ISSN: 2091-2331 (PRINT) 2091-234X (ONLINE)

Journal of Nobel Medical College
Available Online: www.nepjol.info, www.nobelmedicalcollege.com.np
Volume 5, Number 1, Issue 8, January-July 2016, 61-65

Original Article

Characterisation and Isolation of Candida Species from ICU Patients in Nobel Medical College Teaching Hospital, Biratnagar

Bigu Kumar Chaudhari, Ganesh K.Singh, Kamal Prasad parajuli, Kewal Shrestha, Dharmendra Shah
Department of Microbiology, Nobel Medical College Teaching Hospital, Biratnagar
Received: 27th May, 2016; Revised after peer-review: 24th June, 2016; Accepted: 13th July, 2016

Abstract

Background

Fungi have emerged as major causes of human diseases. Intensive Care Units (ICU), harbor almost all the risk factors for opportunistic fungal infections. Among these, Candida infections are very common with recent trends being rise in the non-Candida albicans (NCA) species along with an increase in resistance of these species to antifungal drugs. Increament in invasive Candidasis during last three decades have been reported in several studies, among all Candida species the Candida albicans is considered as the most common infectious agent the other non-albicans like C. Tropicalis, C. glabrata, C. parapsilosis C. krusei, and C. dubliniensis were raised as infectious agents. The aim of current study is to characterize the candida species from the clinical specimens of patients admitted in the ICU of Tertiary Care hospital in Eastren Nepal and to perform their antifungal susceptibility.

Material and Methods

The study was carried out in the tertiary care hospital Nobel Medical College and Teaching Hospital Biratnagar Nepal over a period of 6 monthsbetween September 2015 to February 2016. The following techniques were employed to characterize the isolates in the study – Gram's stain, culture on Sabouraud's Dextrose Agar, Germ Tube test, morphology in Cornmeal Agar and chromogenic agar media, sugar fermentation and sugar assimilation tests, and the results were interpreted by using standard protocols.

Results

Out of 50 candida isolates from different clinical samples including 37 blood samples, 9 urines and 4 Endo Tracheal (ET) tube, the most common species was C.albicans (44%) followed by C.tropicalis (26%), C.Glabrata (18%), C.Parapsilosis (08%), C.Krusei (02%), and C.Dubliniensis(02%).

Conclusion

The purpose of the study is to show the value of species isolation, identification and antimicrobial sensitivity testing of the ICU & NICUs patients.

Key Words: *Antifungal susceptibility, Candida species, Candida bloodstream infections (BSIs), Intensive Care Unit, Non- Candida albicans species.*

Introduction

For over 20 years, Candida bloodstream infections (BSIs) have been increasing significantly worldwide, representing an important infective complication in hospitalized patients with medical and

Correspondence Address:** ***Bigu Kumar Chaudhari, Lecturer | E-mail: biguchaudhari@gmail.com

surgical disorders [1]. Candida spp. especially Candida albicans consider as one of the most common cause of fungal infections leading to a range of life-threatening invasive too non-lifethreatening mucocutaneous diseases [2]. According to nosocomial Infection Surveillance systems of the United State, candida spp. is the 7th most common nosocomial pathogens [3]. There are several Candida species involve in causing disease, but the most commonspecies is Candida albicans. Many other documents and records are showing the increasing incidence of other non-albicans among hospitalised and immune suppressed patients. The emergence of this opportunistic pathogen is favoured by the change in the most susceptibility due to the growing number of immune compromised individuals in the population as a result of HIV pandemic and the use of long-term immunosuppressive therapy in cancer and organ transplant patients [4]. Candida infection in hospitalized patient is increasing significantly over the last 10 years. Particularly the patients in the Intensive Care Unit who have invasive monitoring lines. Candida infection leading to prolonged hospitalization and significant mortality which dictate the need to take all the possible measures to prevent this infection particularly at the high risk patients.

Among the ICU patients Candida bloodstream infection has become the fourth common organism because of the invasive infection in critical patients) [5,6]. In recent year there has been increase in isolation of other species from neonate's patients but still the Candida albicans is the most common one with invasive Candidiasis [7]. The causative agents of bloodstream infection is most commonly by Candida albicans with prevalence rate is near by 50%) [8]. The increasing nosocomial UTI subgroups is because of fungal infection and all fungal UTI are due to Candida species) [9].

Materials and Methods

The study was undertaken with the Candida isolates obtained from clinical specimens of patients admitted in the ICU for a period of 6 months from September 2015 to February 2016, in the Department of Microbiology in a tertiary care hospital Nobel Medical College and Teaching Hospital Biratnagar Nepal, It has a 30 bedded ICU. The study was recruited after approval of Institutional Review Committee (IRC).

Selection of cases & inclusion criteria:

1. patients with different indwelling devices admitted in ICU.
2. patients with long term corticosteroids therapy like immune compressive and HIV.
3. premature low birth baby admitted in ICU
4. patient with major surgery admitted in ICU.

Exclusion Criteria

1.Those clinical samples which shows growth other than Candida from ICU patients.

2. Patients with antifungal therapy.

Microbiological records of ICU from during the period of 6 months were reviewed to identify the Patients with positive candida cultures. Sample collected were used to inoculate on Sabourad Dextrose Agar with chloramphenicol slant and for direct microscopy with Gram's stain. Culture growths were subjected to Germ tube test and corn meal agar(CMA) morphology (Dalmau technique) & CHROM agar [10]. Carbohydrate assimilation & fermentation test to differentiate between Candida albicans and non-albicans groups. The susceptibility to antifungal drugs of Candida isolates was determined by using the Disk Diffusion method described by the Clinical and Laboratory Standards Institute (CLSI, USA), document M44-A [11]. The

specimen collected was blood, ET tube and urine. Positive Candida cuture patient's data, clinical features, risk factors, and results were taken for further management.

Results

A total of 186 samples were received, from the ICU of our hospital during the study period. Out of these 50 samples showing growth of candida were included. In this study, among the 50 Candida species isolated, C. Albicans were more in number than NCA species. Out of 50 candida isolates from different clinical samples including 37 blood samples, 9 urines and 4 Endo Tracheal (ET) tube [table no1], the most common species was C. albicans (44%) followed by NCA species C. tropicalis (26%), C. Glabrata (18%) %), C. Parapsilosis (08%), C. Krusei (02%), and C. Dubliniensis (02%) [table no 2].

The highest number of samples were received in the age group of 0-2 Month (42%), followed by the age groups of 3-11 Month (04%), 1-5 year (04%), between 6-12 year (04%), between 13-19 year(02%), Between 20-30 year(08%) Between 31-40 year(10%) Between 42-50year(10%) Between 51-60 year (08%), Between 61-70 year (08%) [table no 3].

Candida species were isolated more in males (54%) than females (46%) in the present study. [Table 4] Candida species were isolated mainly from blood samples (74%), followed byurine(18%) and ET tube (8%). The distribution of candida spp. From various clinical samples is in [Table No. 6]

Table 1: Distribution of samples showing growth of candida species

Samples	NO- %
Blood	37 (74%)
Urine	9 (18%)
E .T tube	4 (8%)
Total	50

Table 2: Species of candida isolated

Candida spp.	NO of isolates (%)
C.albicans	22 (44%)
C.tropicalis	13 (26%)
C.parapsilosis	04 (08 %)
C.glabrata	09 (18%)
C. krusei	01 (02 %)
C. dubliniensis	01 (02 %)
Total	50

The most common species of candida isolated was C.albicans forming 44 % of the total isolates . The non-albicans candida species form the remaining 56 % of the total isolates.

Table 3: Age, sex-wise and species-wise distribution of C.species

Age (month & year)	No (%)
Between 0-2 Month	21(42%)
Between 3-11 Month	02 (0 4%)
Between 1-5 year	02 (04%)
Between 6-12 year	02 (04%)
Between 13-19 year	01 (02 %)
Between 20-30 year	04 (08 %)
Between 31-40 year	05 (10%)
Between 42-50year	05 (10%)
Between 51-60 year	04 (08%)
Between 61-70 year	04 (08%)
Total	50

Table 4: Sex wise

Males No (%)	Females No (%)
27 (54%)	23(46%)

Table No 5: Distribution of Candida spp. From various clinical samples

Candida spp.	Blood	Urine	E. T.Tube	Total (%)
C. albicans	19(51.3 %)	02(22.2 %)	01(25 %)	22(44 %)
C.tropicalis	09 (24.3%)	03(33.3 %)	01(25 %)	13(26 %)
C.glabrata	07(18.9 %)	01(11.1 %)	01(25 %)	09(18 %)
C.parapsilosis	01(02.7 %)	02(22.2 %)	01(25 %)	04(08 %)
C. dubliniensis	01(02.7 %)	-	-	01(02 %)
C. krusei	-	01(11.1 %)	-	01(02 %)
Total	37	09	04	50

Discussion

Candida species are the most common cause of fungal infections, leading to a range of life-threatening invasive too non-life-threatening mucocutaneous diseases. The species are endogenous in nature and are usually responsible for opportunistic infections. In addition, using several broad spectrum antibiotics, immunosuppressive and corticosteroids drugs in ICUs and NICUs wards, were increased candiduria. During the several last decades, an increasing in several opportunistic fungal infection was observed.

This is newly recognized opportunistic pathogen that has been linked too oropharyngeal Candidiais in HIV-infected patients. It has also been observed in blood isolates from bone marrow transplant patients, denture wearers, cancer patients, infants and elderly, oral and vaginal isolates from non-HIV-infected patients [5,6,7]. In this study total 186 patient's samples were screened, out of these 50 were positive for Candida. Candida was mainly isolated from blood (74%) and urine (18%) & ET Tube 8%. Candida albicans, Candida tropicalis, Candida glabrata causes 44%, 26% and 18% of the infections respectively. Our study shows Candidaalbicans is the most predominant one. Candida species are the 4th most causative agents of bloodstream infection in hospitalised patients due to the normal microbes of human, which enhance to colonise the implanted devices of the host [12]. Although there is low sensitivity of conventional culture to detect yeast but the invasive nature of the fungi causing opportunistic infecting which causes morbidity and mortality in hospitalized patients. The main aim of our study is to show the morbidity of fungal infection in ICU patients [13]. The advance technique of modern medicine is able to treat many kinds of disease. The incidence of nosocomial infection of the fungi have been increasing due to immunosuppressive drugs,use of broad-spectrum antibiotics, indwelling devices, prolonged stay in the ICU, intra-abdominal surgery, immuno-compromised and immunosuppressive drugs[14].Various reports regarding the prevalence of Candida albicans and non-Candida albicans bloodstream infection have been reported from India. From the neonates Candida albicans as the most common isolates Candida few case have documented [15,16]. Candida bloodstream infection has become challenge in critically ill patient due to immuno-compromised, diabetes, intravenous cannulae, and intravenous drug use) [17]. Candidiasis is most commonly seen in ICUs patients. C. albicans is the predominant cause of Candida bloodstream infection [18]. The major component of cell membrane is phospholipids which is catalyses by phospholipase which acts as a virulent factor and due to this there was hydrolysis of phospholipids and lyses of cell [19]. Invasive fungal infections caused by a variety of fungal species are becoming an increasing problem worldwide. In particular, Candida species have become the seventh most common cause of nosocomial sepsis in children [20].ICU patients are at higher risk for Invasive Candidiasis than patients the general ward patients [21].Among the known risk factors for Invasive Candidiais, colonization is the most important one as it indicates that patients have an endogenous source of candida [22].The present study highlights the change in epidemiology in the species distribution of Candida and also highlights a rise in the infections by NCA species as compared to those by C. albicans, along with an increase in resistance of these NCA species to the routinely used antifungals. Therefore, knowledge of the local species distribution of Candida through presumptive identification, followed by confirmation, is essential to initiate early empirical therapy,

especially in an ICU setup, which harbors a lot of immunocompromised patients and other patients susceptible for acquiring various fungal infections. Moreover, antifungal susceptibility testing, though not as routinely performed as antibacterial susceptibility testing, should be carried out, along with MIC values of the isolates for the various groups of antifungals, in order to optimize therapy and outcome.

Conclusion

This study demonstrates the importance of species identification and antifungals usceptibility testing for hospitalized patients in ICUs and NICUs wards. In the NICU, it is associated with a very high mortality, especially with antibiotic uses. Major risk factors are prolonged ventilation (>7 days). Antenatal care has a protective impact on neonatal fungal infection in our settings.

References

[1] D. Diekema, S. Arbefeville, L. Boyken, J. Kroeger, and M. Pfaller, The changing epidemiology ofhealthcare-associated candidemia over three decades, Diagnostic Microbiology and Infectious Disease. 73 (2012) 45–48.

[2] Achkar JM, Fries BC, Candida infections of the genitourinary tract, Clin Microbiol Rev. (2010) 253-73.

[3] Saha R, Das SD, Kumar A, et al., Pattern of Candida isolates in hospitalized children, Indian JPediatr. 75 (2008) 858-60.

[4] Raut SH, VaraiyaA, Differentiation of candida dublinenesis on Chrom Agar & Pals agar JMM.10 (2009) 55-58.

[5] Shaseen MA, Taha M, Species identification of Candida Isilates obtained from oral lesion ofhospital and non-hospitalised patient with oral candidiasis, Egyptain Dermatology Online Journal.2 (2006).

[6] Eggimann P, Garbino J, Pittet D, Epidemiology of Candida species infections in critically ill non-immunosuppressed patients, Lancet Infect Dis. 3 (2003) 685-702.

[7] Ostrosky-Zeichner L, Pappas PG, Invasive candidiasis in the intensive care unit, Crit Care Med.34 (2006) 857-63.

[8] Avila-aguero M.L, Canas-Coto A, Ulloa-Gutierrez R, Caro M.A, Alfaro B, Paris M.M, Riskfactors for Candida infections in a neonatal intensive care unit in Costa Rica, International Journal of Infectious Diseases. 9 (2005) 90-95.

[9] Bukhary ZA, Candiduria: a review of clinical significance and management, Saudi J Kidney DisTranspl.19 (2008) 350-60.

[10] Horvarth LL, Hospenthal DR, Murray CK,Dooley DP, Direct isolation of Candida spp,From bloodcultures on the chromogenic mediumHROMagar Candida, J Clin Microbiol.41 (2003) 2629-32.

[11] Clinical and Laboratory Standards Institute/National Committee for Clinical LaboratoryStandards, Method for Antifungal Disk Diffusion Susceptibility Testing of Yeasts: ApprovedGuideline, Document M44-A, Wayne, PA: Clinical and Laboratory Standards Institute,(2004).

[12] Vinitha M, Ballal M, Biofilm as virulence marker in Candida isolated from blood, World J MedSci. 2:1 (2007) 46-8.

[13] Meersseman W, Lagrou K, Maertens J, Van Wijngaerden E, Invasive aspergillosis in theintensive care unit, Clin Infect Dis.45 (2007) 205–16.

[14] Kam LW, Lin JD, Management of systemic candidal infections in the intensive care unit, Am JHealth Syst Pharm.59 (2002) 33-41.

[15] Narain S, Shastri JS, Mathur M, Mehta PR, Neonatal systemic candidiasis in a tertiary care Centre, IJMM. 21:1 (2003) 56-8.

[16] Narang A, Agarwal PR, Chakrabarti A, Kumar P, Epidemiology of systemic candidiasis in a tertiarycare neonatal unit, J Trop Pediatrics. 44:2(1998) 104-8.

[17] Pfaller MA Nosocomial candidiasis: Emerging species, reservoirs and modes ofTransmission, Clin Infect Dis. 22 (1996) 89-94.

[18] Edmond MB, Wallace SE, McClish DK, Pfaller MA, Jones RN, Wenzel RP, Nosocomialbloodstream infections in United States hospitals: a three-year analysis, Clin Infect Dis.29(1999) 239-244.

[19] Mohan das V, Ballal M Proteinase and phospholipase activity sas virulence factors in Candida species isolated from blood, Rev IberoamMicol. 25 (2008) 208-210.

[20] Kumar D, Kumar A, Singh S, Tilak R , Candidemia-induced pediatric sepsis and its association with free radicals, nitric oxide, and cytokine level in host, J Crit Care. 30 (2015) 296–303.

[21] Wisplinghoff H, Bischoff T, Tallent SM, Seifert H, Wenzel RP, Edmond MB, Nosocomial bloodstream infections in US hospitals: analysis of 24,179 cases from a prospective nationwide surveillance study, Clin Infect Dis. 39:3 (2004) 309–17.

[22] Miranda LN, van der Heijden IM, Costa SF, Sousa AP, Sienra RA, Gobara S, et al., Candida colonisation as a source for candidaemia, J Hosp Infect. 72:1 (2009) 9–16.