pattern of fungal infections in Intensive Care Unit patients. The Sir Sunderlal Hospital, BHU, is one of the tertiary care centers catering a large number of patients from the states of Uttar Pradesh, Bihar, Madhya Pradesh, Jharkhand and Chhattisgarh. Critically ill patients from various departments like medicine, surgical, orthopedic, pulmonology, cardiac, gastroenterology, obstetrics and gynecology

are admitted in the intensive care unit (ICU). It is presumed that the findings of this study would faithfully reflect the pattern of fungemia in seriously ill patients from this part of our country, and to an extent that of developing nations like ours. According to surveillance data from the US Centres for Disease Control and Prevention, Candida now accounts for 12% of all hospital-acquired blood stream infection[13].

2. METHOD

A total of 412 patients study was conducted prospectively, patients were included both sex of age range lowest being 18 years and highest being 80 years during the period of June 2009 to October 2011 in the 16-bed medical surgical adult ICU of SSL Hospital, BHU,Varanasi, India. In the present study, 218(52.9 %) were males and 194 (47.1 %) were females, incidence of male ratio is high most probably due to working conditions outside the room, as in female usually they remain inside the room in Indian scenario. The cases were classified according to the responsible Candida species in the C. albicans and non-albicans candidemia groups in different clinical condition like in diabetes mellitus patient, TPN administration, neutropenic patient and cancer patient in the ICU. No informed consent was obtained from the individual patients whose data were analyzed in this observational, no interventional study. All decisions regarding diagnostic testing and treatment were made by the attending consultant.

Inclusion criteria:-If they had candida ICU-acquired blood stream, length of stay >48 h after ICU admission included in this study.

Exclusion criteria: - Immunosuppressed patients those with neutropenia (neutrophil count <1000/mm3) and/or those who treated with an antifungal drug before ICU admission and patients with a diagnosed candidemia before ICU admission not included in the study.

In the study all age group patients were included lowest being 18 years and highest being 80 years. 77.2% (i.e. 318 out of 412) of patients stayed for around 2-3weeks in ICU. Mean duration of stay of study population was 17.07 days. Candidemia was defined as at least one positive blood culture for Candida species in patients hospitalized for more than 48 h with signs or symptoms of infection.Candiduria was taken when there was presence of more than 100,000 cfu/mL of the same Candida species in two distinct urine samples obtained within 1 wk. All patients admitted to ICU were screened and presence of risk factors, like broad spectrum antibiotics > 3 days , duration of mechanical Ventilation, Malignancy, Diabetes mellitus, Neutropenia, Endotracheal intubation, Total parenteral Nutrition, Haemodialysis, and Central venous line sent samples for evaluatation for fungal infections. Any of the following samples were obtained, those associated with risk factors, blood at least from two sites (10 ml), endotracheal tube secretions collected in mucus extractor, bronchial aspirate, high vaginal swab, urine (50 ml), Central line tip were the samples under strict aseptic precautions sent for culture in Sabouraud dextrose broth in mycology division of the department of microbiology. Patient of diabetes mellitus , persistent neutropenia, those receiving TPN and cancer , request microbiologist to find out different type Candida species among candida positive patient.

After completion of the study, the data were entered into the statistical software package SPSS 16.

The recorded data was analyzed using chi-square test and Fisher's exact test. Data of this test was analyzed in MICROSOFT EXCEL 2007.SPSS VERSION 16.

RESULT

During the study period, 106 patients with candidemia were identified among 412 patients admitted to the ICU.Table 1 compares distribution of risk factors among Candidaemia patients. 124 patients were diabetic in them 20 were found to have Candida in blood (i.e. $P \leq 0.05$).36 patients had malignancy, among these 16 were having candida in their blood, P = 0.05 which was significant. 178 patients received total parenteral nutrition 58 patients were having Candidaemia, P = 0.049 which was significant. 4 were having HIV and all of them were having Candidaemia,p = 0.016 which was significant. 82 patients had neutropenia in them 36 were found to have invasive Candidiasis with P = 0.03 which is significant. The P value for all other risk factors like receipt of broad spectrum antibiotics (P = 0.24), Central line (P = 0.33), ventilator (P =

0.20), organ failure (P = 0.23), intubation (P = 0.64), hemodialysis (P = 0.91) came as insignificant i.e P> 0.05.

Table 2 shows the distribution of Candida species among culture positive patients. 106 of blood Candida isolates were 52 (49.0%) Candida tropicalis, 28 (26.6%) C. albicans, 14(13.5%) Candida gullerimondi, 8 (7.8%) Candida glabrata and 4 (3.8%) were Candida krusei.

Table 3 showed Candida tropicalis is common in patients receiving TPN (51.7%). C. albicans stands second (20.7%) followed by C. gullerimondii (17.2%). C. glabrata accounts for 6.9% of Candidaemia and C. krusei 3.5%.

Table 4 showed that C.tropicalis (35.0%) is common in diabetic patients, followed by C. albicans (30.0%). C. gullerimondi accounts for 20.0%, C.glabrata (10%), C. krusei(5%)of Candidaemia cases.

In neutropenic patients, C. tropicalis incidence is 44.4%. Strikingly C. gullerimondii incidence is more (22.2%) than C. albicans (16.7%). C. krusei accounts for 11.1% of cases and C. glabrata is present in 5.6% of neutropenic patients as shown in Table 5.

Table 6 showed C. tropicalis is present in 50% of cancer patients followed by C. gullerimondi (25.0%). C albicans and C. krusei account for 12.5% each. 4 HIV positive patients 3 were having C. glabrata and one was having C. tropicalis.

3. DISCUSSION:

The most common cause of invasive fungal disease is infection with Candida species. Candidiasis is the fourth common cause of nosocomial bloodstream infections worldwide, accounting for 9% of all such infections in the United State[1.]Despite the availability of an expanded antifungal medicine, the mortality associated with invasive Candida infections remains high, ranging between 19 and 49 %.[14,15]. In the present study of 106 (25.72%) patients out of 412 ICU patients with candidemia were identified. In which 106 of blood Candida isolates were 52 (49.0%) Candida tropicalis, 28 (26.6%) C. albicans, 14(13.5%) Candida gullerimondi, 8 (7.8%) Candida glabrata and 4 (3.8%) were Candida krusei .Among candida species predominance of candida tropicalis was more as compare to candida albicans in the patient of receiving TPN, diabetes, neutropenic and cancer patient as shown in Table2,3,4,5&6. Over the last decade, the proportion of infections caused by C albicans has fallen16[11], At the time of 1990, 80% of all fungal blood stream isolates were caused by C albicans[11]. Study from Italy ,candida albicans in decreasing pattern , decreasing from 62% of all candidal isolates in 1999 to only 24% in 2003[17].While C. glabrata, is increasing in trend surprising 0% in 1999 to 26% in 2003.[17] The authors noted a strong correlation of the rise in non-albicans species with the use of fluconazole[17]. Common risk factors predisposing to candidemia include candidal colonization, prior exposure to antibiotics, renal failure, presence of a central venous catheter, and need for total parenteral nutrition (TPN) [18, 19]. In a large 3.5-year multicenter prospective observational study of candidemia within the United States, Nguyen and colleagues[19] demonstrated that nearly 50% of all candidal isolates were of non-albicans species, with C. glabrata (6.3%) and C krusei (4.3%) representing over 10% of all cultures.

Although Candida albicans is the most commonly identified species, the incidence of infection with non-albicans Candida species are increasing[20].In a multicenter surveillance study conducted in the United States from 2004 to 2008, 46% of 2,019 bloodstream isolates[20] were C albicans, whereas 54% were non-albicans Candida species, including Candida glabrata (26% of all cases), Candida parapsilosis (16%), Candida tropicalis (8%), and Candida krusei (3%).·A program of epidemiology and fungal susceptibility performed in the USA, Canada and South America, called SENTRY,[21] demonstrated that 47% of the Candidaemia were caused by non-albicans species and that Torulopsis glabrata had become the second most frequent species causing Candidaemia. Invasive fungal diseases are known to cause significant morbidity and mortality in immunosuppressed patients, but the incidence of invasive fungal disease in immunocompetent critically ill patients is difficult to find out because of the lack of clear definitions and inaccurate diagnostic procedures. Mlinaric Missoni et al. from Croatia had reported the fungal incidence in tissue biopsy specimens of diabetic patients who had clinical evidence of fungal infections. The predominant isolates were C. parapsilosis (45.5%), C. tropicalis (22.7%), C. albicans (9.1%), and C. glabrata (9.1%). Bansal et al. from India had reported 9% isolation of fungi from superficial swabs taken from 103 patients

with diabetic foot wounds. The predominant species were C. tropicalis (29%), C.albicans (14%), and C. guillerimondii (7%), followed by Aspergillus flavus (21%), Aspergillus niger (14%), and Fusarium species (14%). In present study among candida species, C.tropicalis was also predominant species in diabetes patient. Non-C.albicans Candida species particularly C. glabrata and C. krusei were more common in haemodialysis recipients than in Candidaemic patients not receiving haemodialysis. Some studies showed that Candida glabrata is the predominant species in clinical materials of patients receiving total parenteral nutrition.

India scenario: The rise in frequency of infection of non-albicans Candida species has been also observed in tertiary care centres in India, with the isolation rate ranging from 50% to 96%.[22,23] [,24] Predominant isolation of C.tropicalis instead of C. glabrata or C .parapsilosis in all age groups in the Indian scenario is unique. Occasionally, outbreaks due to unusual yeast had also been observed in Indian hospitals, as in the Pichia anomala (C .pellicosa) outbreak in paediatric wards of PGI, Chandigarh during 1996-1997 [25]. Somansu Basu et al in 2003 reported that C. albicans was the predominant species isolated from all clinical specimens, species were identified, viz.,C. albicans (45.8%), C. tropicalis (24.7%), C. parapsilosis (10.5%), C. krusei(7.0%), C. kefyr (7.0%), C. guilliermondii (3.5%), and C. glabrata (1.1%).[26] C.glabtara was the most common species isolated in patients with diabetes mellitus and its frequency was significantly higher in them when compared to control group[27].some species show particularities as Candida tropicalis shows a higher invasive capacity and 50 to 60% of the colonized patients develop disseminated Candidiasis; Candida parapsilosis is associated with total parenteral nutrition and with central venous catheters and infection by this species is often not preceded by colonization. The incidence of invasive Aspergillosis is also increasing in the last two decades, namely in patients with haematological malignancies, bone-marrow or solid-organ transplantation, chronic corticosteroids users and even COPD patients. [28]

CONCLUSIONS

The continuously using of invasive monitoring and aggressive use of broad-spectrum antibiotics and surgical technologies in the ICU has not only improved survival of critically ill patients with life-threatening illnesses but has also increased the risk for fungal infections. Fungal infections can become severe and rapidly progressive and are often difficult to diagnose and treat. Although diagnostic and therapeutic modalities for some of the fungal infections are improving such as for C albicans**,** more studies are needed for non-albicans Candida species especially *C.tropicalis*.

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Table 1: Distribution of risk factors for Candidaemia.

Risk factor	No of patients (%)		P value
	All (n=412)	Positive Cases (n=106) =25.72%	
Central venous catheter ;yes/no	314(76.2) /98(23.8)	86(81.1)/40(18.9)	0.33
Ventilator; yes/no	370(89.8)/ 42(10.2)	200(94.3)/ 6(5.7)	0.20
Diabetes; yes /no	124(30.1)/ 288(69.9)	20(18.9)/ 86(81.4)	0.04*S
Malignancy; yes/no	36(8.7)/ 376(91.3)	16(15.1)/ 90(84.9)	0.05*S
Neutropenia; yes/no	82(19.9)/ 330(80.1)	36(34.0)/ 70(66.0)	0.003* S
Endotracheal tube; yes/no	366(88.8)/ 46(11.2)	96(90.6)/ 10(9.4)	0.64
Total parenteral nutrition; yes/no	178(43.2)/ 234(56.8)	58(54.7)/ 56(45.3)	0.049*S
Hemodialysis;yes; yes/no	72(17.5)/ 340(82.5)	18(17.0)/ 88(83.0)	0.91
HIV; yes/no	4(1.0)/ 408(99)	4(3.8)/ 102(96.2)	0.016*S
Broad spectrum antibiotics; yes/no	392(95.1)/ 20(4.9)	104(98.1)/ 2(1.9)	0.24
Organ failure; yes/no	306(59.7)/ 166(40.3)	56(52.8)/ 50(47.2)	0.23
Tracheostomy; yes/no	138(33.5)/ 274(66.5)	40(37.7)/ 66(62.3)	0.45

*S-Statistically Significant (P<0.05)

Table 2: Distribution of Candida species isolates recovered from blood

Candida species	No. of cases (n = 106)	Percentage (%)
Candida albicans	28	26.6
Candida tropicalis	52	49.0
Candida gullerimondi	14	13.5
Candida glabrata	8	7.8
Candida krusei	4	3.80

Table 3: Distribution of Candida species among patients receiving TPN
(Positive 58 out of 178 patient)

Candida species	Number of cases	%
Candida tropicalis	30	51.7
Candida albicans	12	20.7
Candida glabrata	4	6.9
Candida gullerimondi	10	17.2
Candida krusei	2	3.5
Total	58	100.0

Table 4: Distribution of Candida species among Diabetics patient
(Positive 20 out of 120 patients)

Candida species	Number of cases	%
Candida albicans	6	30
Candida tropicalis	7	35
Candida glabrata	2	10
Candida gullerimondi	4	20
Candida krusei	1	5
Total	20	100.0

Table 5: Distribution of Candida spp among neutropenic patients
(36 candida positive patient out of 82 patients)

Candida species	**Number of cases**	**%**
Candida tropicalis	16	44.4%
Candida gullerimondi	8	22.2%
Candida albicans	6	16.7
Candida glabrata	2	5.6
Candida krusei	4	11.1
Total	36	100.0

Table 6: Distribution of Candida species among cancer patients
(Candida positive 16 out of 36)

Candida species	Number of cases	%
Candida albicans	4	25
Candida glabrata	0	0
Candida gullerimondi	4	25.0
Candida krusei	2	12.5
Candida tropicalis	6	37.5
Total	16	100.0