Journal of Advances in Microbiology

12(1): 1-7, 2018; Article no.JAMB.42991
ISSN: 2456-7116

Aerobic Bacteria and Fungi on the Surfaces of a Tertiary Assistance Hospital from Northern Brazil

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Authors' contributions

This work was carried out in collaboration with all authors. Authors JVBS and ACAC designed the study, managed the literature searches and wrote the first draft of the manuscript. Authors LGCL and MAS performed the laboratorial work and wrote the first draft of the manuscript. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/JAMB/2018/42991
Editor(s):
(1) Dr. Foluso O. Osunsanmi, Department of Biochemistry and Microbiology, University of Zululand, South Africa.
Reviewers:
(1) Alvaro Francisco Lopes de Sousa, University of São Paulo, Brazil.
(2) Iramiot Jacob Stanley, Busitema University, Uganda.
(3) S. Thenmozhi, Department of Microbiology, Periyar University, India.
Complete Peer review History: http://www.sciencedomain.org/review-history/26183

Original Research Article

Received 27 June 2018
Accepted 29 August 2018
Published 11 September 2018

ABSTRACT

Background and Objectives: It is very common that hospital environment is colonised by microorganisms. This colonisation is a potential threat for hospitalised patients, especially in high-risk services. Quantification of microorganisms from surfaces is an important strategy to control the hospital infection.
Materials and Methods: Total 70 samples were collected and seeded in the Mueller Hinton and Sabouraud Agar culture media. Media cultures in Petri dishes were incubated and microbial growth was quantified (CFU/cm^2). Bacteria cultures were characterised by Gram stain. Fungal cultures were submitted for micromorphological evaluation and CHROMagar™ incubation.
Results: Bacterial population higher than 250 CFU/cm^2 was found in 11 out of 70 samples. Fungi were found in 3 out of 70 samples. Common manipulation surfaces as water drinking unit, shelf and water tap presented the highest contamination rates. Bacterial cultures (n= 95) showed mostly the presence of Gram-negative bacteria (81%). In addition, regarding the fungal cultures (n= 129) mostly filamentous fungi (72%), from *Aspergillus* genera were obtained.

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Conclusion: Most of the investigated hospital surfaces presented low contamination (< 5 CFU/cm^2). However, microbiological studies should be regular in critical areas in order to reinforce measures to control and prevent hospital infection.

Keywords: Bacteria; fungi; surfaces; hospital.

1. INTRODUCTION

The hospital environment is largely contaminated with pathogenic microorganisms. This colonization is a threat for hospitalised patients, especially in high-risk services [1,2]. This prevalence is quantitatively and qualitatively variable from one institution to another and in the same establishment according to the services. It can constitute a risk factor for nosocomial infections [3-5].

Although it is difficult to establish a direct link between environmental contamination and the occurrence of nosocomial infections, several studies have shown that microorganisms from human and/or environmental origin contaminate hospital surfaces, and play an important role in the occurrence of these infections [6,7]. Microbiological monitoring of the environment in health facilities is part of preventing the transmission of nosocomial infections [8]. The microbiological controls of the environment are one of the measuring tools that make it possible to evaluate a starting situation and the effectiveness of corrective measures, therefore, they must be implemented in a relevant way and obey very precise objectives while avoiding the inflation of useless analyses, consuming time and financial means [9-11].

There are no systematic studies on the relative importance of various environmental factors to ensure a safe environment although it has always acknowledged that pathogenic microorganisms in fomites may be a possible risk factor for nosocomial infection. In Northern Brazil, there are no studies to quantify or reduce contamination of the hospital environment for the last 10 years. The hypothesis that environmental microorganisms cause human disease results from two facts: first, our interaction with inanimate and, secondly, environmental objects, which are generally contaminated with potential human pathogens [4]. The objective of this study was to quantify and qualify microorganisms present on hospital surfaces from a tertiary hospital in Northern Brazil.

2. MATERIALS AND METHODS

2.1 Location and Type of Study

A descriptive cross-sectional study was conducted at the General Hospital of Brazil (GHB) from 1st January to 30th June 2018. This is a tertiary health facility located in the north region. This hospital has a capacity of 102 beds and has all the major medical and surgical specialities. Surfaces were selected by convenience and focused on important hospital services: ICU- Intensive Care Unit; SC- Surgery centre; NS- Infirmary service; OC- Outpatient care; OS- Oncology service; EC- Emergency care; BN- Baby nursery and NS- Nutrition service.

Microbiological analyses were carried out at the Mycology laboratory from the National Institute for Amazon Research. These samples were taken between 5 AM and 8 AM (24hs after cleaning procedures) using a sterile swab previously moistened with sterile saline 0.9%. The swabs were passed in parallel streaks by slightly turning them, on defined areas of 4 cm^2.

2.2 Microbial Isolation

The swabs from the different sampling sites were immediately inoculated into Mueller Hinton Agar and Sabouraud Agar. The petri dishes were incubated in a bacteriological incubator at 35°C for 48 hours for the bacteria, and at 25°C for 5 days for the fungi. The number of colony forming units are expressed as per cm^2 [12,13].

2.3 Colonies Identification

Petri dishes after incubation were observed for colony counting and macroscopic classification (shape, size, colour) [13,14]. Bacterial identification was carried out by morphological analysis by Gram staining. Fungal identification was carried out by macromorphological and micromorphological characterisation (Cotton Blue Stain). Specifically, *Candida* spp. isolates were cultivated and classified on CHROMagar™ (CHROMagar Microbiology, France).

3. RESULTS

In order to investigate bacteria (CFU/mL) in hospital surfaces, isolation tests were performed by using Petri dishes containing Mueller Hinton Agar (Fig. 1). At least, one sample per type of investigated surface presented more than 250 CFU/cm^2 (except bed grid). The water taps were the most contaminated surfaces, five samples presented more than 250 CFU/cm^2.

In order to investigate fungi (CFU/mL) in hospital surfaces, isolation tests were performed by using Petri dishes containing Sabouraud Agar (Fig. 2). Three samples presented bacterial population more than 250 CFU/mL (Fig. 2).

Micro-morphological analysis was performed to characterise the isolates. Fig. 3 summarises the results of these tests. Out of the 95 bacterial isolates, the majority (74%) was Gram-negative cocci and most of the 129 fungal cultures were molds, a significant part of them were from *Aspergillus* genera.

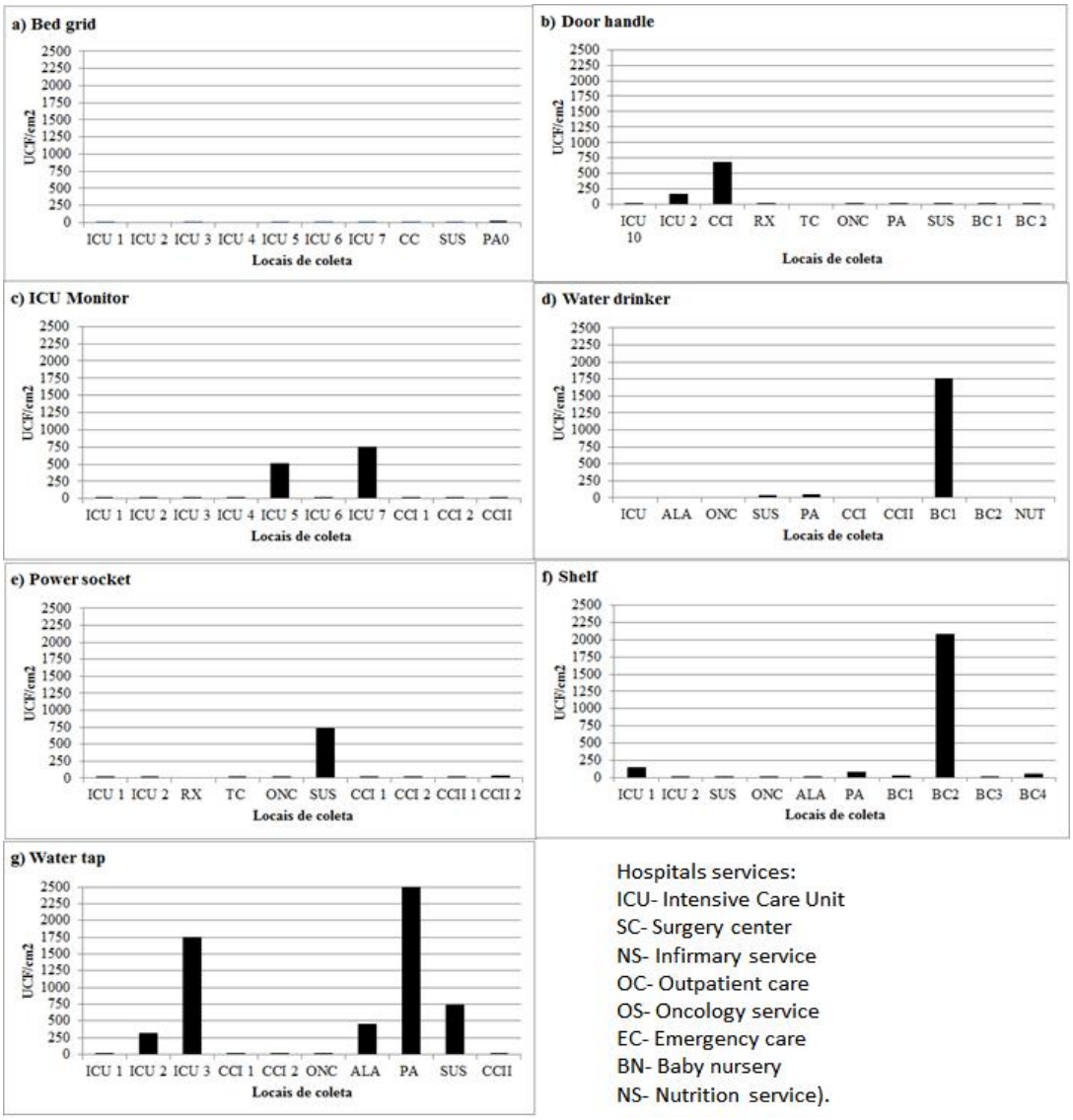

Fig. 1. Number of cultivated bacteria (CFU/cm^2) from the surfaces of a tertiary assistance hospital from northern Brazil (a-Bed grid, b-Door handle, c-Cardiac monitor, d-Water drinker, e-Power socket, f-Shelf and g-Water tap)

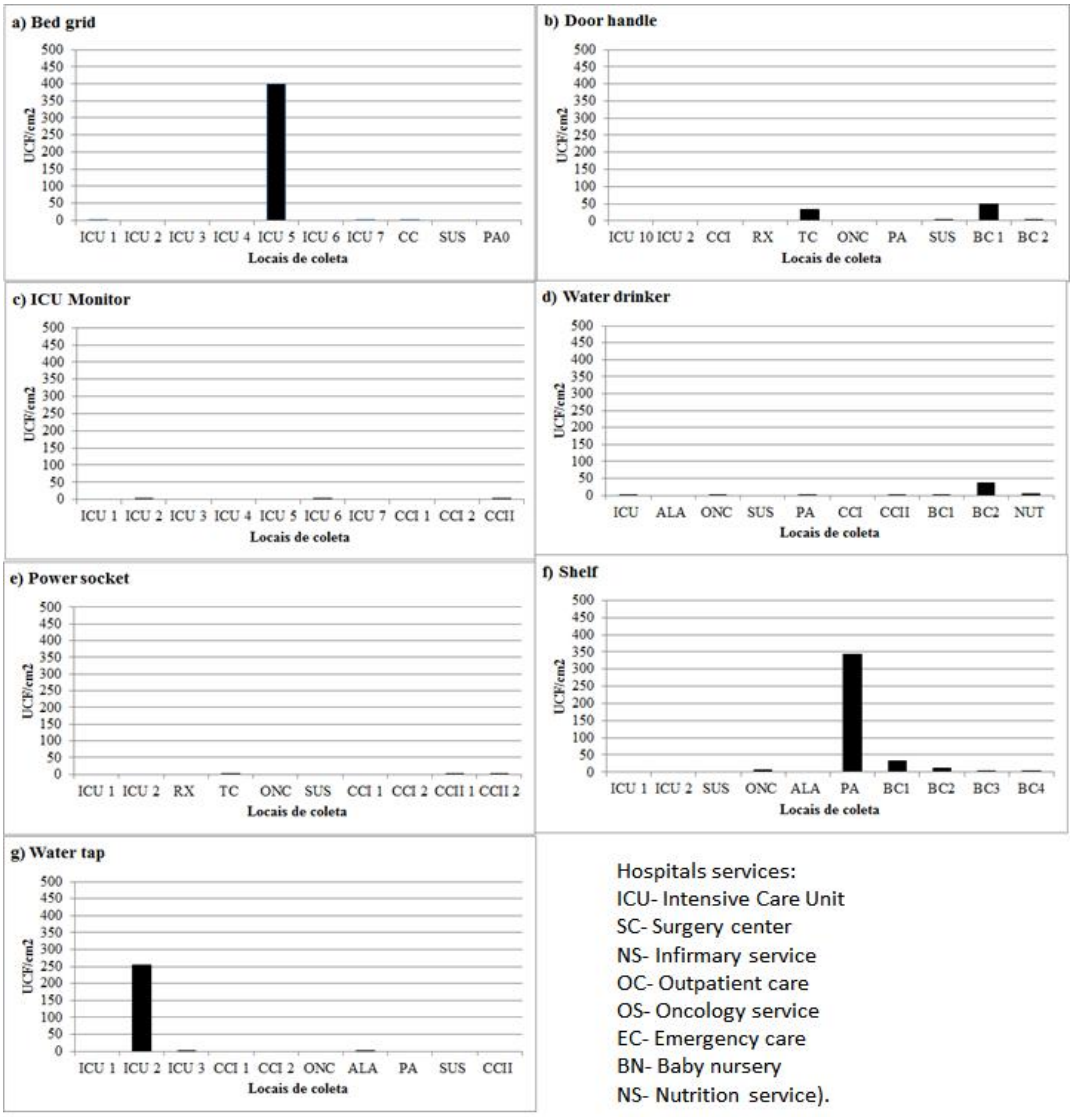

Fig. 2. Number of fungi (CFU/cm^2) from surfaces of a tertiary assistance hospital from northern Brazil (a-Bed grid, b-Door handle, c-Cardiac monitor, d-Water drinker, e-Power socket, f-Shelf and g-Water tap)

4. DISCUSSION

In the present study bacteria and fungi were found in higher number in common manipulation surfaces like drinking water, shelf and tap water. They presented higher contamination rates. Bacteria were mostly Gram-negative while most of the fungi were molds. However, microbiological studies should be regular in critical areas in order to reinforce measures to control and prevent hospital infection [5].

Some workers suggested for bacteriological standards to assess clinical surface hygiene in hospitals [15]. Dancer [15] proposed that the first standard concerns any finding of a specific 'indicator' organism, the presence of which suggests a requirement for increased cleaning. Indicators include *Staphylococcus aureus*, including methicillin-resistant *S. aureus*, *Clostridium difficile*, vancomycin-resistant enterococci and various Gram-negative bacilli. The second standard concerns a quantitative aerobic colony count of 5 CFU/cm^2 on frequent hand touch surfaces in hospitals.

In the present study, most of the investigated hospital surfaces presented low

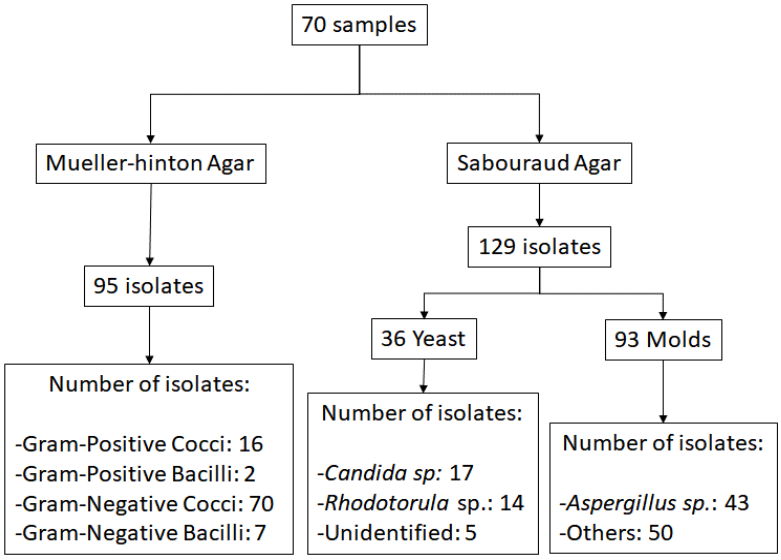

Fig. 3. Identification of microorganisms isolated from the surfaces samples from a tertiary care hospital in Northern Brazil

contamination (<5 CFU/cm^2). Recently, Ebongue et al. [1], has found several colonised sites with 10^5 CFU/cm^2 microorganisms on various surfaces and medical devices in the General Hospital of Douala. As previously described, common manipulation surfaces like water drinking unit, shelves and water tap show higher contamination rates [16-20]. This information is important and demonstrates that in the investigated facilities, more attention should be given to this type of surfaces.

In the present study, 95 bacterial cultures were obtained. Mostly Gram-negative bacteria (81%) were observed. This was an expected response since previous and recent studies have demonstrated similar results. These organisms are the potential causes of hospital infections and their hospital population must be monitored and controlled [21,22].

In addition, in the present study, 129 isolates were fungi, out of which, 36 isolates were identified as yeast and 93 were filamentous fungi. Among the fungi, 43 isolates were identified as *Aspergillus* spp. Most *Candida* infections comes from endogenous origin, however, this organism may survive few days in the hospital setting and be transmitted by the hands of caregivers and surfaces [23,24]. The most isolated filamentous fungus was *Aspergillus* spp. with 70.5% of the isolates, in a multicenter study in hospitals. Mousa et al. (1999) verified the correlation between fungal isolates of burns with the environmental surfaces of the burn therapy unit (UTQ), isolating *Aspergillus niger* in greater numbers in patients and environment [25-27].

The present work had the following limitations: a) The organisms were not identified upto the species level; b) the organisms were not evaluated for susceptibility to antibiotics. However, the study reported robust sampling and simple assessment methodologies as the tools to be used routinely for the evaluation of hospital colonisation. As well as, the data generated here may serve for future discussions.

5. CONCLUSION

The present work described the microbial distribution in a hospital in the north of Brazil. Surfaces from water drinking unit, shelf and water tap presented higher contamination rates. The microbiological controls make possible to evaluate a starting situation and evaluate the effectiveness of corrective measures. The study suggests that microbiological studies should be regular in critical areas in order to reinforce measures to control and prevent hospital infection.

ACKNOWLEDGEMENTS

This project was conducted with support from the CNPq, FAPEAM and CAPES.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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Peer-review history:
The peer review history for this paper can be accessed here:
http://www.sciencedomain.org/review-history/26183