

ORIGINAL ARTICLE

PROFILE OF MICROORGANISMS ISOLATED FROM INPATIENTS IN A HOSPITAL IN PARANÁ

Amanda Ribeiro Espírito Santo¹, Ricardo Castanho Moreira², Leopoldo Sussumu Matsumoto³, Eliane da Luz Furtado⁴, Cláudio Queniti Hirai⁵

ABSTRACT

Objective: to identify the profile of microorganisms isolated from biological samples of patients admitted to a hospital in Paraná.

Method: descriptive and retrospective research, with data collection from a secondary source, developed in a hospital in Paraná (Brazil), referring to 2018. The study population consisted of biological samples from 226 patients admitted during the period from January 1st to December 31st, 2018. Data analysis was performed using descriptive statistics.

31st, 2018. Data analysis was performed using descriptive statistics. Result: the mean age of the patients was 60.41 years old and the majority were women. Of the biological samples, 70 had positive results for bacterial identification. The tracheal aspirate showed greater positivity to microorganisms, with 30%. The most frequent microorganisms were *Escherichia coli*, coagulase-negative *Staphylococcus*, *Pseudomonas*, *Staphylococcus aureus*, *Enterobacter sp.* and *Proteus sp.*

Conclusion: the study contributes to hospital care, as it produces knowledge of the microbiological profile and its resistance to antibiotics.

DESCRIPTORS: Epidemiological Profile; Microbiota; Hospital Infection; Bacterial Infections; Microbial Drug Resistance.

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PERFIL DE MICRORGANISMOS ISOLADOS DE PACIENTES INTERNADOS EM UM HOSPITAL DO PARANÁ RESUMO

Objetivo: identificar o perfil de microrganismos isolados de amostras biológicas de pacientes internados em um Hospital do Paraná.

Método: pesquisa descritiva e retrospectiva, com coleta de dados de fonte secundária, desenvolvida em um hospital do Paraná (Brasil), referente a 2018. A população do estudo foi composta por amostras biológicas de 226 pacientes admitidos durante o período de 01 de janeiro a 31 de dezembro de 2018. A análise dos dados foi realizada pela estatística descritiva. Resultado: a idade média dos pacientes foi de 60,41 anos e maioria mulheres. Das amostras biológicas, 70 tiveram resultados positivos para identificação bacteriana. O aspirado traqueal apresentou maior positividade a microrganismos, com 30%. Os microrganismos mais frequentes foram Escherichia coli, Staphylococcus coagulase negativa, Pseudomonas, Staphylococcus aureus, Enterobacter sp. e Proteus sp.

Conclusão: o estudo contribui para a assistência hospitalar, por produzir conhecimento do perfil microbiológico e sua resistência a antibióticos.

DESCRITORES: Perfil Epidemiológico; Microbiota; Infecção Hospitalar; Infecções Bacterianas; Resistência Microbiana a Medicamentos.

PERFIL DE MICROORGANISMOS AISLADOS DE PACIENTES INTERNADOS EN UN HOSPITAL DE PARANÁ

RESUMEN:

Objetivo: identificar el perfil de microorganismos aislados de muestras biológicas de pacientes internados en un hospital de Paraná.

Método: investigación descriptiva e retrospectiva, con recolección de datos de una fuente secundaria, desarrollada en un hospital de Paraná (Brasil), con referencia al año 2018. La población del estudio estuvo compuesta por muestras biológicas de 226 pacientes internados durante el período del 1 de enero al 31 de diciembre de 2018. El análisis de los datos se realizó por medio de estadística descriptiva.

Resultado: la media de edad de los pacientes fue de 60,41 años y la mayoría eran mujeres. De las muestras biológicas, 70 tuvieron resultados positivos con respecto a la identificación bacteriana. El aspirado traqueal presentó mayor positividad a microorganismos, con 30%. Los microorganismos más frecuentes fueron Escherichia coli, Staphylococcus coagulasa negativa, Pseudomonas, Staphylococcus aureus, Enterobacter sp. y Proteus sp.

Conclusión: el estudio contribuyó a la asistencia hospitalaria, por generar conocimiento del perfil microbiológico y su resistencia a antibióticos.

DESCRIPTORES: Perfil epidemiológico; Microbiota; Infección hospitalaria; Infecciones bacterianas; Resistencia microbiana a medicamentos.

Healthcare-Associated Infections (HAIs) represent a serious public health problem, as they generate social and financial impacts⁽¹⁾. They are characterized by infections related to health care provided in different settings, such as hospitals, long-term institutions, basic health units, clinics, offices, and home care⁽²⁾.

In the hospital context, these infections are acquired after the patient's admission and manifest themselves during hospitalization or after hospital discharge, related to hospitalization or health procedures. When the incubation period of the microorganism is not known, the National Health Surveillance Agency (*Agência Nacional de Vigilância Sanitária*, ANVISA) has agreed that Healthcare-Associated Infections (HAIs) are any clinical manifestation of infection that appears after 72 hours of the patient's admission⁽³⁾.

The HAIs terminology started to be used in 2007, with the publication of the Centers for Disease Control and Prevention (CDC) replacing the term Hospital Infection, due to the inability to determine for sure where the pathogen was acquired, as the patients are seen at different points in the care network, being exposed to several pathogens⁽²⁾.

In the Brazilian health services, there is a high incidence of HAIs, with a consequent increase in the inappropriate and indiscriminate use of different antimicrobials, resulting in the selection of resistant bacteria(4). In a reference university hospital for the northern region of Paraná, in the period from 2009 to 2011, of the 11,117 patients seen, 889 cases of HAIs were recorded, for an occurrence of 8%. Among the patients diagnosed with HAIs, the mortality rate was estimated at 38.4%, with the majority of deaths being related to these infections⁽⁵⁾.

Microbial resistance is considered a major threat to public health worldwide, as it increases the length of hospital stay, morbidity and mortality, in addition to reducing protection for surgical patients undergoing chemotherapy, and transplanted. Therapeutic options are increasingly scarce and expensive, and sometimes even non-existent^(4,6). The bacteria become resistant to antimicrobials due to four mechanisms: destruction or inactivation of the drug by enzymatic function, blocking the bacteria's porins in order to prevent the entry of the antimicrobial, changes in the drug's target site, and an efflux pump that eliminates the antimicrobial from the bacteria⁽⁷⁾.

A complication of the HAIs is sepsis, a host's unregulated response to infection, which leads to dysfunction of vital organs and is life-threatening, as it has a mortality rate greater than 50%. Another aspect to be highlighted is the severity of sepsis when acquired in the hospital compared to the acquisition of the pathogen in the community. A study carried out between 2010 and 2015 in a hospital of the southern region of Brazil, found a longer hospital stay (23 days versus 8 days) and twice the chance of hospital mortality (30.7% versus 15.6%) of patients with hospital sepsis in relation to community-acquired sepsis⁽⁸⁾.

To regulate the control and prevention measures for the HAIs, Ordinance No. 2,616, of May 12th, 1998, was drafted and published by the ANVISA, with guidelines and rules that establish the necessary actions for the prevention and control of HAIs, being instituted in public and private organizations that provide health care throughout the national territory⁽³⁾.

The Hospital Infection Control Program (*Programa de Controle de Infecções Hospitalares*, PCIH) is also included in this Ordinance and aims to reduce the incidence and severity of the HAIs, by means of a set of predetermined actions, guiding that "for the proper execution of the PCIH, the hospitals must constitute the Hospital Infection Control Commission (*Comissão de Controle de Infecção Hospitalar*, CCIH), the institution's advisory body and for the execution of hospital infection control actions"⁽³⁾.

For specialists in this area, conducting basic, epidemiological and translational research studies, to fill knowledge gaps that respond to emerging threats, represents one

of the four strategic pillars of actions to eliminate HAIs⁽⁹⁾.

The objective of the study was to identify the profile of the microorganisms isolated from biological samples of patients admitted to a hospital in northern Pioneer region of Paraná.

METHOD

This is a descriptive and retrospective research study. "Descriptive research aims to describe the distributions of the existing variables, without worrying about chance or other hypotheses"^(10:3).

The research was carried out in a Philanthropic Hospital, located in the northern Pioneer region of Paraná, which is part of the Regional Health Master Plan of the State of Paraná. The Hospital has Emergency Department, Maternity, Pediatrics, Medical-surgical Clinic, Surgical Center, and Intensive Care Unit (ICU) for adults, with 150 beds registered in the Unified Health System (*Sistema Único de Saúde*, SUS)⁽¹¹⁾.

The institution has a CCIH and uses the Online Hospital Infection Control Notification System (*Sistema Online de Notificação de Controle de Infecção Hospitalar*, SONIH). This system was implemented by the state of Paraná with the purpose of providing agility in the sending of hospital infection notification forms and data analysis by hospitals and by the Health Surveillance department⁽¹²⁾.

The study population consisted of patients admitted during the period from January 1st to December 31st, 2018. The variables were the following: number of patients hospitalized per month, hospitalization sector, gender, age, type and location of the culture material collected, analysis laboratory, morphology, degree of pathogenicity and Gram of the bacterium, and resistance of the microorganism to antimicrobials.

The source of data collection was secondary, using the SONIH, results of laboratory tests, and the CCIH record books. Data collection took place in August and September 2019 in the hospital itself. The data were entered into a spreadsheet, analyzed based on descriptive statistics and presented in absolute numbers and percentages.

With regard to the ethical aspects, the project was approved by the Ethics Committee for Research with Human Beings with opinion No. 3,438,955.

For this research, the use of the Free and Informed Consent Term was waived, as it is a retrospective descriptive study, which used only information from institutional information systems and/or other sources of data and clinical information available in the institution without the forecast of using biological material; because all data were handled and analyzed anonymously, without nominal identification of the research participants; because the results of the study were presented in an aggregated form, not allowing for the individual identification of the participants; and because it is a non-interventionist study (without clinical interventions) and with no changes/influences on the routine/treatment of the research participants and, consequently, without adding risks or losses to their wellbeing.

RESULTS

In 2018, according to data from DATASUS, 3,277 patients were admitted to the institution. 226 cultures of biological samples from patients were performed, 70 (30.97%)

with positive test results for bacterial identification. These positive tests came from biological samples from 53 patients, as some of them had positive results for bacterial identification in more than one site on the body.

The mean age of the participants was 60.41 years old, 32 (60.4%) were female and 21 (39.6%) were male. Figure 1 shows the distribution of the patients with a positive culture according to the hospitalization sector.

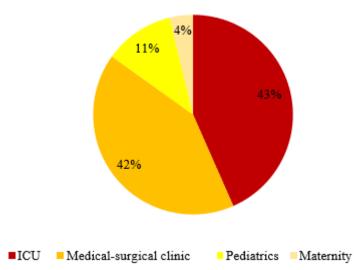


Figure 1 – Distribution of the patients (n=53) according to the hospitalization sector. Bandeirantes, PR, Brazil, 2019

The sectors of the ICU and medical-surgical clinic were those whose patients had the highest proportion of bacterial identification (Figure 1). The type of material with the highest number of microbial growth was tracheal aspirate with 30% (21/70), followed by urine with 28.7% (20/70) and by wound secretion with 27.1% (19/70).

The main bacteria isolated during 2018 are shown in Figure 2.

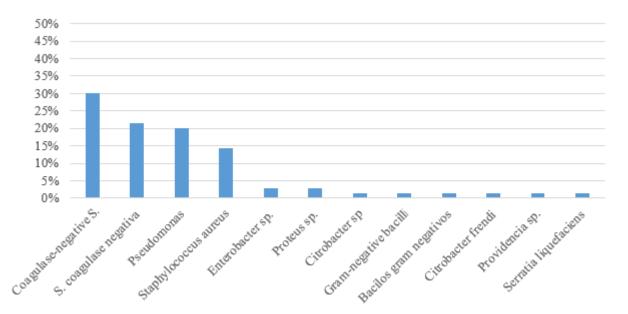


Figure 2 – Proportion of microorganisms identified (n=70) in samples from patients hospitalized in a hospital institution. Bandeirantes, PR, Brazil, 2019

The most frequent microorganisms were *E. coli* (n=21; 30.0%) coagulase-negative Staphylococcus (n=15; 21.4%), Pseudomonas (n=14; 20%), Staphylococcus aureus (n=10; 14.3%), Enterobacter sp. (n=2; 2.9%), and Proteus sp. (n=2; 2.9%). As for the classification by the Gram staining technique, it was observed that 26 (37.1%) are Gram-positive and 44 (62.9%), Gram-negative.

Figure 3 shows the association of the most frequent microorganisms according to the type of material collected for culture.

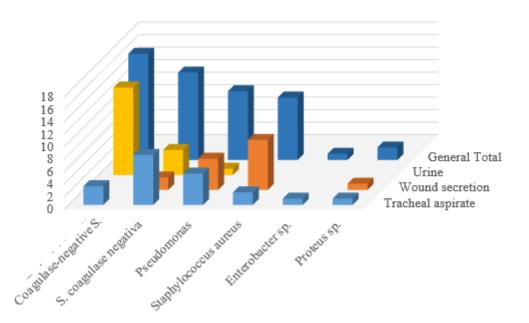


Figure 3 – Association of the material used for culture and the most frequent microorganisms of patients admitted to a hospital institution. Bandeirantes, PR, Brazil, 2019

There is a variety of bacteria isolated from material of the tracheal aspirate, with a greater number of coagulase-negative *Staphylococcus* (n=8; 40%), *Pseudomonas* (n=5; 25.0%), and *Escherichia coli* (n=3; 15.0%). In samples obtained from wound secretion, *Staphylococcus aureus* (n=8; 50%) had a higher incidence, followed by *Pseudomonas* (n=5; 31,3%). In turn, when analyzing urine, there was predominance of *Escherichia coli* (n=14; 73.7%). Table 1 shows the profile of bacterial resistance to the antibiotics tested.

Antibiotics		Antimicrobial resistance profile of the isolated bacteria (%)				
	E. coli	Pseudomonas	S. aureus	Coagulase- negative S.	Enterobacter sp.	Proteus sp.
	n=17	n=14	n=10	n=8	n=1	n=1
Nalidixic Acid	-	100	_	-	-	100
Pipemidic Acid	-	100	-	-	-	-
Amikacin	-	_	50	-	_	-
Amoxillin	0	100	50	0	100	-
Ampicillin	20	100	77.8	50	100	100
Azithromycin	-	66.7	-	-	_	-
Cephalothin	0	100	100	0	_	-
Cefepime	-	100	66.7	-	-	-
Cefotaxime	0	100	66.7	100	_	-
Cefoxitin	-	100	100	25	100	-
Ceftriaxone	100	100	100	100	_	100
Cefuroxime	30	100	100	50	100	-
Ciprofloxacin	23.5	53.8	100	50	_	100
Clindamycin	-	100	71.4	100	_	-
Chloramphenicol	-	100	66.7	_	_	-
Phosphomycin	100	16.7	50	100	100	-
Gentamicin	20	70	50	50	100	100
Imipenem	100	30	10	100	100	0
Levofloxacin	25	85.7	100	25	0	100
Lomefloxacin	100	66.7	100	100	0	-
Meropenem	-	50	0	-	_	-
Nitrofurantoin	0	100	80	0	_	100
Norfloxacin	23.5	25	100	0	_	100
Piperacillin	100	71.4	50	100	100	0
Sulf+Trimet	-	100	100	-	-	-
Sulfazotrim	-	_	100	-	-	100
Ticarcillin	0	50	100	0	-	-
Vancomycin	-	100	42.9	100	-	-

Table 1 - Antimicrobial resistance profile of the most frequently isolated bacteria in patients admitted to a hospital in northern Paraná. Bandeirantes, PR, Brazil, 2019

Profile of microorganisms isolated from inpatients in a hospital in Paraná

In the last three months of 2018, the hospital used the following antibiotics (injectables, tablets, and suspension): Amikacin, Amoxillin, Ampicillin, Ampicillin, + Sulbactam, Azithromycin, Sulfamethoxazole + Trimethoprim, Cephalexin, Cephalothin, Cephazolin, Cefepime, Ceftazidime, Ceftriaxone, Ciprofloxacin, Clindamycin, Amoxacillin +Clavulanate, Gentamicin, Levofloxacin, Meropenem, Metrodinazole, Norfloxacin, Oxacillin, Penicillin, Piperacillin + Tazobactam, and Vancomycin.

Among them, the most prescribed and administered antimicrobials in hospitalized patients were Ceftriaxone (n=2,188), Oxacillin (n=1,750), Clindamycin (n=1,018), Vancomycin (n=813), and Piperacillin + Tazobactam (n=651).

In the study, resistance of the *E. coli*, *Pseudomonas*, coagulase-negative *Staphylococcus*, *Staphylococcus aureus*, and *Proteus sp.* bacteria was observed for Ceftriaxone. Clindamycin and Vancomycin are antibiotics that showed sensitivity in few *S. aureus* bacteria, 71.4% and 42.9%, and without any sensitivity to *Pseudomonas* and coagulase-negative *S.*. Piperacillin + Tazobactam, the fifth most widely used antibiotic, was found to be effective for *Proteus sp.*, all being sensitive and showing 100% resistance in the *E. coli*, coagulase-negative *Staphylococcus*, and *Enterobacter sp.* bacteria.

DISCUSSION

The study carried out in a hospital in Goiânia-GO⁽¹³⁾ verified that 51.8% of the positive results for microbiological identification came from women and 48.2% from men, corroborating the results of this research. In contrast, another study⁽¹⁴⁾ demonstrated a higher incidence in males (56.8%).

The literature corroborates that the tracheal aspirate has higher incidence, representing 65.3% in one study⁽¹⁵⁾ and 45.9% in another⁽¹⁶⁾. However, other studies⁽¹³⁾ consider urine as the material with the highest microbial growth rate: urine culture showed 49.0% microbial growth and tracheal aspirate with only 11.8% and, in another⁽¹⁴⁾ study, microbial growth in the urinary tract was 40.90% in urine.

A study carried out in *Clínica Los Rosales*, in Colombia⁽¹⁷⁾, showed results similar to the microbiological profile of our study (Figure 2), as *E. coli* was present in 30.6% of the samples and *Staphylococcus aureus* in 9.7%. In addition, 74.19% (n=46) of the bacteria samples were classified as Gram-negative.

The most frequent site for the presence of microorganisms (Figure 3) was the tracheal aspirate with 36.4% and with a greater presence of coagulase-negative *Staphylococcus* and *Pseudomonas*, considering that they are mostly patients admitted to the ICU sector, who usually have higher susceptibility to bacterial infections due to invasive procedures, for example, mechanical ventilation through orotracheal intubation or tracheostomy, impaired immune system, indiscriminate use of antimicrobials for prophylaxis of infections, and presence of highly virulent microorganisms. Therefore, there is an increase in bacterial resistance and a consequent increase in the use of antimicrobials of new generations⁽¹⁸⁾.

According to the Anvisa⁽¹⁸⁾, *Pseudomonas* and *Enterobacter sp.* are the most common bacteria found in respiratory tract infections. In contrast, in the present study, coagulasenegative *Staphylococcus* was the most frequently isolated on this site. This microorganism is found on the skin, but it can also take shelter in the oropharynx and in the gastrointestinal and urogenital tracts. There are studies⁽¹⁹⁾ that report the opportunistic character of these bacteria, which can cause infections in the primary colonization sites or spread to other sites. Another hypothesis presented in the literature⁽¹⁹⁾ is the occurrence of colonization of health professionals by coagulase-negative *Staphylococcus* and possible transmission to the patients, still under study. *Pseudomonas* are bacilli distributed in large quantities in soils and in water sources, and can proliferate in any humid environment, capable of forming biofilm for their protection and with resistance to various disinfectants and antibiotics⁽⁷⁾.

In the microbiological culture of urine, the presence of microorganisms was more frequent in women, with *Escherichia coli* identified in most of the samples. This profile is in line with the literature, which points to this bacterium as the most frequent in urine samples. In the Hospital of Granada, Spain, analyses carried out from 2013 to 2016 showed that this bacterium represented the highest proportion among those isolated in the samples⁽²⁰⁾.

The *E. coli* bacterium is present in the microbiota of the human digestive tract. Due to the anatomical characteristics of women, who have a less extensive urethra compared to men, and in patients with a bladder catheter, this bacterium is the main responsible for urinary tract infections, especially for being transported by the ascending route (urethra, bladder, ureter, and kidney)⁽¹⁸⁾. It was not possible to analyze the association between the presence of the bacterium and the use of a bladder catheter due to the lack of this information in the researched data sources.

In the wound secretion material, there was predominance of the *Staphylococcus aureus* microorganism, which corroborates with a study carried out in 66 cultures of wound secretion of patients admitted to a Public Hospital⁽²¹⁾, as well as with the results of the research carried out in the National Wound Institute Foundation (*Fundación Instituto Nacional de Heridas*, FINH) in Chile⁽²²⁾. According to a study⁽⁷⁾, the two genera of bacteria that most often affect skin infections are *Staphylococcus* and *Streptococcus*.

The *Proteus sp.* bacterium was found in the current study both in the tracheal aspirate and in the wound secretion. It is common to find it in the digestive tract as part of the normal microbiota. In a hospital environment, the bacterium is a causative agent of infections in the urinary tract and in surgical wounds. Infections in surgical wounds have the patients themselves as source of bacteria, from other colonized sites such as the nostrils, oral cavity, female genital tract, feeding tract, and the skin. Still, the hospital environment itself and the medical and nursing teams also represent a potential source for infection⁽¹⁸⁾.

It is noteworthy that other factors influence on the pathogenesis of surgical wound infections, such as host-related risk factors (obesity, diabetes mellitus, vascular insufficiency, and immunodeficiencies), microbiological factors (microbial load and virulence of each germ), and factors related to the surgery (potential for contamination of the surgical site, duration of the surgery, and intra- and peri-operative complications)⁽¹⁸⁾.

In the present study, antimicrobial resistance by *E. coli* was found to Ampicillin and to Ceftriaxone in 20% and 100% of the sample, respectively. These findings were in disagreement with another study⁽¹⁷⁾, which identified 95% of the *E. coli* bacteria with resistance to Ampicillin and 56% to Ceftriaxone. In a survey⁽²³⁾, the data are also divergent, as it presents 20% of the *E. coli* bacteria resistant to Ceftriaxone and 100% sensitivity to Piperacillin + Tazobactam, but corroborates the percentage of 20% resistance to Ciprofloxacin and Gentamicin.

According to a study⁽²⁴⁾, conducted by means of electronic reports of patients admitted to six private hospitals in Cascavel-PR, also show different results from the present study, with 28% of the *E. coli* bacteria showing resistance to Ceftriaxone, 7% to Imipenem, and 10% to Piperacillin. As for the resistance to Gentamicin, the result is similar, with 21% of resistant bacteria.

The resistance results of the *Pseudomonas* bacterium corroborates with a research study⁽¹⁶⁾, showing that 100% of the *Pseudomonas* shows resistance to Ampicillin, Cefotaxime, and Nitrofurantoin. It also shows that some bacteria are resistant to Ciprofloxacin (57.1%), Cefepime (42.8%), Imipenem (53.3%), Meropenem (46.6%), and Piperacillin + Tazobactam (66.6%).

According to a study⁽²³⁾, the resistance results of the *Staphylococcus aureus* bacterium

in relation to the antibiotics, show disagreement for Amikacin, Ceftriaxone, Gentamicin, and Sulfametoxazole + Trimethoprine, all without resistance, Ciprofloxacin (14.3%), and with a close result in Clindamycin (42.9%). Another study also showed disagreement for Ciprofloxacin, with 28.5% resistance, as well as for Vancomycin, with 100% sensitivity⁽¹⁶⁾.

According to a research study⁽¹⁶⁾, the resistance profile of the coagulase-negative *Staphylococcus* bacterium for Ciprofloxacin and Gentamicin presents values similar to the present study, 54.5% and 58.3%, respectively. Clindamycin and Vancomycin show 58.3% and 0.0% resistance, presenting contradictory values.

The study has methodological limitations that must be considered for the interpretation of the results. The sample was heterogeneous, composed of patients in intensive care units and inpatient clinics. The collection of data from secondary sources, with a retrospective view, limited the exploration of other variables associated with antimicrobial resistance. The culture and antibiogram of biological samples in the institution under study is not routine for all the patients who used antibiotics. Therefore, the percentage of antimicrobial resistance presented was calculated based on the available tests, and not on the total number of patients who used each type of antibiotic, a fact that can overestimate the percentage of bacterial resistance to the antibiotics presented.

CONCLUSION

The microbiological profile of the patients admitted to the hospital in northern Paraná showed a diversity of bacteria and resistance to several antibiotics, with a higher frequency of *E. coli*, coagulase-negative *Staphylococcus*, *Pseudomonas*, *Staphylococcus aureus*, *Enterobacter sp.* and *Proteus sp.*, with predominance of Gram-negative bacteria.

The antibiotic resistance found was worrying, as it was observed that, with the exception of *Proteus sp.*, the five most frequent bacteria showed 100% resistance to Ceftriaxone, the most prescribed antibiotic for treating bacterial infections in the hospital.

The present study contributes to hospital care, especially for the study locus, as it produced knowledge about the profile of the most frequent microorganisms in the patients treated there and an approximation to the antibiotic resistance profile.

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Corresponding author: Ricardo Castanho Moreira Universidade Estadual do Norte do Paraná Rod. BR 369, Km 54 - 86.360-000 - Bandeirantes, PR, Brasil E-mail: ricardocastanho@uenp.edu.br

Role of Authors:

Substantial contributions to the conception or design of the work; or the acquisition, analysis, or interpretation of data for the work - RCM, LSM, ELF

Drafting the work or revising it critically for important intellectual content - RCM, LSM, ELF, CQH Final approval of the version to be published - RCM, LSM, ELF, CQH

Agreement to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved - ARES



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