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RESEARCH ARTICLE

Epidemiology of *Candida* colonization in medical surgical intensive care unit of a tertiary care teaching hospital of North India

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ABSTRACT

Objective: Invasive candidiasis is associated with increased morbidity and mortality in critically ill patients. Current study was undertaken to study the colonization trend in critically ill patients admitted to a medical /surgical ICU of a tertiary care teaching hospital.

Methodology: Data for the current study has been derived from a larger database generated for external validation of risk prediction scores for invasive candidiasis conducted in a 12 bedded medical/surgical ICU of a tertiary care hospital of North India. Non neutropenic adult patients with >48 hours of ICU stay were included in the study. Colonization surveillance samples were collected from oral cavity, endotracheal aspirates, axilla, perineum and urine at the time of admission and then on 3rd, 7th, 14th and 21st day of ICU stay. Blood culture samples were taken at admission and then as per physician's discretion.

Results: Total 200 patients were enrolled from July 2013 to November 2014. Ninety five percent patients were colonized with *Candida* either at admission or during their stay in ICU. The most common species responsible for colonization was *Candida glabrata* (27%) followed by *C. tropicalis* (20.5%) and *C. auris* (18%). Seventeen patients developed *Candida* blood stream infection. *C. tropicalis* was the most common species causing candidemia. C.auris was most frequent colonizer of axilla (54.2%), while rectal swabs had high growth of *C. glabrata* (44.9%).

Conclusion: Our study population had high rate of *Candida* colonization. *C. glabrata* was the most common colonizer followed by *C. tropicalis*. *J Microbiol Infect Dis 2018; 8(4):147-152.*

Keywords: *Candida auri*, *Candida* colonization, *Candida glabrata, Candida tropicalis;* critically ill

INTRODUCTION

Healthcare associated infections are a major cause of morbidity and mortality in hospitalized patients [1]. Recent literature has shown carbapenem resistant *Enterobacteriaceae*, *Candida* and *Clostridium difficile* are the three most common emerging hospital acquired pathogens [2,3]. Progressive colonization is an important step leading to subsequent invasive *Candida* infection in the predisposed host [4]. In their landmark paper, Pittet et al. designed colonization index (based on number of sites colonized and density of colonization) as a prediction tool for subsequent invasive candidiasis in critically ill patients [5]. Since then *Candida* colonization has been studied extensively by various authors [6]. Leon et al. developed and validated "*Candida* score"as a prediction tool for identifying patients at risk for invasive candidiasis [7,8]. The score was calculated by giving one point each to multifocal colonization, total parental nutrition, and surgery while severe sepsis was given two points. A score equal or above 3 was considered high risk for invasive candidiasis.

There are also reports of *Candida* co-colonization with multidrug resistant organisms in patients exposed to broad spectrum antibiotics [9].Respiratory tract colonization with *Candida* has been shown to be associated with multidrug resistant ventilator associated pneumonia and poor outcome [10,12].

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Though *Candida* is considered to be a normal flora of human gut, the mechanism of transition from being commensal to pathogen remains poorly understood. Analysis of the various risk factors for invasive candidiasis suggests that it is probably the density of colonization which once crosses a critical threshold in a predisposed host plays the key role in pathogenesis of invasive candidiasis. Lau et al. showed that colonization of two or more sites and heavy colonization are independent risk factors significantly associated with invasive candidiasis [12].

There is limited literature in the field of *Candida* colonization in critically ill patients from tropical countries. Current study was undertaken to study the trend and pattern of *Candida* colonization and bloodstream infection in critically ill patients admitted to a medical surgical intensive care unit of a tertiary care teaching hospital of North India.

METHODS

Study setting and Patient population

Data for current study has been collected from a larger database generated for an external validation study of risk prediction scores for invasive candidiasis done at Department of Critical Care Medicine, Sanjay Gandhi Institute of Postgraduate Medicine (SGPGIMS), Lucknow under Senior Research Associateship (SRA) program of Council of Scientific and Industrial Research (CSIR), New Delhi. The external validation of risk prediction scores for invasive candidiasis has already been published elsewhere [13].

Sanjay Gandhi Institute of Postgraduate Medicine is a 1050 bedded tertiary care teaching hospital in North India. The Department of Critical Care Medicine runs a 12 bedded medical/surgical ICU. The patient population comes from all over Lucknow and nearby area. We admit patients from other ICUs within the institute as well as other hospitals.

From July 2013 to November 2014, 200 patients were enrolled into a prospective observational study. Patients with neutropenia (absolute count $<0.5 \times 10^9$/l), hematological malignancy, bone marrow transplantation, age <18 years and ICU stay less than 48 hours were excluded. Informed consent was taken from the patient or nearest kin of the patient.

Sample collection and laboratory method

Surveillance samples (oral swab, urine, tracheal aspirate, axillary swab and rectal/perinealswab) were collected at the time of admission, on 3rd, 7th, 14th and 21st day of ICU stay. Oral sample was collected by sweeping the swab stick over anterior fauces. Urine was collected from Foley's catheter under strict asepsis. Endotracheal aspirate was collected via mucous extractor attached to suction apparatus. Axillary and rectal/perineal (area just around the anus) samples were collected by sweeping the swab stick in the respective area. Paired blood cultures were also sent at admission and then as per decision of the treating physician.

To test the presence of *Candida*, a loop full of specimen was streaked on Sabouraud dextrose agar (SDA) plate and incubated at 37 ^{0}C. Plates were examined for growth of *Candida* at 24 and 48 hrs. Blood cultures were done using automated BACTEC system (Becton Dickinson Diagnostic Instrument System).Phenotypic species identification was done using germ tube testing, sugar assimilation, CHROM agar and Tetrazolium Reduction medium (TRM).

MALDI-TOF (Matrix Assisted Laser Desorption Ionization -Time of Flight) was not available in our institute at the time of the study, therefore isolates were sent to Microlab, Coimbatore for confirmation of species after phenotypic identification. Due to limited finances we could not sent all the samples, so isolates from blood and 200 isolates from other sites with doubtful phenotypic identification (as guided by our Microbiology experts) were sent for MALDI-TOF(Bruker Daltonik MALDI Biotyper) analysis for confirmation of the *Candida* species.

Definition

Candida colonization was defined as positive culture of any of the surveillance sample for any *Candida* species. Candidemia was defined as one or more positive blood culture for any *Candida* species. Multispecies candidemia was defined as growth of more than one *Candida* species from same blood culture bottle or samples collected within 72 hours [14].

Statistical method

SPSS version 17 was used for statistical analysis. Categorical variables were expressed

as frequencies and percentages. Continuous variables expressed as means with standard deviation when the distribution was Gaussian and medians with interquartile range when the distribution was non-parametric.

Ethical issues

The study was approved by the ethics committee of the institute.

RESULTS

There were 305 admissions to the ICU during the study period, out of which a total of 200 patients qualified the inclusion criteria. Mean age of the patients were 45 ± 16.7 years. There were 124 males and 76 females (Table 1). Median duration of hospitalization before ICU admission was 7 days with IQR of 3-13 days.

Candida colonization

Two thousand six hundred and forty four (2644) samples from oral cavity, endotracheal aspirate, urine, rectal and axillary swabs were tested during the study period. One hundred ninety one (95%) patients were colonized with *Candida* species either at admission or during their stay in ICU. One hundred sixty two (81%) patients had multifocal (more than one site colonization).

Site specific species distribution

C. glabrata (27%) was the most common species responsible for colonization followed by *C. tropicalis* (20.5%) and *C. auris* (18%) (Table 2).

Table 1. Demographic data of the patients.

Variable	Total population, n=200
Age*	45.5 (30-60)
Medical	166 (83%)
Surgical	34 (17%)
SOFA^ at ICU admission*	10 (7-13)
APACHE** II at ICU admission*	17(12-22)
Days of ICU stay*	12.5(7-29)
Days of Hospital stay*	23 (15-46)
Days of hospitalization before ICU# admission*	7 (3-13)
Survivors at 28 days	114 (57%)
Survivors at discharge from ICU	102 (51%)
Duration of antibiotic therapy*	22.5 (15-40)
Abdominal surgery	29 (14%)
Diabetes Mellitus	41 (20%)
Pancreatitis	30 (15%)
Organ transplant	9 (4.5%)
Total parenteral nutrition/Partial parenteral nutrition	130 (65%)
Number of invasive lines and catheters*	4 (3-4)
Septic shock at admission	139 (69.5%)
Total duration of septic shock in days*	5 (2-9)
Use of mechanical ventilation	181 (90%)
Duration of mechanical ventilation in days*	12 (6-25)

*Inter-quartile range 25-75, **Acute Physiology and Chronic Health Evaluation, ^Sepsis related organ failure assessment score, # Intensive care Unit,

Table 2. Site specific species distribution of *Candida*.

Species	No. of Oral cavity isolates	No. of Urine isolates	No. of Endotracheal aspirate isolates	No. of Axillary swab isolates	No. of rectal swab isolates	Total (No. of isolates)
C. albicans	85 (21.6%)	12 (5.6%)	8 (10.9%)	10 (4%)	31(8.2%)	146 (11%)
C. tropicalis	92 (23.4%)	69 (32%)	22 (30%)	18 (7%)	67 (17.7%)	268 (20.5%)
C. parapsilosis	71 (18%)	28 (13%)	15 (20.5%)	38 (15.5%)	30 (7.9%)	182 (13.9%)
C. glabrata	81(20.6%)	57 (26.5%)	18 (24.6%)	27(11%)	170 (44.9%)	353 (27%)
C. auris	26 (6.6%)	29 (13.5%)	10 (13.6%)	133 (54.2%)	37(9.7%)	235 (18%)
C. krusei	16 (4%)	7 (3.2%)	0	0	12 (3.1%)	35 (2.6%)
C. keyfr	5 (1.3%)	6 (2.7%)	0	1 (0.04%)	5 (1.3%)	17 (1.3%)
C. rugosa	3 (0.7%)	5 (2.3%)	0	18(7.3%)	20 (5.2%)	46 (3.5%)
C. lusitaniae	3 (0.7%)	1 (0.4%)	0	0	3 (0.7%)	7 (0.5%)
C. fabianii	1 (0.2%)	1 (0.4%)	0	0	0	2 (0.15%)
C. guilliermondii	0	0	0	0	1 (0.26%)	1 (0.07%)
Unidentified	10 (2.5%)	0	0	0	2 (0.5%)	12 (0.9%)
Total	393	215	73	245	378	1304

Table 3. The percentages of Candida colonization trend at various sites.

Samling day	Oral colonization	Endotracheal colonization	Axilla colonization	Rectal colonization	Urine colonization	All sites colonization
Admission	73	25	29	53	32	44
Day 3	72	32	38	68	36	51
Day 7	51	19	43	68	43	47
Day 14	49	9	44	67	46	45
Day 21	27	2	41	53	39	34

We found site preferences by some species. Samples collected from axillary skin showed very frequent growth of *C. auris* (54.2%), while rectal swabs had high growth of *C. glabrata* (44.9%).Among the species isolated from oral secretions *C. tropicalis* (23.4%), *C. albicans* (21.6%), *C. glabrata* (20.6%) and *C. parapsilosis* (18%) were the most common isolates. *C. tropicalis* (32%) and *C. glabrata* (26.5%) were more frequent than other species in urine sample.In endotracheal aspirate commonest isolates were *C. tropicalis* (30%), *C. glabrata* (24.6%) and *C. parapsilosis* (20.5%).

Colonization trend

Oral colonization decreased from 73% to 27% during three weeks ICU stay (Table 3). Urine colonization increased from 32% to 46% from admission to 14 days then dropped to 39% on 21 days. Endotracheal colonization was highest on third day of ICU admission but declined steadily over next three weeks. Axillary colonization increased from 29% at admission to 43% on day 7, and then remained static for next two weeks. Rectal colonization ranged from 53% to 68% during three weeks period.

Candidemia

Seventeen patients developed bloodstream *Candida* infection during their stay in ICU (Table 4). Out of these, three patients had multispecies candidemia. *C. tropicalis* was the most species responsible for candidemia. Thirteen patients had first blood culture positive at the time of admission while rest of the four showed positive culture during their stay in ICU on 13th, 15th, 21st, 39th day of ICU admission respectively.

Antifungal prescription

Out of 200 patients 131 received antifungal therapy during their stay in ICU. Indication for starting antifungal therapy was prophylaxis (risk factor driven) in 25 patients, empirical (fever driven) in 90 patients and targeted (microbiology driven) in 6 patients. For 10 candidemia patients, empirical therapy was starting at the time of drawing blood culture sample and was later modulated as per the culture sensitivity report once the blood culture came positive.

DISCUSSION

We found a very high rate of *Candida* colonization (95% unifocal and 81% multifocal) in the study cohort. Various factors predisposing for such high rate could be broad spectrum antibiotic use, average duration of Pre-ICU hospitalization being 7 days (IQR 3-13), tropical climate etc. Moreover, as discussed earlier our ICU is a tertiary ICU and we take patients from other HDUs and ICUs within the hospital as well as from nearby hospitals. Most of these patients are already on broad spectrum antibiotics by the time they reach us.

Schulte et al studied 485 patients at hospital admission and found that patients who were able to ambulate were less likely to be colonized with *Candida* (odds ratio; OR=0.45, 95 % CI: 0.27–0.73) and patients with history of fluoroquinolones use (OR=3.01, 95 % CI: 1.80-5.01) were more likely to be colonized with *Candida* [9]. Rate of *Candida* colonization in the study population was 19%.

To the best of our knowledge, largest study in this field was conducted by Lau et al in seven Australian ICUs [12]. Out of 6,015 patients included in the study, fifty eight percent (n=3511) patients were colonized with *Candida* at the time of first sample collection (at 72hrs following ICU admission) and 48% (n=1671) had multifocal colonization. *C. albicans* (81%) was the most frequent colonizer. These findings are in contrast to our study, where we found *C. albicans* forming only 11% of the isolates. Our findings confirm the high prevalence of non-albicans *Candida* in Indian subcontinent. *C. tropicalis* was the most common species causing blood stream infection in our study population. These findings are in agreement with Chakarbarti et al. who reported *C. tropicalis* as the most common cause of candidemia in a study including 1400 candidemia episodes in 27 ICUs across the country [15].

C. glabrata was the most frequent colonizer of the gut while C.auris preferentially colonized the axilla. The site specific predilection for these species could be due to expression of certain types of receptors on the host surface or due to favourable host environment at these sites. Site specific predilection by *Candida* has never been reported earlier. This could be an important finding because local application of antifungal can bring down colonization at specific sites. Further research in this field can bring new insight into colonization pattern of *Candida*.

Regarding colonization trend we found site related variation. Oral care with Chlorhexidine mouth wash is routine practice in our ICU. This probably explains the improvement in oral as well as endotracheal *Candida* colonization over three weeks from admission. High rectal/perineal and axillary colonization represents the areas frequently missed in patient care and may require special attention as a source of endogenous infection.

Strengths and limitations

This is the first study showing *Candida* colonization trend in critically ill patients admitted to adult medical surgical ICU from a tropical country. The study highlights the importance of cleaning of axilla and perineal area to break *Candida* colonization. The study is limited by its single centre design and microbiological technique in species identification. As already mentioned we could send only 200 isolates (including all blood stream isolates) for MALDI-TOF analysis.

Another limitation of the study was inability to analyse the risk factors for colonization due to almost universal colonization of critically ill patients (95%) at admission or during their stay in ICU.

Besides this our data should be interpreted keeping in mind the frequent use of antifungal agent as prophylaxis, empirical or targeted regimen.

Conclusion

Rate of *Candida* colonization was high in our study population. *C. tropicalis* was the most frequent isolate from the blood. *C. glabrata* was

the most common colonizer followed by *C. tropicalis*. *C. glabrata* preferentially colonized rectum/perineal area while *C. auris* showed preference for axilla in our study population.

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Declaration of Conflicting Interests: The authors declare that they have no conflict of interest.

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