Journal of Hospital Infection 85 (2013) 60–65

Available online at www.sciencedirect.com

Journal of Hospital Infection

journal homepage: www.elsevierhealth.com/journals/jhin

Possible transmission of *Candida albicans* on an intensive care unit: genotype and temporal cluster analyses

F. Hammarskjöld [a,b,*], S. Mernelius [c], R.E Andersson [d], S. Berg [e], H. Hanberger [b], S. Löfgren [c], B.-E. Malmvall [b,f], M. Petzold [g], A. Matussek [c]

[a] *Department of Anaesthesia and Intensive Care, Ryhov County Hospital, Jönköping, Sweden*
[b] *Division of Infectious Diseases, Department of Clinical and Experimental Medicine, Faculty of Health Science, Linköping University, Sweden*
[c] *Microbiology Laboratory, Department of Laboratory Services, Division of Medical Services, Ryhov County Hospital, Jönköping, Sweden*
[d] *Department of Surgery, Ryhov County Hospital, Jönköping, Sweden*
[e] *Division of Cardiovascular Anaesthesia, Department of Medical and Health Science, Faculty of Health Science, Linköping University, Linköping, Sweden*
[f] *Futurum the Academy for Health Care, Jönköping County Council, Jönköping, Sweden*
[g] *Centre for Applied Biostatistics, Sahlgrenska Academy, University of Gothenburg, Sweden*

ARTICLE INFO

Article history:
Received 13 November 2012
Accepted 3 June 2013
Available online 5 August 2013

Keywords:
Candida
Intensive care unit
Molecular typing
Nosocomial infections

SUMMARY

Background: Nosocomial transmission of *Candida* spp. has not been fully explored and previous studies have shown conflicting results.
Aim: To evaluate the possible nosocomial transmission of *Candida* spp. on an intensive care unit (ICU).
Methods: A prospective study was conducted for a period of 19 months, including all patients on our ICU with growth of *Candida* spp. from surveillance and directed cultures. Molecular typing with repetitive sequence-based polymerase chain reaction was used to define genotype relationships between the *Candida albicans* and *Candida glabrata* isolates. Candida isolates obtained from blood cultures taken from patients in our county outside the ICU were used as a reference. Temporal cluster analysis was performed to evaluate genotype distribution over time.
Findings: Seventy-seven patients with 78 ICU stays, representing 12% of all ICU stays, were found to harbour 180 isolates of *Candida* spp. Molecular typing revealed 27 *C. albicans* genotypes and 10 of *C. glabrata*. Possible clustering, indicated by overlapping stays of patients with indistinguishable candida genotypes, was observed on seven occasions with *C. albicans* and on two occasions with *C. glabrata*. Two *C. albicans* genotypes were found significantly more often in the ICU group compared with the reference group. Moreover, *C. albicans* genotypes isolated from more than one patient were significantly more often found in the ICU group. Temporal cluster analysis revealed a significantly increased

* Corresponding author. Address: Department of Anaesthesia and Intensive Care, Ryhov County Hospital, 551 85 Jönköping, Sweden.
E-mail address: fredrik.hammarskjold@lj.se (F. Hammarskjöld).

http://dx.doi.org/10.1016/j.jhin.2013.06.002

number of pairs with indistinguishable genotypes at a 21-day interval, indicating clustering.

Conclusion: This study indicates possible transmission of *C. albicans* between ICU patients based on genotyping and temporal cluster analysis.

Introduction

Fungal infections are an increasing problem in intensive care units (ICUs) and contribute to increased mortality. Furthermore, ICU patients with longer ICU stays are more prone to be colonized with *Candida* spp. compared to those with short stays.[1] Nosocomial candida infections on ICUs have previously been considered to be endogenously acquired.[2] However, healthcare workers may carry candida on their hands and nosocomial outbreaks of *Candida* spp. have been reported, suggesting that transmission is a possibility.[3–7] A few studies have addressed transmission of *C. albicans* within hospitals and units; the results, however, are conflicting.[4–6,8–12] Previous studies of transmission have been based on the distribution of genetic variants of *Candida* spp. in patients and the environment. No study has attempted a temporal cluster analysis. The distribution of genotypes within a suitable reference group is required if one is to evaluate the transmission of *Candida* spp. on the ICU. The role of the inanimate environment as a source of *C. albicans* transmission has hardly been investigated and has not been confirmed in the ICU environment.[13] In a previous study on catheter-related bloodstream infections on our ICU, a low incidence was found, but the proportion of candida isolates was high.[14]

The aim of this study was to evaluate the possibility of nosocomial transmission of *Candida* spp. by comparing the distribution of patients on our ICU with a reference group from patients outside the ICU. This was complemented by temporal cluster analysis which was performed to evaluate genotype distribution among ICU patients over time. The genetic relatedness of *C. albicans* and *C. glabrata* isolates was investigated using a commercially available repetitive sequence-based polymerase chain reaction (rep-PCR) technique, DiversiLab (bioMérieux, Marcy l'Etoile, France). DiversiLab has proven its utility as a standardized and reproducible system useful for local epidemiological studies of *Candida* spp., and is easy to use in clinical laboratories at a low cost.[15]

Methods

Setting

Ryhov County Hospital is a 500-bed public hospital supporting most medical and surgical specialties. The ICU is a seven-patient (two single rooms, one double room and one four-bedded room) general ICU with patients having a median Acute Physiology and Chronic Health Evaluation II (APACHE II) score of 18. The nurse:patient ratio is 1.3:1.

Study design

A prospective cohort study was performed between 1 January 2007 and 31 July 2008 collecting all candida isolates ($N = 180$) from surveillance and directed cultures. All patients ($N = 77$) with a positive candida culture were included and patient data were recorded (age, gender, APACHE II score, length of ICU stay, antimicrobial drugs, surgery and ICU mortality). The surveillance cultures (tracheal secretion, catheter urine, perineal swab, wounds and incision sites) were collected every Monday from all patients with an ICU stay longer than 72 h. Directed cultures were collected at the request of the physician responsible or the daily visiting infection specialist. All patients who had a blood culture isolate of *C. albicans* or *C. glabrata* from 2006 to 2008 in the county but not treated at this ICU were chosen as reference group.

Microbiology

Samples were cultured on BBL™ CHROMagar™ Candida medium (bioMérieux), incubated at 35 °C overnight. Species determination of candida isolates was performed on the VITEK2 compact, using the YST card, according to the manufacturer's recommendation (bioMérieux).

Extraction and purification of DNA were performed using the Ultra Clean™ Microbial DNA Isolation Kit (MO BIO Laboratories, Inc., Carlsbad, CA, USA) in accordance with the manufacturer's instructions for fungi, with the following modifications: after addition of the MD1™ solution the samples were incubated at 80 °C for 30 min and after 45 min of bead beating the samples were centrifuged for 2 min. The isolates from blood cultures were also incubated with 15 units of Zymolyase (Zymo Research, Irvine, CA, USA) at 37 °C for 30 min prior to the addition of the MD1™ solution. DNA samples were amplified using the candida fingerprinting kit according to the manufacturer's instructions (bioMérieux). The fragments were separated using an Agilent 2100 Bioanalyzer (Agilent Technologies, Santa Clara, CA, USA). Finally, web-based software (DiversiLab v3.4) was used to analyse the genotypic similarity of isolates, by the Kullback–Leibler method.[15] An internal positive control was chosen based on the number and distribution of produced fragments. Repeated analysis of the internal positive control gave an inter-run similarity of 97%. The limit for indistinguishable isolates, and thus our definition of a genotype, was therefore set at 97% similarity instead of the recommended 95% (bioMérieux).

Definitions

Candida infection and colonization were defined as candida growth with or without clinical symptoms related to candida as judged by the attending ICU physician in consultation with the daily visiting infection specialist.

Statistics

Significant differences between groups were determined using χ^2- or Fisher's exact test for categorical variables and Student's *t*-test for continuous variables. Analysis was performed with Statistica 10 (Statsoft, Tulsa, OK, USA).