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# Epidemiology of Bacterial Contamination of Inert Hospital Surfaces and Equipment in Critical and Non-critical Care Units: A Brazilian Study

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***Authors' contributions***

*All authors contributed to the study conception and design. Development of methodology, performing the experiments were performed by authors LPP, ETMB, JRG, BCG, LGA, JCOL, JLA and DSC under supervision of the author DOR. Material preparation, data collection and analysis were performed by all authors. The first draft of the manuscript was written by author DOR and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.*

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## ABSTRACT

**Aims:** The hospital environment is an important reservoir of microorganisms, including multidrug-resistant pathogens, which can cause in-patient contamination and healthcare-related infections. The objective of this study was to describe the epidemiology of bacterial contamination (contaminated sites, pathogen species and their antimicrobial susceptibility, and identifying of

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multidrug-resistant microorganisms - MDR) of inert hospital surfaces and medical equipment in two public hospitals in Northern Brazil.
**Methods:** This was a cross-sectional study with 243 samples (*n* = 208, from Hospital A; and *n* = 35, from Hospital B) collected by friction with humidified swabs from inert surfaces and equipment. Sequentially the samples were cultivated and bacterial species were identified by culture-based methods and tested for their susceptibility through agar diffusion assay according to the Clinical and Laboratory Standards Institute (CLSI).
**Results:** Most inert surfaces and equipment analyzed presented bacterial contamination (95.5%). *Staphylococcus aureus* was the main pathogen of clinical significance detected both in Hospital A (61.8%) and B (68.6%). Hospital A showed higher rates of isolated MDR bacteria than Hospital B, especially in the Adult Intensive Care Unit, which included methicillin-resistant *Staphylococcus aureus* (MRSA) (52.7%), Enterobacteria resistant to 4$^{th}$ generation cephalosporins (19.4%), and multidrug-resistant *Pseudomonas aeruginosa* (2.8%).
**Conclusion:** The failures in the control of bacterial contamination of inert surfaces and equipment in the two hospitals analyzed reinforce the need for a revised protocol for cleaning and disinfection of the inert surfaces and equipment, and for regulation of antibiotic dispensing, mainly in the AICU of Hospital A, which was found to be a reservoir of MDR pathogens.

*Keywords: Infections; pathogens; cross-transmission; reservoir.*

## 1. INTRODUCTION

Healthcare-associated infections (HAI) are a major public health concern commonly associated with extended length of hospital stay, invasive procedures and antimicrobial resistance. HAI account for high hospital costs and contribute to increased morbidity and mortality of infected patients [1].

HAI are usually caused by pathogenic bacteria that may emerge from the patient's endogenous microflora during antibiotic therapy in approximately 70% of the cases [2,3]. HAI may also be acquired from the exogenous environment (30% of the cases) in that the hospital setting plays a significant role in contagion and transmission of the pathogens to patients [3,4]. Thus, epidemiological studies involving environmental contamination are important.

In the hospital setting, patients, staff and visitors represent the main reservoir of microorganisms, whereas secondary reservoirs include all environments where nutrients, moisture, and temperature are suitable for microbial survival, such as air humidifiers and nebulizers [5,6]. In addition, dry and inanimate surfaces can also serve as a reservoir of pathogens [3,5,7-9], as in mattresses and bed frames [4,10,11], door knobs [11,12], and even in medical equipment such as stethoscopes and ultrasound devices [7-11]. Contamination of these surfaces contributes to pathogen spreading and, as a result, development of infections by horizontal transmission [13-14].

Overall, surfaces can be directly contaminated by bacteria from colonized and/or infected patients or from the hands of health professionals [5,7]. Highly touched surfaces (e.g. bed frames, stethoscopes, bedside tables, and door knobs) [11-13] may be contaminated by common bacteria of the hand microbiota. More importantly, MDR bacteria have been detected on medical equipment and contact surfaces, especially in critical care units [10,15,16].

Studies carried out in Brazil and North America have reported contamination of hospital surfaces by bacteria resistant to antibiotics, especially methicillin-resistant *S. aureus* (MRSA) [4,17,18] and vancomycin-resistant enterococci [19,20]. These findings indicate that patients and staff are at risk of contamination by pathogens associated with high mortality rates against which treatment options are restricted.

Given the importance of in-hospital transmission, HAI studies have looked into the epidemiology of hospital-related bacterial contamination to propose preventive measures to reduce the contamination and dissemination of resistant pathogens [5]. This study aimed to describe the epidemiology of bacterial contamination (contaminated sites, pathogen species and their antimicrobial susceptibility, and identifying of multidrug-resistant microorganisms - MDR) of inert hospital surfaces and medical equipment in two public hospitals in Northern Brazil.

## 2. MATERIALS AND METHODS

This was a cross-sectional study carried out in the *Hospital Geral Público de Palmas* (HGPP) [Palmas Public General Hospital] (A) located in the state of Tocantins, Brazil, and in the *Hospital Municipal Eurico Dutra* [Eurico Dutra Municipal Hospital] (B) located in the city of Barreiras, Western Bahia, Brazil.

Hospital A is a large teaching hospital which provides tertiary care and is a reference center for medium and high complexity healthcare assistance. The hospital contains approximately 400 beds and is a major health center for the state of Tocantins and neighboring states in Brazil. It includes two intensive care units (ICUs) – a pediatric unit (PICU) with 3 rooms and 8 beds, and an adult unit (AICU) with 26 beds distributed among 18 rooms; pharmacy; laboratory; operating room (OR); elective and emergency surgery services; emergency room (ER) with three on-call specialties – orthopedics, internal medicine, and surgery; conventional hospital ward, and home care and outpatient facilities, with an average 3,500 appointments per month.

Hospital B is a medium-sized and medium complexity general hospital which provides services such as diagnostic and therapeutic support services, emergency and outpatient care. Patients are admitted through spontaneous and referred demand from the city of Barreiras and surrounding region. The hospital has a total of 10 beds in the Internal Medicine Ward (IMW), 29 beds in the Surgical Ward (SW) and 4 OR with 5 post-anesthesia recovery beds in the SW.

### 2.1 Sample Collection

There was a substantial difference in the number of samples between the two hospitals because the Hospital A and B are the large and medium-sized, respectively. A total of 243 samples (*n* = 208, Hospital A; *n* = 35, Hospital B) were collected from the following surfaces and medical equipment: Hospital A - ER (door knobs, *n* = 56); OR (door knobs, *n* = 20); AICU (heart monitors, *n* = 18; infusion pumps, *n* = 18; medication tables, *n* = 18; side bed frames, *n* = 18; with a total *n* = 72); and PICU (side bed frames, *n* = 16; bed headboards, *n* = 8; bed frame feet, *n* = 8; mattresses, *n* = 8; bedside tables, *n* = 8; stethoscopes, *n* = 8; samples from the sinks, *n* = 4; with a total *n* = 60); Hospital B - IMW (bed headboard, *n* = 3, mattresses, n = 2; side bed frame, *n* = 2; saline stand, *n* = 1; door knobs, *n* = 3, with a total *n* = 11); SW (bed headboard, *n* = 5, mattresses, *n* = 3; side bed frame, *n* = 1; saline stand, *n* = 3; countertops, *n* = 1; door knobs, *n* = 4; with a total *n* = 17); OR (surgical light, *n* = 4; stretcher, *n* = 3; with a total *n* = 7). The selected hospital areas included those with obtained access and represented the most important areas of hospital. The selected hospital surfaces included those highly touched or nearby patients.

Our team conducted the sampling and we ensure that everyone's sampling techniques were standardized. We collected on the touch-screen and buttons from the heart monitors, on the buttons from the infusion pumps, on the lateral from the mattress, and from the knobs sinks. We collected the samples with the patients in the room/bed at the time of sampling.

The samples were collected in the morning and afternoon from July to October 2018 (Hospital A) and April 2018 (Hospital B) by friction with sterile swabs moistened in broth Brain Heart Infusion (BHI, Oxoid, Basingstoke, Hampshire, England) from selected surfaces. The swabs were placed immediately into sterile tubes with broth BHI and transported in insulated boxes to the *Centro Universitário Luterano de Palmas* (CEULP) [Palmas Lutheran University Center] (Hospital A) and to the *Universidade Federal do Oeste da Bahia* (UFOB) [Federal University of Oeste da Bahia] (Hospital B) and incubated at 37 °C for 24 h.

### 2.2 Bacterial Culture

After incubation, the samples in the tubes with broth BHI were vortexed and subcultured onto Mannitol Salt Agar, MacConkey, and Pseudomonas agar supplemented with blood to isolate *S. aureus,* coagulase-negative staphylococci (CoNS), and Gram-negative bacteria (Enterobacteriaceae family, non-fermenters), respectively, in the aerobic conditions.

Next, clinically significant bacterial species [21] were identified by the classical approach as follows:

- *S. aureus* and CoNS: fermentation of Mannitol Salt Agar, Gram staining, catalase test, coagulase test, DNase Agar;
- Enterobacteriaceae family: growth on MacConkey agar, cytochrome oxidase,

Gram staining, lactose fermentation (Triple Sugar Iron Agar -TSI), biochemical tests (B Enterokit Probac of Brazil);

- Non-fermenter Bacilli (*Pseudomonas aeruginosa*): growth on Pseudomonas agar, odor, colony morphology, cytochrome oxidase, growth at 42°C, Gram staining, oxidation in Hugh Leifson medium;
- *Micrococcus* spp.: Gram staining, growth on Blood agar, no growth on Mannitol Salt Agar, catalase test, lysostapine resistance
- Gram-Positive Bacilli: Gram staining;

The bacterial strains were then tested for their antimicrobial susceptibility in vitro by the agar diffusion technique as recommended by the Clinical and Laboratory Standards Institute [22]. The tested antibiotics included imipenem (10 mcg), cefepime 30 (mcg), chloramphenicol (30 mcg), vancomycin (30 mcg), oxacillin (1 mcg), ciprofloxacin (5 mcg), co-amoxiclav (30/10 mcg), gentamicin (10 mcg), norfloxacin (10 mcg), cefoxitin (30 mcg), cefotaxim (30 mcg), amikacin (30 mcg), piperacillin (100 mcg), linezolid (30 mcg) and sulfamethoxazole (25 mcg).

Bacterial strains were considered as multidrug-resistant (MDR) if showing resistance to at least three classes of antimicrobials [23,24] and associated phenotypes. Some examples included MRSA, Enterobacteria resistant to 4$^{th}$ generation cephalosporins and quinolone-resistant *P. aeruginosa*, according to definitions of the European Center for Disease Prevention and Control [23].

## 3. RESULTS

Bacterial contamination was detected in 94.7% of the 208 sampled surfaces and equipment in Hospital A. A total of 233 bacterial isolates were obtained and no microbial growth was observed on 11 surfaces sampled in the PICU. In Hospital B, all 35 surfaces that were sampled were identified with bacterial contamination.

The microbiological analysis of the surfaces in Hospital A showed a predominance of *S. aureus* (62.0%), common bacteria of the human flora, and nosocomial bacteria, such as Enterobacteria (24.0%) and *P. aeruginosa* (7%). In Hospital B, there was a predominance of environmental microorganisms, such as Gram-positive Bacilli (41%) and *Micrococcus* spp. (9.0%) (Fig. 1).

In Hospital A, the ER door knobs were mostly contaminated with *S. aureus* (53.3%), but also with enteric bacteria (30.4%), with a high frequency of samples contaminated with *P. aeruginosa* (64.7%) (Table 1). The AICU showed the highest frequency of *S. aureus* colonization, particularly on heart rate monitors (83.3%). Infusion pumps and side bed frames were mostly contaminated with *P. aeruginosa* (16.7%). The PICU was frequently contaminated not only with *S. aureus* on all stethoscopes analyzed, but also with CoNS. As shown in Table 1, 10 out of the 16 CoNS samples found throughout the study originated from the PICU.

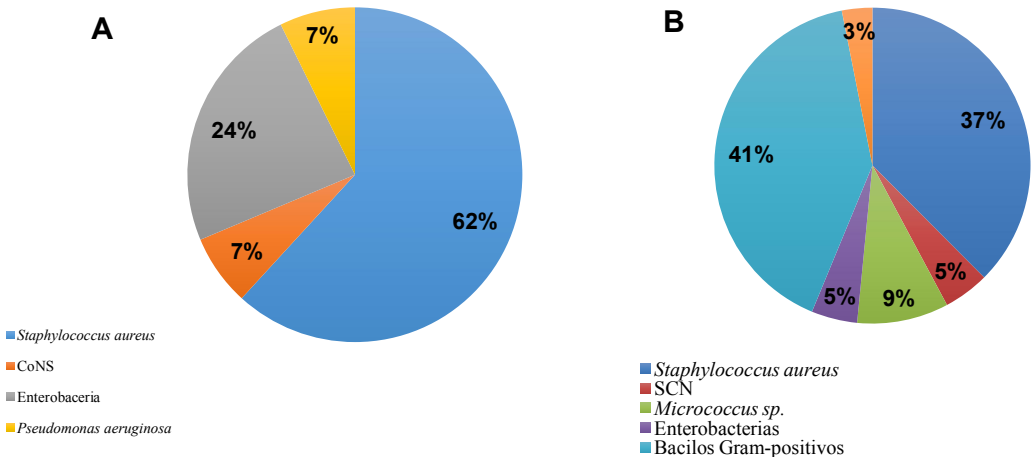


**Fig. 1. Frequency of microorganisms recovered from inert surfaces and medical equipment in the Hospital Geral Público de Palmas-TO, Brazil (A) and Hospital Municipal Eurico Dutra in the city of Barreiras, BA, Brazil (B)**

*CoNS: Coagulase-Negative Staphylococci*

**Table 1. Frequency of clinically important microorganisms isolated from inert surfaces and medical equipment in the Hospital Geral Público de Palmas -TO, Brazil (A).**

| Hospital units | Surfaces (N) | *S.aureus* N (%) | CoNS N (%) | Enterobacteria N (%) | *P. aeruginosa* N (%) | Total of bacteria (N) |
|---|---|---|---|---|---|---|
| **ER** | door knobs (56) | 49 (53,3) | 4 (4,3) | 28 (30,4) | 11 (11,9) | 92 |
| **OR** | door knobs (20) | 12 (60,0) | 2 (10,0) | 5 (25,0) | 1 (5,0) | 20 |
| **AICU** | heart monitors (18) | 15 (83,3) | - | 3 (16,7) | - | 18 |
| | infusion pumps (18) | 13 (72,2) | - | 3 (16,7) | 2 (11,1) | 18 |
| | medication tables (18) | 14 (77,8) | - | 4 (22,2) | - | 18 |
| | side bed frames (18) | 11 (61,1) | - | 6 (33,3) | 1 (5,6) | 18 |
| **PICU** | side bed frames (16) | 8 (57,1) | 1 (7,4) | 5 (35,7) | - | 14 |
| | bed headboards (8) | 3 (50,0) | 2 (33,3) | 1(16,7) | - | 6 |
| | bed frame feet (8) | 3 (50,0) | 2 (33,3) | - | 1 (16,7) | 6 |
| | mattresses (8) | 3 (75,0) | 1 (25,0) | - | - | 4 |
| | bedside tables (8) | 3 (42,8) | 3(42,8) | 1(14,3) | - | 7 |
| | stethoscopes (8) | 8 (100,0) | - | - | - | 8 |
| | samples from the sinks (4) | 2(50,0) | 1(25,0) | | 1(25,0) | 4 |
| **Total (N)** | **208** | **144 (61,8)** | **16 (6,9)** | **56 (24,0)** | **17 (7,3)** | **233** |

*N: total sample size; %: Percentage; ER: Emergency Room; OR: Operating Room; AICU: Adult Intensive Care Unit; PICU: Pediatric Intensive Care Unit; CoNS: Coagulase-Negative Staphylococci*

**Table 2. Frequency of clinically important microorganisms isolated from inert surfaces and medical equipment in the Hospital Municipal Eurico Dutra in the city of Barreiras, BA, Brazil (B)**

| Hospital units | Surfaces (N) | *S. aureus* N (%) | CoNS N (%) | Enterobacteria N (%) | *Micrococcus* N (%) | Total of bacteria (N) |
|---|---|---|---|---|---|---|
| **IMW** | bed headboard (3) | 2 (66,7) | - | 1 (33,3) | - | 3 |
| | mattresses (2) | 1 (50,0) | - | - | 1 (50,0) | 2 |
| | side bed frame (2) | 1 (50,0) | - | - | 1 (50,0) | 2 |
| | saline stand (1) | - | - | - | 1 (100,0) | 1 |
| | door knobs (3) | 2 (66,7) | - | 1 (33,3) | - | 3 |
| **SW** | bed headboard (5) | 3 (60,0) | 1 (20,0) | 1 (20,0) | - | 5 |
| | mattresses (3) | 2 (66,7) | - | - | 1(33,3) | 3 |
| | side bed frame (1) | 1 (100,0) | - | - | - | 1 |
| | saline stand (3) | 3 (100,0) | - | - | - | 3 |
| | countertops (1) | 1 (100,0) | - | - | - | 1 |
| | door knobs (4) | 2 (50,0) | 1 (25,0) | - | 1 (25,0) | 4 |
| **OR** | surgical light - BU (2) | 2 (100,0) | - | - | - | 2 |
| | surgical light - AU (2) | 2 (100,0) | - | - | - | 2 |
| | stretcher - BU (1) | 1 (100,0) | - | - | - | 1 |
| | stretcher - AU (2) | 1 (50,0) | 1 (50,0) | - | - | 2 |
| **Total (N)** | **35** | **24 (68,6)** | **3 (8,6)** | **3 (8,6)** | **4 (11,4)** | **35** |

*N: total sample size; %: Percentage; IMW: Internal Medicine Ward; SW: Surgical Ward; OR: Operating Room; BU: Before Use; AU: After Use; CoNS: coagulase-negative staphylococci*

**Table 3. Multi-drug resistant microorganisms recovered from inert surfaces and equipment of hospitals located in Northern Brazil.**

| Hospital | Hospital units | Number of samples | Culture positive | Microorganisms (N) | MDR* isolated (Frequency) |
|---|---|---|---|---|---|
| **Hospital A (Palmas-TO)** | **ER** | 56 | 92 | *S.aureus* (49)<br>CoNS (4)<br>Enterobacteria (28)<br>*P.aeruginosa* (11) | MRSA (28,3%)<br>MR CoNS (4,3% )<br>Enterobacteria resistant to 4$^{th}$ generation cephalosporins (8,7%)<br>- |
| | **OR** | 20 | 20 | *S.aureus* (12)<br>CoNS (2)<br>Enterobacteria (5)<br>*P.aeruginosa* (1) | MRSA (25,0%)<br>MR CoNS (5,0%)<br>-<br>- |
| | **AICU** | 72 | 72 | *S.aureus* (53)<br>Enterobacteria (16)<br>*P.aeruginosa* (3) | MRSA (52,7%)<br>Enterobacteria resistant to 4$^{th}$ generation cephalosporins (19,44%)<br>multi-resistant *P.aeruginosa* (2,78%) |
| | **PICU** | 60 | 49 | *S.aureus* (30)<br>CoNS (10)<br>Enterobacteria (7)<br>*P.aeruginosa* (2) | MRSA (4,08%)<br>MR CoNS (20,40%)<br>Enterobacteria resistant to 4$^{th}$ generation cephalosporins (6,12%)<br>multi-resistant *P.aeruginosa* (2,04%) |
| **Hospital B (Barreiras-BA)** | **IMW** | 11 | 11 | *S.aureus* (6)<br>Enterobacteria (2)<br>*Micrococcus*. (3) | MRSA (27,27%)<br>-<br>- |
| | **SW** | 17 | 17 | *S.aureus* (13)<br>CoNS (2)<br>Enterobacteria (1)<br>*Micrococcus*(1) | MRSA (29,4%)<br>MR CoNS (5,88%)<br>-<br>- |
| | **OR** | 7 | 7 | *S.aureus* (6)<br>CoNS (1) | MRSA (14,3%)<br>- |

**Multidrug-resistant pathogen; ER: Emergency Room; OR: Operating Room; AICU: Adult Intensive Care Unit; PICU: Pediatric Intensive Care Unit; IMW: Internal Medicine Ward; SW: Surgical Ward; MRSA: Methicillin-Resistant Staphylococcus aureus; MR-CoNS: Methicillin-Resistant Coagulase-Negative Staphylococci*

The surfaces of Hospital B were contaminated with *S. aureus* in the following sites: bed headboards (66.7%) in the IMW, saline stands (100.0%) in the SW and surgical lights (100.0%) in the OR (Table 2).

Hospital A showed higher rates of MDR bacteria recovered from surfaces than Hospital B, especially in the AICU, which included MRSA (52.7%), Enterobacteria resistant to 4$^{th}$ generation cephalosporins (19.4%) and multi-resistant *P. aeruginosa* (2.78%). The OR in both Hospital A and B had the lowest frequency of MDR bacteria, as shown in Table 3.

## 4. DISCUSSION

Inert hospital surfaces and medical equipment can be a reservoir of MDR pathogens. Understanding the epidemiology of bacterial contamination in this setting is essential to prevent in-patient contamination and HAI development.

Bacterial pathogens can survive and remain viable on inert surfaces and equipment due to their ability to form biofilms and to environmental factors such as surface porosity and humidity [7,25]. This ability to adapt to environmental stress works as a major factor driving pathogen proliferation and dissemination. In this context, our results appointed that 94.7% (Hospital A) and 100.0% (Hospital B) of the inanimate surfaces analyzed ($n$ = 243) in general hospitals located in Northern Brazil were contaminated with bacterial pathogens. The contamination of hospital surfaces is dependent on a number of factors including proximity of colonized and/or infected patients to the surfaces analyzed; the physical structure of the hospital; the adoption of antibiotic administration programs; as well as issues with the cleaning and disinfection protocols of surfaces and medical equipment. In the hospitals studied, routine disinfection and cleaning occurs 3 times a day in critical units (Operating Room, Adult Intensive Care Unit, Pediatric Intensive Care Unit) and 2 times a day in semi-critical units (Emergency Room, Internal Medicine Ward, Surgical Ward) using 70º alcohol. Terminal disinfection and cleaning that encompasses all horizontal and vertical surfaces, occurs after bed clearance, and usually occurs weekly in critical units and biweekly in n semi-critical units. While adopting nationally recommended cleaning and disinfection protocols, our results on environmental contamination corroborate those of national surveillance studies carried out in ICUs in Center-Western [26] and Northern Brazil [15]. Consistent with this, international studies [2,5,27] have shown that only less than 50% of hospital surfaces are properly cleaned and disinfected with germicides [28-30]. These alarming findings strongly suggest that the hospital environment can act as a reservoir of pathogens and enable their cross-transmission to the patient.

While bacteria, viruses, and fungi can commonly thrive in environmental reservoirs [21,31,32], the etiology of microbial contamination in our study was specific to each hospital. Hospital A had surfaces and equipment contaminated with human pathogens, whereas Hospital B was contaminated with environmental bacterial contaminants. These differences may be explained by several factors [19,33] such as hospital size and cleaning/disinfecting regimens. Hospital A is a large hospital providing medium and high complexity healthcare assistance, with an ICU demanding appropriate cleaning/disinfection. In addition, admitted patients have an increased length of hospital stay and are often prescribed antibiotics indiscriminately. A combination of these circumstances may lead to selection of nosocomial pathogens such as MRSA, *P. aeruginosa* and Enterobacteria, which ultimately contaminate the surrounding environment. On the other hand, Hospital B is a medium-sized hospital providing medium complexity healthcare assistance, which does not present all the aggravating factors described for Hospital A. The medium complexity is based on ambulatory attending and is composed of actions and services aimed at attending the main health problems of the population, whose complexity of care in clinical practice demands the availability of specialized professionals and the use of technological resources for diagnostic support and treatment. As examples we cited The Reference Center for Men's Health and The Center for Children's Specialties in Campo Grande, Brazil. In Hospital B, admitted patients have a better prognosis and have not an increased length of hospital stay, so the presence of common bacterial contaminants on hospital surfaces (e.g., Gram-positive Bacilli and *Micrococcus* spp.), which are widely distributed in the environment, is justified [21]. The bacterial contamination present therein may result from the patient's transient skin microbiota, even though opportunistic infections may develop in immunocompromised individuals [21].

*S. aureus* was the main microorganism recovered from the surfaces and equipment of both Hospital A (61.8%) and B (68.6%). In Hospital A, there was a predominance of *S. aureus* on door knobs in the ER (53.3%), heart monitors (83.3%) and medication tables (77.8%) in the AICU, and stethoscopes (100.0%) and mattresses (75.0%) in the PICU. In Hospital B, *S. aureus* was the main pathogen recovered from door knobs and bed headboards (66.7%), saline stands (100.0%) and mattresses (66.7%). These percentages are similar to those reported in the national literature [4] for mattresses (72.0%) and higher than international percentages [34] for beds (34%) and door knobs (26.0%). These differences can be attributed to the greater effectiveness of international cleaning and disinfection protocols. Our data show that highly touched surfaces are prone to be contaminated with *S. aureus*, a microorganism commonly found on the skin. This suggests that the hands of health professionals, the patients themselves or visitors were the main vector of contamination of surfaces in the units analyzed, although this was not investigated. Surfaces and equipment can be contaminated and, if not are properly disinfected, re-contaminate sanitized hands during the interruption of patient care to touch the surfaces and equipment such as keyboards, tables, door knobs, stethoscopes etc. The surfaces can become persistently contaminated by pathogens [5,7,19] and the hospital environment turns into a reservoir of highly transmissible microorganisms.

In our study, surfaces of the PICU were found to be contaminated with CoNS (24,4%), as observed in the Neonatal Intensive Care Unit in the California, USA, where CoNS were the more abundant on the surfaces [35]. CoNS are main cause of late-onset sepsis in developed countries[36] and early neonatal infections in Canadá [36], USA [37] and Brazil [38]. Despite the fact that CoNS are not considered pathogenic under normal circumstances, their presence on objects with frequent manual contact (e.g., bedside tables and areas close to patients such as bed headboards and bed frame feet) may pose a risk of contamination and development of infection in hospitalized children. This fact requires attention by local health teams and indicates a need for revision of surface cleaning/disinfection protocols at the PICU in Hospital A.

Our findings showed a high percentage of methicillin-resistant isolates of CoNS and most *S. aureus*. The frequency of MRSA recovered from the ER (28.3%) was similar to that reported in the literature [12] (20.0%), while the frequency of MRSA recovered from the AICU (52.7%) was higher than that found in Brazil [39] (41.8%) and lower than international rates [40] (67.3%). These differences in MRSA rates may be justified by adaptations in the methodology. The presence of MRSA in both hospitals analyzed reflects the importance of this pathogen in the environment [34], as observed in another tertiary hospital in the Middle East in five rooms and two nursing stations in the AICU and PICU. MRSA strains often exhibit multidrug resistance, including resistance to beta-lactam antibiotics, macrolides, fluorquinolones, and exhibit strong virulence as a result of a multiplicity of factors acting simultaneously to evade the host's defenses. Some of which include the production of enzymes and toxins, intracellular invasion and proliferation, dissemination into tissues and organs, and ability to form biofilms [4,27,41-43]. These virulence factors mediate bacterial adhesion to inert hospital surfaces and medical equipment, which may explain the high prevalence rates of MRSA found in this study, although biofilm production was not investigated herein.

The contamination of surfaces and equipment with Gram-negative bacilli is not as studied as the contamination with MRSA, VRE and *C. difficile* [3]. A study [44] reported the presence of MDR Enterobacteria in 22.2% of 18 sampled surfaces, which is consistent with our findings (19.44%) for the AICU of Hospital A. European data [45] showed a high level of contamination by *P. aeruginosa* in sink isolates and tap biofilms. In line with this, our study recovered *P. aeruginosa* from 25% of sink samples, which attests that this pathogen is commonly associated with humid sites in the hospital environment and is the main microorganism isolated in ICUs [46]. In contrast, from a total of seventeen *P. aeruginosa* isolates found in this study, eleven were non-MDR isolates from door knobs of the ER, which is not a humid environment. This may be associated with the presence of *P. aeruginosa*-infected patients in beds near contaminated door knobs. Once more, this reinforces the understanding that health professionals' hands work as a vector of pathogen transmission. In our study, MDR *P. aeruginosa* isolates were recovered at a low frequency from AICU (2.78%) and PICU (2.04%) facilities, which confirm from the literature showing (2,96%) [47] prevalence of MDR *P. aeruginosa* in transplantation unit in Germany.

Even so, our results confirm the imminent risk of contamination of critically ill patients.

In our study, we observed that the AICU of Hospital A was highly contaminated with MDR pathogens. This is in line with observational studies that recovered MDR microorganisms from critical care units [5,27,40]. The environmental contamination of ICUs has been a worrisome issue, since severely ill patients are prone to develop infections. It is fundamental to adopt standard measures to prevent and control hospital-acquired infection, considering the patients' risk of death, the proximity of the beds and the presence of monitoring and support equipment, which are highly touched and susceptible to contamination. It is also worth noting the widespread use of antibiotics in critical units, which select MDR clones both in the patient and in the environment, in addition to the fact that viable MDR bacteria have been isolated from biofilm surfaces and furniture after terminal cleaning of ICUs [5,48]. Evidence has suggested that ICUs are epicenters of MDR microorganisms, which was confirmed in our study.

Adding, these pathogens, mainly MRSA, were commonly (71.4%) isolated from oropharyngeal colonization of patients admitted to Hospital 2 [49] in a study conducted by our team.

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## 5. CONCLUSION

Our results showed a high frequency of contaminated surfaces and equipment, with isolation of difficult-to-treat MDR phenotypes, e.g. MRSA, MR-CoNS, Enterobacteria resistant to 4$^{th}$ generation cephalosporins and MDR *P. aeruginosa.* These pathogens, mainly MRSA, were commonly (71.4%) isolated from oropharyngeal colonization of patients admitted to Hospital 2 in a study conducted by our team. In this investigation, we concluded that current antibiotic use, advanced age and, a previous hospitalization were statistically significant risk factors for the risk of colonization by multidrug-resistant bacteria. The fact of the presence of the same pathogens phenotypes, (e.g., MRSA and others) detected in the patients and in the hospital environment in the same hospital validated our results, although to have disparity in the sample number in this study. These findings raise concern and point to the need to review the protocols of prevention and control of infections in the hospitals analyzed. The cleaning/sanitizing of inert surfaces and equipment and antibiotic dispensing procedures should also be looked at in critical units, since antibiotics can act as a selective force of resistant bacterial strains. Routine and terminal disinfection of environmental surfaces and medical equipment with germicides is recommended in the literature [32] to decrease the frequency and level of contamination, especially highly touched surfaces near the patient. There is a need for greater attention into hand hygiene before and after contact with patients as much as with the areas close to them. These actions together could reduce contamination of hospital surfaces and equipment and the possibility of cross-transmission of pathogens while minimizing the risk of contamination of patients and the development of HAI.

Taken altogether, the association between environmental contamination and the epidemiology of HAI is complex. This study is the pioneer in the Northern region of Brazil, area that is lacking in scientific data, and presented limitations, e.g. absence of use molecular biology tools to identify species, to describe the Staphylococcal cassette chromosome mec (SCCMEC) type of MRSA and presence or absence of extended-spectrum beta-lactamase (ESBL) among the Enterobacteria or *P.aeruginosa* isolates circulating within the hospital environment. The molecular biology would be useful to determine the similarity of strains recovered from surfaces, in order to track the circulation and origin of MDR pathogens during cross-infection. The absence of microbiota analysis from the hands of health professionals was other limitation of this study, since we tried to correlate environmental isolates with their possible sites of origin, e.g. hands of the health care, considering the knowledge of the literature. However, the development of this study established partnerships with hospitals and the possibility of futures researches, that can better understand the correlation of bacterial contamination with the underlying pathology of hospitalized patients close to contaminated surfaces using molecular biology tools. Our results showed a high frequency of contaminated surfaces and equipment, and critical care units acting as reservoir of MDR pathogens, including MRSA, MR-CoNS, Enterobacteria resistant to 4$^{th}$ generation cephalosporins and MDR *P. aeruginosa*. These results may assist health professionals of the analyzed hospitals in the adoption of preventive and control measures for

HAI and reveal the need for reflection on the indiscriminate use of antimicrobials and on the cleaning/sanitizing of inert surfaces and equipment.

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## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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