EPIDEMIOLOGICAL PROFILE AND ANTIMICROBIAL SUSCEPTIBILITY OF MICROORGANISMS ISOLATED FROM NOSOCOMIAL INFECTIONS

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ABSTRACT

The aim of this study was to investigate the main microorganisms involved in nosocomial infection cases between June/2012 and June/2014 in a hospital in Rio de Janeiro, Brazil and determine their antimicrobial susceptibility. This study was developed from the hospital's microbiology lab database. One hundred fifty-nine cultures were analyzed: 71 from surgical incisions, 46 from tracheal fluid, 22 from non-surgical wounds, seven from pleural fluids, five from venous access catheters, three from ascitic fluid, three from pericardial fluid, and two from cerebrospinal fluid. Seventy-six samples (47.80%) had a positive result. Eighty-four microorganisms were isolated. The most commonly isolated bacteria were tested for antimicrobial susceptibility. Coagulase-negative staphylococci were the most frequently isolated bacteria (21.42%), followed by *Staphylococcus aureus* (16.67%), and *Enterobacter* sp. (11.90%). Amoxicillin/clavulanate and oxacillin had the highest levels of resistance. The majority (83.34%) of Coagulase-negative staphylococci had oxacillin resistance and all isolates showed susceptibility to vancomycin. A case of *Enterococcus* sp. vancomycin- resistance was identified and reported. The analyzed samples showed a significant resistance to various classes of antibiotics, highlighting the importance of antimicrobial susceptibility tests for the choice of an appropriate drug to be used for the treatment of the patient and prevention of nosocomial infection.

Keywords: Cross infection; Health profile; Drug resistance; Microbiology;

PERFIL EPIDEMIOLÓGICO E SUSCEPTIBILIDADE ANTIMICROBIANA DE MICRORGANISMOS ISOLADOS DE CASOS DE INFECÇÃO HOSPITALAR

RESUMO

O objetivo deste estudo foi investigar os principais microrganismos envolvidos em casos de infecção hospitalar entre Junho/2012 e Junho/2014 em um hospital no Rio de Janeiro, Brasil e determinar o perfil de susceptibilidade antimicrobiana dos mesmos. O estudo foi desenvolvido a partir do banco de dados do laboratório de microbiologia do hospital. Cento e cinquenta e nove culturas foram analisadas: 71 swabs de incisões cirúrgicas, 46 lavados traqueais, 22 swabs de feridas não-cirúrgicas, sete líquidos pleurais, cinco pontas de cateteres venosos, três líquidos ascíticos, três líquidos pericárdicos e dois líquidos cefalorraquidianos. Setenta e seis amostras (47,80%) resultaram em culturas positivas. Oitenta e quatro microrganismos foram isolados. *Staphylococccus* coagulase negativa foi o microrganismo isolado em maior proporção (21,42%), seguido por *Staphylococcus aureus* (16,67%) e *Enterobacter* sp. (11.90%). Amoxicilina/clavulanato e oxacilina apresentaram os maiores índices de resistência. A maioria (83,34%) dos *Staphylococcus* coagulase-negativa apresentaram resistência à oxacilina e todos os isolados apresentaram susceptibilidade à vancomicina. Um caso de *Enterococcus* sp. resistente à vancomicina foi identificado e reportado. As amostras analisadas mostraram resistência significativa a várias classes de antibióticos, destacando a importância dos testes de sensibilidade aos antimicrobianos para a escolha do medicamento apropriado para ser utilizado no tratamento do paciente e prevenção de infecção nosocomial.

Palavras-chave: Infecção hospitalar; Perfil epidemiológico; Resistência a medicamentos; Microbiologia;

PERFIL EPIDEMIOLÓGICO Y LA SUSCEPTIBILIDAD ANTIMICROBIANA DE MICROORGANISMOS AISLADOS DE INFECCIONES NOSOCOMIALES

RESUMEN

El objetivo de este estudio fue investigar los principales microorganismos implicados en los casos de infección nosocomial entre junio/2012 y junio/2014 en un hospital de Rio de Janeiro, Brazil y determinar su sensibilidad a los antimicrobianos. Este estudio se desarrolló a partir de la base de datos del laboratorio de microbiología del hospital. Se analizaron cien cincuenta y nueve culturas: 71 de incisiones quirúrgicas, 46 de fluidos traqueales, 22 de las heridas no quirúrgicas, siete de los fluidos pleurales, cinco de los catéteres de acceso venoso, tres de líquido ascítico, tres de líquido pericárdico, y dos de fluido cerebroespinal. Setenta y seis muestras (47.80%) tuvieron un resultado positivo. Ochenta y cuatro microorganismos fueron aislados. Los microorganismos más frecuentemente aislados fueron testados cuanto su susceptibilidad antimicrobiana. Estafilococos coagulase negativa fue la bacteria más frecuentemente aislada (21,42%), seguido por *Staphylococcus aureus* (16,67%), y *Enterobacter* sp. (11.90%). Amoxicilina/ácido clavulánico y oxacilina presentaron los más altos niveles de resistencia. La mayoría (83,34%) de los estafilococos coagulase-negativa tenía resistencia a la oxacilina y todos los aislados mostraron susceptibilidad a la vancomicina. Se identificó un caso de *Enterococcus* sp. con resistencia a la vancomicina. Las muestras analizadas mostraron resistencia significativa a varias clases de antibióticos, poniendo de relieve la importancia de las pruebas de susceptibilidad antimicrobiana para la elección de un medicamento apropiado para ser utilizado para el tratamiento del paciente y la prevención de la infección nosocomial.

Palabras clave: Infección hospitalaria; Perfil de salud; Resistencia a medicamentos; Microbiología;

Introduction

Nosocomial infection (NI) is defined as an infection acquired after the patient's admission that manifests itself during hospitalization or after discharge and is related to hospitalization or hospital procedures [1]. Recently, infections acquired within health-care facilities have become more common [2].

NI emerged with the advent of hospitals and has become a large problem for global public health. The morbidity and mortality of patients and hospital costs have increased due to the use of more sophisticated procedures, pathogenic microorganisms, inappropriate antibiotic use, and microbial resistance to such drugs [3]. Three risk factor categories traditionally associated with NI have been described as factors inherent to patient, invasive procedures, and the hospital environment [4].

Worldwide, infectious diseases are responsible for the death of 17 to 20 million people a year. In addition, about 10 million acquire NI and of those, nearly 300 thousand are unable to resist the bacteria [5].

Different pathogens, such as bacteria, fungi, and viruses, cause NI. Bacteria from human microbiota are the most common group of agents. Usually, these bacteria are not a risk to healthy subjects due to low virulence. However, they can cause infection in individuals with compromised clinical conditions, and thus are considered opportunistic bacteria [6]. The most common sites of severe infections in adults are the urinary tract, respiratory and gastrointestinal systems, skin, and soft tissues [7].

The most common agents of NI are Gram negative bacteria (*Escherichia coli*, *Pseudomonas* sp., *Klebsiella* sp., *Proteus* sp., *Enterobacter* sp., *Serratia* sp.) and Gram positive bacteria (*Streptococcus* sp, *Staphylococcus aureus*, *Staphylococcus epidermidis*) [6]. However, the epidemiology of infectious organisms has changed significantly. The relative frequency of Gram negative bacteria isolated from a general population of ICU (Intensive Care Unit) patients has declined, while the pathogenic role of Gram positive organisms, especially *Staphylococcus aureus* and Coagulase-negative staphylococci, has increased significantly [7].

Another medically important group involved in NI are fungi. *Candida albicans* and *Aspergillus* spp. are the most frequent pathogens. Fungi are responsible for about 8% of hospital infections [6].

Regarding viruses, hepatitis B, hepatitis C, enteroviruses, and viruses associated with nosocomial pneumonia are commonly recorded. Viruses represent about 5% of infections [6].

Antibiotics are widely used for the treatment and prophylaxis of infections. These drugs comprise between 30% and 50% of hospital drug spending. Erroneous treatment is another factor contributing to the development of bacterial resistance. After several decades of successful antibiotic therapy, we confront a worrying prospect: the accelerated evolution of antibiotic resistance by important human pathogens [8].

The incidence of nosocomial resistant microorganisms has increased worldwide. Antibiotic resistance has developed as a natural consequence of bacteria's ability to adapt. The indiscriminate use of antibiotics has increased selective pressure and also the opportunity of being exposed to these bacteria. This facilitates the acquisition of resistance mechanisms, making bacterial resistance the main public health problem affecting both developed and undeveloped countries [9,10]. The increase of modern antimicrobials is needed because the process of bacterial resistance is very fast compared to the process of new drug development[11].

It is estimated that about one third of NIs could be avoided. Programs on effective prophylactic measures are among the main challenges for the management of hospitals [12]. To avoid an increase in bacterial resistance, the rational use of antibiotics, especially broad-spectrum antibiotics, needs to be monitored and the adoption of preventive and educational measures need to be put into place [11]. Despite the epidemiological surveillance programs and existing preventive measures, the risk for development of NI remains unacceptably high [13].

From this perspective, the purpose of study is to determine the main microorganisms involved in cases of nosocomial infections between June 2012 and June 2014 in a hospital in Rio de Janeiro, Brazil, and to analyze their sensitivity and resistance to antibiotics.

Methodology

This is an epidemiological, cross-sectional, retrospective study. The study was conducted at Casa de Saúde São Lucas, a private hospital situated in Rio de Janeiro, Brazil. This hospital serves several localities in the state of Rio de Janeiro and is referral center of cardiac surgery for the Sistema Único de Saúde (a governmental healthcare assistance program).

This study was properly approved by the ethics committee of Federal Fluminense University, under the number 00887812.1.0000.5243, process 146.816. This study was developed respecting the rules contained in Resolution number 196/96 of the Brazilian Council of Health. This resolution establishes criteria about beneficence, non-maleficence, autonomy, justice, and equity. The anonymity and confidentiality of the used information were ensured.

The access to the patient database was authorized by the technician responsible for the institution's clinical laboratory The data was collected from records of microbiological cultures and sensitivity tests from the laboratory database. The following data were collected: sex, clinical specimen, date of collection, isolated microorganism, antimicrobials, and presented susceptibility.

Data collection was performed in June 2014. The results from cultures of the following clinical specimens were analyzed: swabs from surgical incisions (71 samples), tracheal lavage fluid (46 samples), swabs of non-surgical wounds (22 samples), pleural fluid (7 samples), venous access catheters (5 samples), ascitic fluid (3 samples), pericardial fluid (3 samples), and cerebrospinal fluid (2 samples). There were 159 cultures in total from patients hospitalized from June 2012 to June 2014.

The microbiological diagnosis, bacterial cultures, and identification were performed using conventional methods described in the literature according Koneman *et al* (2008) [14].

The criteria that were used to diagnose infection were in accordance with the Centers for Disease Control and Prevention and the Brazilian Health Surveillance Agency. [15, 16]

The sensitivity test was performed by the disk diffusion method according to the CLSI, 2014 (Clinical and Laboratory Standards Institute) [17]. The tested antimicrobials differed according to the clinical specimen analyzed.

The data were organized in a Microsoft Office Excel® spreadsheets. The prevalence rates of identified microorganisms and the antimicrobial susceptibility profile were presented as simple percentages. Finally, the data were expressed in tables and discussed according to the relevant literature.
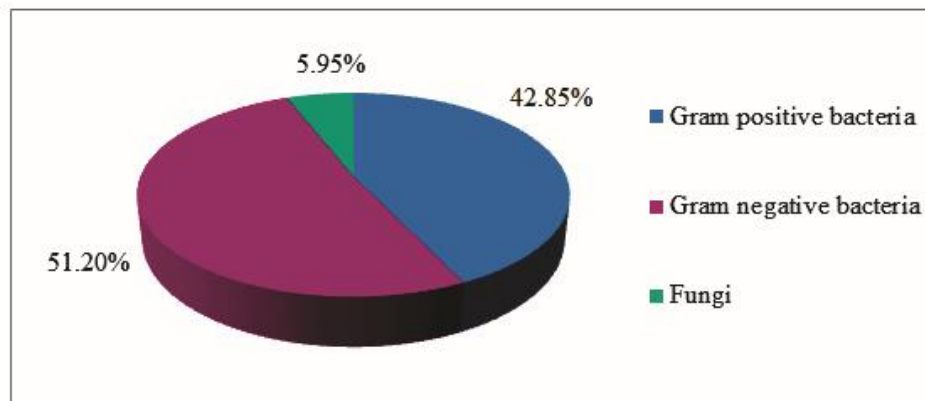
Results

During this study period, the clinical laboratory of Casa de Saúde São Lucas conducted bacteriological tests of 159 clinical samples from suspected cases of nosocomial infections. According to the database, 76 samples (47.80%) had positive cultures, whereas 83 samples (52.20%) resulted in negative cultures.

Of the total positive cultures, 44.74% (34 samples) were from female patients and 55.26% (42 samples) were from male patients.

Of the 159 cultivated samples, 71 were swabs from surgical incisions, 46 from tracheal lavage fluid, 22 were swabs of non-surgical wounds, 7 from pleural fluid, 5 from venous access catheters, 3 from ascit fluid, 3 from pericardial fluid, and 2 from cerebrospinal fluid.

This study shows the presence of Gram-negative bacilli in 43 samples (51.20%), Gram-positive cocci in 36 samples (42.85%), and fungi in 5 samples (5.95%), according to Figure 1.

Figure 1 - Profile of microorganisms isolated from nosocomial infections of a private hospital in Rio de Janeiro, Brazil, from June 2012 to June 2014.
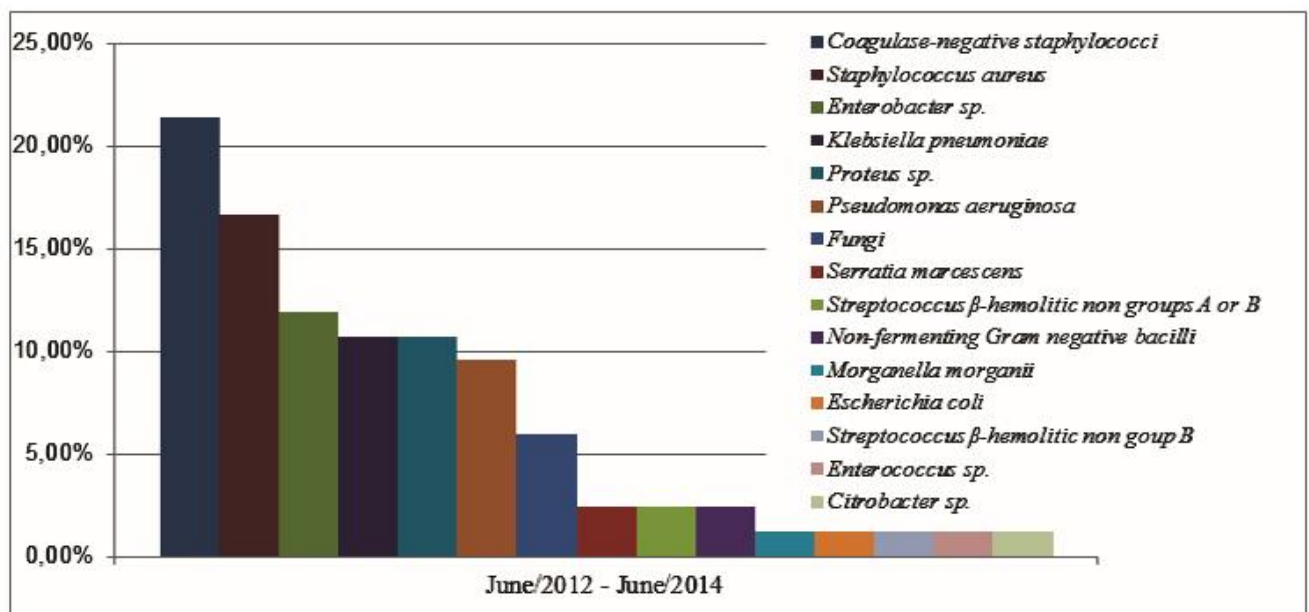

Reference: The authors

Bacterial cultures had 84 isolated microorganisms, with 21.42% Coagulase-negative staphylococci (18 samples), 16.67% *Staphylococcus aureus* (14 samples), 11.90% *Enterobacter* sp. (10 samples), 10.71% *Klebsiella pneumoniae* (9 samples), 10.71% *Proteus* sp. (9 samples), 9.55% *Pseudomonas aeruginosa* (8 samples), 5.95% fungi (5 samples), 2.38% *Serratia* sp. (2 samples), 2.38% β-hemolytic *Streptococcus* non-group A or B (2

samples), 2.38% Non-fermenting Gram negative bacilli (2 samples), 1.19% *Morganella morganii* (1 sample), 1.19% *Escherichia coli* (1 sample), 1.19% β-hemolytic *Streptococcus* group B (1 sample), 1.19% *Enterococcus* sp. (1 sample), 1.19% *Citrobacter* sp. (1 sample), according to Figure 2.

Figure 2 - Distribution percentage of isolated microorganisms in patients of a private hospital in Rio de Janeiro, Brazil, from June 2012 to June 2014.

Reference*: The authors

The positive swabs from surgical incisions samples (33 samples) were comprised of 24 isolated Gram positive cocci, 8 Gram negative bacilli, and 2 fungi.

Of the 46 total samples of tracheal lavage fluid, microbial growth was observed in 28 specimens (60.86%) and were identified as 28 Gram negative bacteria, 2 Gram positive cocci, and 2 fungi.

Half of the non-surgical wound swabs had culture growth (11 samples). Isolated Gram positive and Gram negative bacteria were found in the same proportions.

Regarding the tip cultures from venous catheters, it was observed that 60% of the cultures (3 samples) showed microbial growth (1 Coagulase-negative staphylococci , 1 *Staphylococcus aureus*, and 1 fungi).

No bacterial growth was observed on the two pericardial fluid samples (66.66%). The only positive pericardial fluid sample showed isolated Coagulase-negative staphylococci.

There was no growth of microorganisms in the pleural fluid samples (7 samples), ascitic fluid (3 samples), and cerebrospinal fluid (2 samples).

After isolation of microorganisms, the most common bacterial species in the population of positive patients were subjected to antibiotic susceptibility testing, or antibiogram.

The majority (83.34%) of Coagulase-negative staphylococci had oxacillin and ciprofloxacin resistance, 88.89% had amoxicillin/clavulanate resistance and 72.23% had cefepime (large spectrum 4th generation cephalosporin) resistance (Table 1).

Table 1 - Percentage of resistance of clinical isolates of Coagulase-negative staphylococci and *Staphylococcus aureus* to antibiotic tests.

Antibiotics	Coagulase-negative staphylococci Resistance rate (%)	*Staphylococcus aureus* Resistance rate (%)
Amikacin	5.56	35.71
Amoxicillin/Clavulanic acid	88.89	50.00
Cefepime	72.23	57.14
Cephalothin	44.45	no tested
Ciprofloxacin	83.34	42.85
Clindamycin	55.56	28.57
Gentamicin	27.78	21.42
Oxacillin	83.34	57.14
Teicoplanin	0.00	0.00
Vancomycin	0.00	0.00

Reference*:* The authors

Staphylococcus aureus isolates exhibit high resistance to oxacillin and cefepime, and resistance rates to amoxicillin/clavulanic acid, ciprofloxacin and amikacin were noted (Table 1).

Enterobacter sp. showed 90% susceptibility to the antibiotics piperacillin/tazobactam. *Klebsiella pneumoniae* showed 100% sensitivity to meropenem and imipenem.

Proteus sp. showed wider resistance pattern.

Pseudomonas aeruginosa showed 62.50% resistance rate to ceftazidime and cefepime (3th and 4th generation cephalosporin), according to Table 2.

A case of *Enterococcus* sp. vancomycin- resistance was identified and reported.

Table 2 - Percentage of resistance of clinical isolates of Gram negative bacteria to antibiotic tests.

Antibiotics	*Enterobacter sp.* Resistance rate (%)	*Klebsiella pneumoniae* Resistance rate (%)	*Proteus* sp. Resistance rate (%)	*Pseudomonas aeruginosa* Resistance rate (%)
Amikacin	30.00	22.23	55.56	25.00
Cefepime	20.00	no tested	55.56	62.50
Cefoxitin	no tested	66.67	44.45	no tested
Ceftazidime	20.00	44.45	44.45	62.50
Ceftriaxone	30.00	33.34	66.67	no tested
Ciprofloxacin	40.00	33.34	55.56	25.00
Gentamicin	20.00	11.12	77.78	12.50
Imipenem	10.00	0.00	33.34	12.50
Levofloxacin	20.00	22.23	44.45	12.50
Meropenem	10.00	0.00	22.23	12.50
Piperacillin/Tazobactam	10.00	33.34	11.12	12.50
Polymyxin B	20.00	11.12	22.23	no tested

Reference*:* The authors

Among the identified microorganisms, vancomycin and teicoplanin are the most sensitive antibiotics, whereas amoxicillin/clavulanic acid, oxacillin, cephalosporins and ciprofloxacin showed the largest resistance profile.

Discussion

Coagulase-negative staphylococci (CoNS) was the microorganism most frequently isolated from cultures in this study (21.42%). CoNS are commonly found on the skin and mucous membranes of humans. In the past, they were considered of little clinical importance. However, the incidence of these microorganisms is increasing. So, CoNS are now recognized as opportunistic agents that cause nosocomial and community infections [18]. It is known that this bacteria's pathogenicity is associated with antimicrobial resistance, which results in a bigger problem, since the number of viable antibiotics for the treatment of multiresistant strains is minimal [5].

Among the microorganisms found, *Staphylococcus aureus* ranks as second most common (16.67%). The distribution of *S. aureus* is very wide. The organism can be found in the environment and humans are its main reservoir. This bacterium is present in various parts of the body such as the nasal cavity, throat, intestines, and skin. Nostrils have the highest rate of colonization, where the prevalence is around 40% in the adult population. This asymptomatic colonization has high clinical importance. Once the nostrils have been colonized, the individual can contaminate their own hands and become a vehicle for contact infection [19]. This bacterium has been isolated with high frequency in hospitals and also in health professionals. The transmission to patients occurs through direct or indirect contact. Thus contaminated professionals and hospitals become a risk factor for the development of opportunistic infections [20].

From positive cultures, 11.90% of *Enterobacter* sp., 10.17% of *Klebsiella pneumoniae* and 10.17% of *Proteus* sp. were isolated. The bacteria of the *Enterobacteriaceae* family are responsible for 50% of nosocomial infections. The bacteria of this family are distributed worldwide. Most inhabit the intestines of humans and animals as either members of the normal flora or as infectious agents. Species of *Enterobacter* and *Klebsiella* may persist for long periods and multiply in non-fecal environments [21, 22].

The *Enterobacter* species are found in the natural environment and can colonize on the mucosal surfaces of mammals. *Enterobacter aerogenes* and *Enterobacter cloacae* are commonly isolated species in biological materials. Both species are often isolated from human feces and animals feces, but there are no reports of these bacteria being enteric

pathogens. Occasionally, they can be isolated as opportunistic agents in the urinary tract and the secretional, respiratory, and genitourinary systems of sick individuals, in wounds, or even in sepsis [21, 23].

Klebsiella pneumoniae can be found in water, soil, plants, and sewage. This bacterium can be found colonizing the oropharynx and feces of healthy people. Members of the genus *Klebsiella* are opportunistic pathogens which can cause bacteremia, pneumonia, urinary problems, and other severe infections in humans [21,24,25].

Proteus are Gram negative bacilli of the *Enterobacteriaceae* family. These bacteria may cause opportunistic infections, mainly in immunocompromised patients. The species *Proteus vulgaris* and *Proteus mirabilis* are widely distributed in nature. Both species are present in the intestines of various animals, including most of the human population [21]. In conditions favorable for their proliferation, the microorganism can act as an important pathogen, particularly in geriatric, psychiatric, paraplegic, and immunocompromised patients[26].

Pseudomonas aeruginosa was isolated in 9.55% of the cultures. This bacterium usually comes from humid environments, transmitted by contact and lack of hygiene [27]. Infections caused by *Pseudomonas aeruginosa* are commonly of hospital origin. During the last four decades, the incidence of this microorganism was responsible for 10% of all nosocomial infections. Most hospital equipment and materials, particularly liquids, may serve as a reservoir for *Pseudomonas* sp. This may facilitate the spread throughout the hospital and community through patient transfers or post-discharge [5].

Surgical site infections are a common infection related to health care in Brazil. Such infections rank third of all infections in the health services and 14% to 16% of those infections found in hospitalized patients [28]. Microorganisms commonly associated with surgical site infection are *Staphylococcus aureus*, coagulase negative *Staphylococcus*, and Gram negative bacilli [29].

The skin is the main source for colonization and infection of venous access catheters. The bacteria that are on the skin of the patient migrate along the surface. The distal end of the catheter is colonized and results in infection. The Gram positive cocci have emerged as the main agents of such infections [30].

Skin infections often arise due to epidermis rupture. Epidermis injury is an opportunity for common microorganisms to develop in pathogenic form. The genus *Staphylococcus*

comprises a heterogeneous group of bacteria. Although they are normal human flora, they can start infectious processes in the skin, soft tissues, and wounds [31].

Before the antibiotic era, *Streptococcus* from infectious pulmonary processes were commonly identified in purulent pericarditis agents. After the introduction of antibiotics, this profile has been modified. *Staphylococcus* has become the most frequently isolated microorganisms, followed by Gram-negative agents. Fungi are rare [32].

Fungal hospital infections have a lot of importance. *Candida* yeasts are the largest hospital infection agents and represent a challenge to the survival of patients with serious disease or who are in the postoperative period. US hospitals reported nosocomial pathogen *Candida* as the 6th and the 4th most common cause of bloodstream infections acquired in hospitals. This was highlighted due to the progressive increase in the number of infections and the high morbidity and mortality rates [6].

Factors related to the increased isolation of fungi are the use of broad-spectrum antibiotics, central venous catheters, parenteral nutrition, dialysis, and administration of corticosteroids. In view of these conditions, it is recommended to monitor the mycological tests of patients' biological samples, such as blood, sputum, venous access catheters, peritoneal fluid, and urine [6,7].

Regarding sensitivity and resistance of microorganisms, vancomycin and teicoplanin had low microbial resistance rate, whereas oxacillin, amoxicillin/clavulanic acid, cephalosporins and ciprofloxacin showed high resistance rates.

The increase in infections caused by strains resistant to oxacillin, especially in hospitals, requires intravenous treatment with glycopeptides, such as vancomycin and teicoplanin. Therefore, this is the antimicrobial treatment of choice for infections caused by MRSA (methicillin resistant *Staphylococcus aureus*) [33].

Due to the introduction of penicillin and the selective pressure from antimicrobials, *Staphylococcus aureus* has become one of the main microorganisms associated with nosocomial infections [34]. Coagulase-negative negative staphylococci pathogenesis is associated with its resistance to antibiotics, production of invasive enzymes, and biofilm formation. This strain is often associated with colonization of catheters and implants and is the etiologic agent of several nosocomial infections [35].

Bernardes *et al*. [33] analyzed the sensitivity of isolated strains of coagulase-positive staphylococci to vancomycin and teicoplanin and achieved 100% sensitivity. The same was observed in this work.

In positive cultures, Coagulase-negative staphylococci was the main isolated microorganism. It showed 83.34% resistance to oxacillin. However, all isolates were susceptible to vancomycin.

Custodio *et al.* [36] evaluated the microbiota of the hands of health professionals. They found that coagulase negative was the main strain found among the isolated microorganisms and that it was 75% resistant to oxacillin.

With the emergence of CoNS strains resistant to oxacillin or methicillin, the incorrect use of drugs to treat infections becomes an unaffordable treatment. When there is no apparent resistance to oxacillin, it is the drug of choice for treatment of staphylococcal infections in hospitals. The correct determination of a staphylococci sample's susceptibility to oxacillin is key. The failure to detect this resistance can result in ineffective treatment and the unnecessary and indiscriminate use of vancomycin and other broad spectrum antibiotics in hospitals [18].

Staphylococcus aureus has occupied a prominent rank in the etiology of nosocomial infections due to its high versatility in acquiring antimicrobial resistance [37]. Strains of *Staphylococcus aureus* that are resistant to methicillin are also resistant to first generation cephalosporins, so the alternative therapies for serious infections are glycopeptides, oxazolidinones, and streptogramins [38]. The data describe considerable percentages of resistance to antimicrobials. Oxacillin showed resistance rate of 57.14% cefepime 57.14%, and ciprofloxacin 42.85%.

Previous studies have shown that strains of *Staphylococcus aureus* isolated from ICU patients have a susceptibility to vancomycin (100%) and a low sensitivity to oxacillin [39]. This study obtained 100% sensitivity to vancomycin, while oxacillin resistance presents itself variably according to the region. Thus, resistance must be analyzed according to the hospital [40].

Klebsiella pneumoniae showed 100% sensitivity to meropenem and imipenem antibiotics and 66.67% resistance to cefoxitin. The resistance of these bacteria to antimicrobials has become a public health problem in recent years. *Klebsiella pneumoniae* species can produce β-lactamases encoded by chromosomes or plasmid. Because of potential induction of β-lactamases in enterobacteria, the use of cefoxitin should be restricted to surgical prophylaxis, and in this situation the time of use should not exceed 48 hours [41].

Proteus sp. showed a wider resistance pattern to antimicrobials. This bacterium has increasing importance in clinical practices. It is an infectious agent that is difficult to eradicate

and a microorganism with ability to produce β-lactamases of extended spectrum. Therefore, its control in the hospital environment has become essential. This microorganism is a frequent cause of nosocomial infections, especially in the patients of intensive care units. In this environment, it has been responsible for autoinfection, antimicrobial selection, bacteremia cases, and transmission by the hands of healthcare professionals. The literature mentioned the importance of this microorganism in the infections of immunocompromised patients and reports its increasing resistance to many antibiotics in recent years. The identification of this bacterium as a pathogen of nosocomial infection outbreaks has increased over time. This organism has a narrow antimicrobial susceptibility profile, due to production of β-lactamases expanded spectrum [42].

Enterobacter sp. showed variable sensitivity. The resistance of these microorganisms to multiple antibiotics explains its emergence among hospital infections. *Enterobacter* has natural resistance to β-lactam antibiotics such as ampicillin, amoxicillin, amoxicillin/clavulanic acid, and first-generation cephalosporins. They also easily develop antibiotic resistance to second and third generation cephalosporins due to beta-lactamase induction [43].

The resistance of *Pseudomonas aeruginosa* to various groups of antibiotics characterizes it as typical organism of nosocomial infections [27]. Overall, cephalosporins and carbapenems had higher percentages of activity, and the classes of aminoglycosides and fluoroquinolones had lower activity [42]. In this study, *Pseudomonas aeruginosa* showed 87.50% sensitivity in the presence of levofloxacin, imipenem, meropenem, piperacillin/tazobactam, and gentamicin, and 62.50% resistant to cephalosporin. The analyzed data reiterated that the incidence of microorganisms and their antimicrobial susceptibility vary according to region.

Although it normally colonizes the gastrointestinal tract and the female genital tract, *Enterococcus* sp. plays an important role in hospital infections. The high prevalence of Coagulase-positive negative staphylococci and the consequent use of vancomycin in Brazilian hospitals, the incorrect use of antimicrobials, and the poor conditions of public health institutions predisposes the appearance of intermediate or shortened susceptibility of strains to vancomycin [45].

Colonization or infection with vancomycin-resistant enterococci (VRE) has been linked to a variety of factors, including length of hospital stay, underlying disease, and liver transplantation. Patients colonized with VRE carry the organism in the intestinal flora where

it can remain colonized for prolongêd periods. After introduction into a given hospital, *Enterococcus* has a great ability to spread, affecting various sectors and creating an endemicity profile that makes it very difficult to eradicate [46]. In this study, we detected the presence of a sample of vancomycin-resistant *Enterococcus* sp. that was also sensitive to penicillin.

Hospital infection is a clinical entity with multiple factors involved. The need to reduce and control the incidence culminates in the application of preventive measures, education, and epidemiological control. These measures are intended, by means of a collective awareness, to control infection rates and reduce them to acceptable limits [5]. The epidemiological information is a key tool for planning, implementation, and evaluation of health actions. Bacterial identification in cultures and the analysis of their susceptibility analysis provide important indicators for mortality reduction through a rational antibacterial therapy [21]. This is a basic stage for establishing strategies regarding the appropriate use of antimicrobials. This allows a proper diagnosis and for possible failures in health services to be corrected [47].

Conclusion

The results showed high variation in antimicrobial resistance profiles and the occurrence of multidrug resistance among some studied strains, highlighting the importance of antimicrobial susceptibility tests for the choice of an appropriate drug to be used for the treatment of the patient and prevention of nosocomial infection.

The analyzed samples showed a significant resistance to various classes of antibiotics. Coagulase negative staphylococci was the most frequently isolated microrganism. It showed significant resistance rates compared to antibiotics of choice: amoxicillin/clavulanic acid, oxacillin, and ciprofloxacin. *Staphylococcus aureus is* a typical organism in hospital infections. In this study, it showed multiple antimicrobials resistance, like oxacillin, amoxicillin/clavulanic, and cephalosporin. *Proteus* showed wider resistance pattern, this bacterium has increasing importance in clinical practices.

This study reaffirm the importance of an effective program for infection control with the involvement of healthcare professionals. Nosocomial infection control measures are

important to prevent emergence and spread of multi-resistant bacteria within the hospital environment.

Acknowledgements

References

1.   Azambuja EP, Pires DP, Vaz MRC. Prevenção e controle da infecção hospitalar: as interfaces com o processo de formação do trabalhador. Texto & Contexto Enferm.2005; 13:79-86.

2.   Teixeira DC, Pedro FL, Carneiro M. Infecção hospitalar na visão de enfermeiros da Santa Casa de Caridade de Bagé – RS. Revista de Epidemiologia e Controle de Infecção 2012; 2(1):14-16.

3.   Carvalho MM, Moura MEB, Nunes MRC, Araújo TME, Monteiro CFS, Carvalho LRBM. Infecções hospitalares nas Unidades de Terapia Intensiva em um hospital público. Revista Interdisciplinar UNINOVAFAPI, Teresina 2011; 4(4):42-48.

4.   Banderó VC, Reschke CR, Hörner R. Perfil epidemiológico das infecções hospitalares na Unidade de terapia intensiva infantil do Hospital de Caridade e Beneficência de Cachoeira do Sul, RS, Brasil. RBAC 2006; 38(4):267-270.

5.   da Mata Abegg PTG, Silva LL. Controle de infecção hospitalar em unidade de terapia intensiva: estudo retrospectivo. Semina: Ciências Biológicas e da Saúde 2011; 32(1):47-58.

6.   Levy CE, Von Nowakonski A, Mendes CMF *et al*. Manual de microbiologia clínica para o controle de infecção em serviços de saúde. Módulo V. Detecção e identificação das bactérias de importância médica. Brasília: Agência Nacional de Vigilância Sanitária. Ministério da Saúde, 2004.

7.   Moraes AAP, Santos RLD. Infecções em UTI Geral de um Hospital Universitário. Revista Brasileira de Terapia Intensiva 2003; 15(4):135-141.

8.   Carneiro LC, Carvalhares TT, Pesquero MA, Quintanal CQ, Feitosa SB, Filho JE, Oliveira MAC. Identificação de Bactérias Causadoras de Infecção Hospitalar e Avaliação da Tolerância a Antibióticos. NewsLab 2008; 86:106-114.

9.   Garcia LM, César ICO, Braga CAB, Souza GAAD, Mota EC. Perfil epidemiológico das infecções hospitalares por bactérias multidrogarresistentes em um hospital do norte de Minas Gerais. Revista de Epidemiologia e Controle de Infecção 2013; 3(2):45-49.

10.   de Queiróz Santos NQ. A resistência bacteriana no contexto da infecção hospitalar. Texto Contexto Enfermagem 2004; 13:64-70.

11.   Grillo VTRS, Gonçalvez TG, Júnior JC, Paniágua NC. Incidência bacteriana e perfil de resistência a antimicrobianos em pacientes pediátricos de um hospital público de Rondônia, Brasil. Revista de Ciências Farmacêuticas Básica e Aplicada 2013; 34(1):117-123.

12.   Alves LNS, Oliveira CR, Silva LAP, Gervásio SMD, Alves SR, Sgavioli GM. Hemoculturas: estudo da prevalência dos microrganismos e o perfil de sensibilidade dos antibióticos utilizados em Unidade de Terapia Intensiva. J Health Sci Inst 2012; 30(1):44-47.

13.   Dal-Bó K, Silva RM, Sakael TM. Infecção hospitalar em uma unidade de terapia intensiva neonatal do Sul do Brasil. Rev Bras Ter Intensiva 2012; 24(4):381-385.

14.   Winn WC, Allen SD, Janda WM, Koneman EW, Procop GW, Schreckenberger PC, Woods GL. Koneman, diagnóstico microbiológico: texto e atlas colorido. 6ª edição. Rio de Janeiro: Guanabara Koogan, 2008.

15.   Brasil. Agência Nacional de Vigilância Sanitária Critérios Diagnósticos de Infecção Relacionada à Assistência à Saúde. Brasília: Ministério da Saúde, 2013.

16.   Horan TC, Andrus M, Dudeck MA CDC/NHSN surveillance definition of health-care associated infection and criteria for specific types of infections in the acute care setting. Am J Infect Control 36:309-32. Erratum in Am J Infect Control 2008; 36:655.

17.   Clinical and Laboratory Standards Institute. Performance Standards for Antimicrobic Disc Susceptibility Tests: Approved Standard, 2006.

18.   Bezerra AB, Araújo EC, Alves WCL, Oliveira RA. *Staphylococcus* coagulase negativa resistente a oxacilina no Hospital Regional Público do Araguaia – Pará Araguaia: Faculdade De Ensino Superior Da Amazônia Reunida; 2010.

19.   Santos AL, Santos DO, Freitas CC, Ferreira BLA, Afonso IF, Rodrigues CR, Castro HC. *Staphylococcus aureus*: visitando uma cepa de importância hospitalar. Bras. Patol. Med. Lab 2007; 43(6):413-423.

20.   Dias RS, Zuccoli PC. Infecção hospitalar por *Staphylococcus aureus* e fatores de risco para o desenvolvimento da Síndrome do Choque Tóxico (TSST-1). NBC - Periódico Científico do Núcleo de Biociências, 2(3):27-37.

21.   Alves RPR. Estudo das principais enterobactérias responsáveis pelas infecções no âmbito hospitalar e comunitário: uma revisão. Paraíba: Universidade Estadual da Paraíba; 2011.

22.   Trabulsi LR, Alterthum F, Gompertz OF, Candeias JAN. Microbiologia. 3ª ed. São Paulo: Atheneu, 2002.

23.   CEFAR, Diagnóstica. Bactérias do grupo coliforme. Informativo Cefar de Microbiologia 2005; 2(11):1-4.

24.   Desimoni MC, Esquivel GP, Merino LA. Fecal colonization by extended spectrum betalactamase-producing *Klebsiella pneumoniae* in a neonatal intensive care unit. Enferm Infecc Microbiol Clin. 2004; 22(9):507-11.

25.   Martinez J, Martínez L., Rosenblueth, M, Silva J., Martínez-Romero E. How are gene sequence analyses modifying bacterial taxonomy? The case of Klebsiella. International Microbiology 2004; 7(4):261-268.

26.   O'Hara CM., Brenner FW, Miller JM. Classification, identification, and clinical significance of *Proteus*, *Providencia*, and *Morganella*. Clinical microbiology reviews 2000; 13(4), 534-546.

27.   Silva NFV, Kimura CA, Coimbra MV. Perfil de sensibilidade antimicrobiana das *Pseudomonas aeruginosa* isoladas de pacientes da unidade de tratamento intensiva de um hospital público de Brasília. Revista de Divulgação Científica Sena Aires 2012; 1(1):19-24.

28. Brasil. Ministério da Saúde. Agência Nacional de Vigilância Sanitária. Definição dos Critérios Nacionais de Infecções Relacionadas à Assistência à Saúde. Prevenção de Infecção do Sítio Cirúrgico (ISC). Brasília: ANVISA/MS, 2009.

29. Assunção TP, Pontes BCD, Damasceno CAV. Prevalência de infecções em suturas de cirurgias de revascularização do miocárdio. Rev Bras Cir Cardiovasc 2011; 26(1):43-46.

30. Esmanhoto CG, Taminato M, Fram DS, Belasco AGS. Barbosa D A. Microrganismos isolados de pacientes em hemodiálise por cateter venoso central e evolução clínica relacionada. Acta Paul Enferm 2013; 26(5):413-420.

31. Martins MA, Bachion MM, Santos SLV et al.. Análise da ocorrência de infecção por Staphylococcus sp. em amostras de úlceras venosas de perna de pacientes atendidos em unidades municipais de saúde de Goiânia. In: Anais do VIII Congresso de Pesquisa, Ensino e Extensão; 2011; Goiás.

32. Martí V, Garrido P, Seixo F, Peset A, Ginelli A. Pericardite purulenta com tamponamento cardíaco causada por agentes anaeróbios. Rev Port Cardiol 2008; 27(7-8):959-964.

33. Bernardes RC, Jorge AOC, Leão MVP. Sensibilidade a oxacilina, vancomicina e teicoplanina de Staphylococcus coagulase positivos isolados de pacientes hospitalizados em São José dos Campos. Revista Biociências 2004; 10(5):73-78.

34. Fernandes AT, Ribeiro-Filho N, Barroso EDAR, Graziano KU, Cavalcante NJF, Lacerda RA. Conceito, cadeia epidemiológica das infecções hospitalares e avaliação custo-

benefício das medidas de controle. In: FERNANDES, A. T. (ED.). Infecção hospitalar e suas interfaces na área da Saúde. São Paulo: Atheneu; 2000. p. 215-265.

35. Michelim L, Lahude M, Araújo RP, Giovanaz DSH, Muller G, Delamare APL, Costa SOP, Echeverrigara YS. Pathogenicity factors and antimicrobial resistance of Staphylococcus epidermidis associated with nosocomial infections occuring in intensive care units. Brazilian Jornal of Microbiology 2005; 36(1):17-23.

36. Custódio J, Alves J F, Silva FM, Dolinger EJO, Santos JGS, Brito D. Avaliação microbiológica das mãos de profissionais da saúde de um hospital particular em Itumbiara, Goiás. Revista de Ciências Médicas 2009; 18(1):7-11.

37. Moreira ACMG, Santos RR, Benedo J. Prevalência e perfil de sensibilidade de *Staphylococcus aureus* isolados em pacientes e equipe de enfermagem. Ciênc. cuid. saúde 2013; 12(3):572-579.

38. Moura JSD, Diniz A, Salgado M *et al*. Fatores de risco associados à infecção e mortalidade por *Staphylococcus aureus* resistentes a oxacilina em um hospital de referência para doenças infectocontagiosas de Goiânia-GO, Brasil. Mundo saúde 2011; 35(1):84-90.

39. Moura MEB, Campelo SMA, Brito FCPB. Infecção hospitalar: estudo de prevalência em um hospital público de ensino. Rev. Bras. enferm 2007; 60(4):4116-4121.

40. Amyes SGB, Gemell C.G. Antibiotic resistance. J Med Microbiol 1997; 46:436-70.

41. Medeiros EAS, Machado A, Ferraz AAB, Ferraz E, Arruda E, Nobre J, Konkewicz LR, Pimentel ML, Leão MTC, Trabasso P, Grimbaum R. Prevenção da Infecção Hospitalar. Projeto Diretrizes 2001;16-17.

42. Michelim L. Abordagem Biotecnológica em Proteus mirabilis. Caxias do Sul: Universidade de Caxias do Sul; 2008.

43. González-Mejía EB, Valenzuela EM., Mantilla-Anaya JR, Leal-Castro AL *et al*. Resistance to cefepime in *Enterobacter cloacae* isolates from hospitals in Bogota, Colombia. Revista de Salud pública 2006; 8(2):191-199.

44. Paviani ER, Stadnik CB, Heinek I. Estudo da epidemiologia e perfil de sensibilidade da Pseudomonas aeruginosa. Infarma 2004; 15(11-12).

45. Oliveira GA, Mamizuka EM. Como triar e identificar *Staphylococcus aureus* com sensibilidade reduzida a vancomicina. Laes & Haes 2000; 21(126):110-120.

46. Renner JDP, Carvalho ED. Microrganismos isolados de superfícies da UTI adulta em um hospital do Vale do Rio Pardo – RS. Revista de Epidemiologia e Controle de Infecção 2013; 3(2):40-44.

47. Mesquita KO, Almeida ECB, Lima GK, Dias ICX, Silva VEA, Linhares MSC. Perfil epidemiológico das infecções hospitalares na UTI neonatal da santa casa de misericórdia de sobral no ano de 2009. SANARE-Revista de Políticas Públicas 2010; 9(2).