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Phenotypic identification of bacteria of the family *Enterobacteriaceae* with resistance profile on inanimate surfaces in a University Hospital

Identificação fenotípica de bactérias da família *Enterobacteriaceae* com perfil de resistência em superfícies inanimadas em um Hospital Universitário

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Abstract

The aim of this study was to identify bacteria belonging to the family *Enterobacteriaceae* with resistance profile on inanimate surfaces and utensils of sectors from a University Hospital of Pernambuco. Initially, the samples were collected on 25 inanimate surfaces and utensils of the Intensive Care Unit (ICU), Hemodialysis Center and Medical Clinic in 2018. The samples were plated on MacConkey and SS agar and incubated at 35 ± 2 °C for 24 hours. After incubation, they were submitted to biochemical tests to confirm the presence of bacteria to the family *Enterobacteriaceae*. Subsequently, *Klebsiella pneumoniae* carbapenemase (KPC) was identified by the disc diffusion method, as well as by Hodge test; extended-spectrum beta-lactamase producing bacteria (ESBL) by the disk diffusion and double disk diffusion tests; and metallo-beta-lactamase producing bacteria (MBL) through the disc diffusion method according to the Clinical and Laboratory Standards Institute (CLSI). Eight surfaces or utensils of the 18 samples with *Enterobacteriaceae* presented resistance profile (44.4%). KPC was identified in 46.7% of the samples that presented bacteria with resistance profile, one presented simultaneously KPC and ESBL (6.65%), another one KPC and MBL concomitantly (6.65%) and 6 samples presented only ESBL (40%). Medical Clinic and Hemodialysis Center had the highest presence of resistant *Enterobacteriaceae*. The detection of resistant microorganisms in hospitals is worrying and emphasizes the importance of health professionals in acquiring knowledge about preventive measures to minimize hospital infections.

Keywords: *Klebsiella pneumoniae* carbapenemase (KPC); Extended-spectrum beta-lactamase producing bacteria (ESBL); Metallo-beta-lactamase producing bacteria (MBL); Inanimate surfaces; Hospital infection.

Resumo
O objetivo deste estudo foi identificar bactérias pertencentes a família *Enterobacteriaceae* com perfil de resistência em superfícies inanimadas e utensílios de setores de um Hospital Universitário de Pernambuco. Inicialmente, as amostras foram coletadas em 25 superfícies e utensílios inanimados da Unidade de Tratamento Intensivo (UTI), Centro de Hemodiálise e Clínica Médica em 2018. As amostras foram laminadas em ágar MacConkey e SS e incubadas a 35 ± 2 °C por 24 horas. Após a incubação, elas foram submetidas a testes bioquímicos para confirmar a presença de bactérias da família *Enterobacteriaceae*. Posteriormente, *Klebsiella pneumoniae* carbapenemase (KPC) foi identificada pelo método de difusão em disco, assim como pelo teste de Hodge; bactérias produtoras de beta-lactamase de espectro estendido (ESBL) pelos testes de difusão em disco e difusão em disco duplo; e bactérias produtoras de metallo-beta-lactamase (MBL) pelo método de difusão em disco de acordo com o *Clinical and Laboratory Standards Institute* (CLSI). Oito superfícies ou utensílios das 18 amostras com *Enterobacteriaceae* apresentaram perfil de resistência (44,4%). O KPC foi identificado em 46,7% das amostras que apresentaram perfil de resistência das bactérias, uma apresentou simultaneamente KPC e ESBL (6,65%), outra KPC e MBL concomitantemente (6,65%) e 6 amostras apresentaram apenas ESBL (40%). Clínica Médica e Centro de Hemodiálise tiveram a mais alta presença de *Enterobacteriaceae* resistentes. A detecção de microorganismos resistentes em hospitais é preocupante e enfatiza a importância dos profissionais de saúde na aquisição de conhecimentos sobre medidas preventivas para minimizar as infecções hospitalares.
Palavras-chave: *Klebsiella pneumoniae* carbapenemase (KPC); Bactérias produtoras de beta-lactamase de espectro estendido (ESBL); Bactérias produtoras de metallo-beta-lactamase (MBL); Superfícies inanimadas, Infecção hospitalar.

Resumen
El objetivo de este estudio fue identificar las bacterias pertenecientes a la familia *Enterobacteriaceae* con perfil de resistencia en superficies inanimadas y utensilios de sectores de un Hospital Universitario de Pernambuco. Inicialmente, las muestras fueron recogidas en 25 superficies inanimadas y utensilios de la Unidad de Cuidados Intensivos (UCI), Centro de Hemodiálisis y Clínica Médica en 2018. Las muestras fueron sembradas en agar MacConkey y SS e incubadas a 35 ± 2 °C durante 24 horas. Tras la incubación, se sometieron a pruebas bioquímicas para confirmar la presencia de bacterias de la familia *Enterobacteriaceae*. Posteriormente, se identificó la *Klebsiella pneumoniae* carbapenemasa (KPC) mediante el método de difusión en disco, así como mediante la prueba de Hodge; las bacterias productoras de betalactamasas de espectro extendido (BLEE) mediante las pruebas de difusión en disco y de doble difusión en disco; y las bacterias productoras de metalobactamasas (MBL) mediante el método de difusión en disco según el *Clinical and Laboratory Standards Institute* (CLSI). Ocho superficies o utensilios de las 18 muestras con *Enterobacteriaceae* presentaron perfil de resistencia (44,4%). Se identificó KPC en el 46,7% de las muestras que presentaban bacterias con perfil de resistencia, una presentaba simultáneamente KPC y ESBL (6,65%), otra KPC y MBL de forma concomitante (6,65%) y 6 muestras presentaban sólo ESBL (40%). La Clínica Médica y el Centro de Hemodiálisis tuvieron la mayor presencia de *Enterobacteriaceae* resistentes. La detección de microorganismos resistentes en los hospitales es preocupante y pone de manifiesto la importancia de que los profesionales sanitarios adquieran conocimientos sobre las medidas preventivas para minimizar las infecciones hospitalarias.
Palabras clave: *Klebsiella pneumoniae* carbapenemasa (KPC); Bacterias productoras de betalactamasas de espectro extendido (ESBL); Bacterias productoras de metalobactamasas (MBL); Superficies inanimadas; Infección hospitalaria.

1. Introduction

Bacterial resistance has been rising worldwide and it has become an increasingly worrying problem. This resistance can occur in antimicrobials that are used as first choice in the treatment of severe infections antimicrobials. One of the main causes of recurrence of infections is the ability of microorganisms to synthesize enzymes responsible for their resistance (Laxminarayan, et al., 2016; Banin, et al., 2017).

The bacteria belonging to the family *Enterobacteriaceae* are pathogens capable of causing different types of hospital-acquired infections, a situation that is aggravated by the bacterial resistance in this family, resulting in a challenging therapeutic plan (Rodríguez-Baño, et al., 2018).

Microorganisms that produce these enzymes, such as *Klebsiella pneumoniae* carbapenemase (KPC), extended-spectrum beta-lactamase producing bacteria (ESBL) and metallo-beta-lactamase producing bacteria (MBL) can degrade antimicrobials from the beta-lactam group, but these drugs are most often used in the treatment of severe infections (Bush, 2015).

The presence of resistant microorganisms in hospital environments has caused great impact both in the clinical aspect, due to the high mortality rates associated with the infections caused by these microorganisms (Geisinger, et al., 2017), and in the economic aspect, due to the increase of costs with drug therapy and with time hospitalization (Bush, et al., 2015; Falcone, et al., 2018).

In general, the patients most affected by infection caused by resistant microorganisms are those hospitalized in the Intensive Care Units (ICUs), in addition to the neonates, the diabetics and the immunocompromised ones (Mutters, et al., 2018). These infections are related to the use of critical utensils, such as surgical instruments, in health care that are in constant contact with patients or health care professionals (Johani, et al., 2018; Mutters, et al., 2018).

The first cases of hospital infections induced by KPC were described in 2000 in the United States and rapidly it was spread with outbreaks in Israel and Greece, as well as in large geographical areas such as South America and China (Chen, et al., 2011). In Brazil, the first cases of KPC were reported in Recife and Rio de Janeiro between 2006 and 2009 (Monteiro, et al., 2009; Peirano, et al., 2009). The high and rapid spread of KPC, ESBL and MBL was possible by the ability of the bacteria to transfer their genetic material and the resistance genes, representing a serious risk for the patient and the health professional (Blair, et al., 2015).

Inanimate surfaces and utensils are a constant source of microbial contamination (Bassetti, et al., 2018), because the exposure of patients to these materials is one of the factors responsible for the occurrence of infections associated with health care. Thus, the sterilization and disinfection of these environments, the correct sanitation of health professionals, as well as the diagnosis and correct treatment of infections are measures that play a fundamental role in the prevention of the emergence of infections by resistant microorganisms (Khan, et al., 2017; Bassetti, et al., 2018).

In this context, the aim of this study was to identify bacteria belonging to the family *Enterobacteriaceae* with resistance profile on inanimate surfaces and utensils of the Clinical Medicine, Hemodialysis, and Intensive Care Unit sectors used in the care of patients at a University Hospital in Pernambuco.

2. Methodology

2.1 Materials

The culture media used in this study were obtained from Himedia® and Kasvi® and all solvents were obtained from Merck®. Antimicrobial agents were supplied by Laborclin®. The bacteria were stored at -80 °C in brain heart infusion broth (BHIB) containing 20% glycerol as a cryoprotectant in the Keizo Asami Immunopathology Laboratory from the Federal University of Pernambuco (LIKA/UFPE).

2.2 Collection of samples

Initially, between February and March 2018, 8 samples of inanimate surfaces and utensils from the from the Medical Clinic (sphygmomanometer for common use, transport box for physiological saline, defibrillator trolley (paddles and knob), table of the nursing station, stethoscope for common use, phone and saline package), 10 samples from the Hemodialysis Center (sink for preparation of medication, handwhasing sink, phone, defibrillator trolley (paddles and knob), table of the nursing station, hemodialysis machine buttons (left and right wing), ECG machine (clip and suction cups) and 7 sample of ICU (Electrocardiogram (ECG) achine button, defibrillator trolley (paddles and knob), handwashing sink, phone, multiparameter monitors buttons and sink for preparation of medication), of a public hospital in the city of Recife – PE, Brazil, were collected with a swab, then transferred to BHIB and stored at LIKA/UFPE.

2.3 Sample Analysis

2.3.1 Identification of Enterobacteriaceae

For the identification of bacteria belonging to the family *Enterobacteriaceae*, the samples of the BHI were incubated at 35 ± 2 °C for 24 h. After microbial growth, the samples were plated in MacConkey and SS agar and incubated again at 35 ± 2 ºC for 24 h. After the incubation, the colonies were submitted to the classic biochemical tests to confirm the presence of this bacterial group.

2.3.2 Identification of antimicrobial resistance profile of bacteria

The identification of KPC, ESBL and MBL was performed according to the *Clinical and Laboratory Standards Institute* (CLSI, 2016). After the identification of the *Enterobacteriaceae*, KPC was identified by the disc diffusion method with antimicrobials from the carbapenemics group, as well as by the modified Hodge test. For the disc diffusion method, inoculums of the microorganisms adjusted to 0.5 of the McFarland scale were prepared and seeded in Müeller Hinton agar. Subsequently, the carbapenem disks (ertapenem, imipenem and meropenem) were deposited on the plates and incubated at 35 ± 2 °C for 18 h. Samples were analyzed through the inhibition halos following the CLSI cut-off points. For the modified Hodge test, initially, *E. coli* ATCC 25922 was seeded on Müeller Hinton agar and the meropenem disk was deposited in the center of the plate. Then, the test bacteria were inoculated straight from the edge of the disc to the periphery of the plate. Finally, the plates were incubated at 35 ± 2 °C for 18 h and the result was based on the distortion of the inhibition halos. The positive quality control was performed using a carbapenemase-positive bacterial clinical strain and the negative one was *E. coli* ATCC 25922.

To identify ESBL, the methods used were disk diffusion with antimicrobial and double disk diffusion tests. In both tests the inoculums were adjusted to 0.5 of the McFarland scale and seeded on Müeller Hinton agar. The disc diffusion test was performed as described above only replacing the carbapenems by ceftazidime, cefotaxime and cefpodoxime. For the double disk diffusion test, it was used the discs of ceftazidime (CAZ), amoxicillin with clavulanic acid (AMX/AC) and aztreonam (ATM) that were distributed at 25 mm between them. Finally, the plates were incubated at 35 ± 2 °C for 18 h. After the incubation, the risinge of an additional inhibition halo between AMX/AC and CAZ and/or ATM (ghost-zone) was considered positive for ESBL production (CLSI, 2014). The positive quality control was performed using *K. pneumoniae* ATCC 700603 and the negative one was *E. coli* ATCC 25922.

To identify MBL, the antimicrobial disc diffusion method associated with a chelating agent was used. Over again, the inoculum was adjusted to 0.5 of the McFarland scale, seeded on Müeller Hinton agar and the imipenem, EDTA and ceftazidime discs were deposited at 10 mm between them. The plates were incubated at 35 ± 2 °C for 18 h and the change in inhibition halos confirmed the presence of MBL. The positive quality control was performed using a MBL-positive bacterial clinical strain and the negative one was *E. coli* ATCC 25922.

3. Results

The results were described after collecting 25 samples and identifying 18 surfaces containing *Enterobacteriaceae* bacteria, which resulted in 33 strains (Table 1). At the Medical Clinic, 87.5% of the surfaces (sphygmomanometers commonly used, stethoscopes in common use, table of the nursing station, transport box for physiological saline, defibrillator trolley (paddles) and phone) presented *Enterobacteriaceae*, among them: *Shigella* spp., *Escherichia coli*, *Enterobacter* spp., *Salmonella* spp., *Klebsiella pneumoniae* and *Providencia* spp. The most found bacterium was *Escherichia coli* (37.5%). In this sector only the packaging of saline did not present of bacteria belonging to the family *Enterobacteriaceae*. While 60% of the surfaces of the Hemodialysis Center presented *Enterobacteriaceae* (sink for preparation of medication, handwashing sink, phone, hemodialysis machine buttons (left wing), defibrillator trolley (knob) and nursing station table), among them: *Enterobacter* spp., *Salmonella* spp., *Providencia* spp., *Citrobacter* spp., *Serratia* spp., *Shigella* spp., *Klebsiella pneumoniae* and *Escherichia coli*. The most prevalent bacteria were *Escherichia coli* (27.7%) and *Enterobacter* spp. (22.2%). The ECG machine, defibrillator trolley (paddles) and the hemodialysis machine button (right wing) of this center did not present *Enterobacteriaceae*.

The ICU presented 71.4% of the surfaces with *Enterobacteriaceae* (ECG machine buttons, defibrillator trolley (knobs and paddles), phone and handswashing sink), among them: *Enterobacter* spp. (8.3%), *Serratia* spp. (8.3%), *Providencia* spp. (8.3%), *Klebsiella pneumoniae* (8.3%), *Shigella* spp. (33.3%) and *Escherichia coli* (33.3%). From the samples collected in this sector, only the buttons of the multi-parameter monitors and the sink for preparation of medication did not present *Enterobacteriaceae*.

Escherichia coli was the most prevalent bacterium among the strains, presenting a percentage of 32.5%, followed by *Enterobacter* spp., *Shigella* spp., *Klebsiella pneumoniae*, *Salmonella* spp., *Serratia* spp., *Providencia* spp. and *Citrobacter* spp., with prevalence of 17.4%, 15.2%, 13%, 8.8%, 6.5%, 4.4% and 2.2%, respectively.

Table 1: *Enterobacteriaceae* identified in the Hemodialysis Center, Medical Clinic, and ICU from a University Hospital of Pernambuco.

Sector	Prevalence of colonization n/N total (%)	Place/object	Bacterial Genus/Species	Genus/species prevalence* n/N total (%)
Medical Clinic	7/8 (87.5)	Sphygmomanometer for common use	*Shigella* spp.	2/16 (12.5)
			Enterobacter spp.	3/16 (18.75)
			Salmonella spp.	2/16 (12.5)
			Escherichia coli	6/16 (37.5)
		Transport box for physiological saline	*Klebsiella pneumoniae*	3/16 (18.75)
			Enterobacter spp.	
		Defibrillator trolley (paddles)	*Klebsiella pneumoniae*	
			Escherichia coli	
		Table of the nursing station	*Klebsiella pneumoniae*	
			Escherichia coli	
		Defibrillator trolley (knob)	*Enterobacter* spp.	
			Escherichia coli	
		Stethoscope for common use	*Salmonella* spp.	
			Escherichia coli	
			Shigella spp.	
		Phone	*Escherichia coli*	
		Saline package	Absent	
Hemodialysis	6/10 (60)	Sink for preparation of medication	*Enterobacter* spp.	4/18 (22.2)
			Salmonella spp.	2/18 (11.1)
		Handwhasing sink	*Providencia* spp.	1/18 (5.5)
			Salmonella spp.	
			Citrobacter spp.	1/18 (5.5)
			Escherichia coli	5/18 (27.7)
		Phone	*Enterobacter* spp.	
			Serratia spp.	2/18 (11.1)
			Shigella spp.	1/18 (5.5)
			Escherichia coli	
		Defibrillator trolley (knob)	*Enterobacter* spp.	
			Escherichia coli	
			Shigella spp.	
			Escherichia coli	
		Table of the nursing station	*Klebsiella pneumoniae*	
			Escherichia coli	2/18 (11.1)
		Hemodialysis machine buttons (left wing)	*Enterobacter* spp.	
			Klebsiella pneumoniae	
		ECG machine (clip)	Absent	
		ECG machine (suction cups)	Absent	
		Defibrillator trolley (paddles)	Absent	
		Hemodialysis machine buttons (right wing)	Absent	
ICU	5/7 (71.4)	ECG machine button	*Serratia* spp.	1/12 (8.3)
			Shigella spp.	4/12 (33.3)
		Defibrillator trolley (knob)	*Enterobacter* spp.	1/12 (8.3)
			Shigella spp.	
			Escherichia coli	4/12 (33.3)
		Defibrillator trolley (paddles)	*Shigella* spp.	
			Escherichia coli	
		Handwashing sink	*Shigella* spp.	
			Escherichia coli	
		Phone	*Providencia* spp.	1/12 (8.3)
			Klebsiella pneumoniae	1/12 (8.3)
			Escherichia coli	
		Multiparameter monitors buttons	Absent	
		Sink for preparation of medication	Absent	
TOTAL	18/25 (72.0)	-	-	-

* Prevalence of bacterial species/genus per identified strain. Source: Authors.

In addition to the presence of enterobacteria, it was also possible to evaluate their resistance profile (Table 2). Eight surfaces or utensils of the 18 samples with *Enterobacteriaceae* presented resistance profile (44.4%). KPC was identified in 46.7% of the samples that presented bacteria with resistance profile, one presented simultaneously KPC and ESBL (6.65%), another one KPC and MBL concomitantly (6.65%) and 6 samples presented only ESBL (40%). In the present study ESBL was detected in 3 genera of distinct *Enterobacteriaceae* (*Klebsiella pneumoniae*, *Escherichia coli* and *Enterobacter* spp.). The sectors with the highest incidence of bacteria with resistance profile were the Hemodialysis Center and the Medical Clinic.

Table 2: *Enterobacteriaceae* with resistance profile identified in the Hemodialysis Center, Medical Clinic and ICU from a University Hospital of Pernambuco.

Sector	Prevalence of colonization n/N total (%)	Place/object	Bacterial Genus/Species with resistance profile	Prevalence of resistance profile
Medical Clinic	2/7 (28.5%)	Defibrillator trolley (knob)	*Enterobacter* spp. (ESBL and KPC)	1/5 (20%)
			Escherichia coli (KPC)	2/5 (40%)
		Table of the nursing station	*Klebsiella pneumoniae* (KPC and MBL)	1/5 (20%)
			Providencia spp. (KPC)	1/5 (20%)
			Escherichia coli (ESBL)	
		Phone	Absent	
		Transport box for physiological saline	Absent	
		Sphygmomanometer for common use	Absent	
		Stethoscope for common use	Absent	
		Defibrillator trolley (paddles)	Absent	
Hemodialysis	4/6 (66.7%)	Phone	*Serratia* spp. (KPC)	4/8 (50%)
			Escherichia coli (ESBL)	4/8 (50%)
		Defibrillator trolley (knob)	*Enterobacter* spp. (KPC)	
			Escherichia coli (ESBL)	
		Table of the nursing station	*Klebsiella pneumoniae* (ESBL)	
			Escherichia coli (KPC)	
		Hemodialysis machine buttons (left wing)	*Klebsiella pneumoniae* (KPC)	
			Escherichia coli (ESBL)	
		Handwhashing sink	Absent	
		Sink for preparation of medication	Absent	
ICU	2/5 (40%)	ECG machine buttons	*Shigella* spp. (KPC)	1/2 (50%)
		Defibrillator trolley (knob)	*Escherichia coli* (ESBL)	1/2 (50%)
		Defibrillator trolley (paddles)	Absent	
		Handwhashing sink	Absent	
		Phone	Absent	
TOTAL	8/18 (44.4%)	-	-	-

KPC: *Klebsiella pneumoniae* carbapenemase; ESBL: extended-spectrum beta-lactamase producing bacteria; MBL: metallo-beta-lactamase producing bacteria. Source: Authors.

4. Discussion

The results found in the present study are relevant since high indices of *Enterobacteriaceae* with and without resistance were found. This fact corroborates the predominance of Gram-negative bacteria, such as *Klebsiella pneumoniae* and *Escherichia coli*, as responsible for cases of nosocomial infection, especially in patients submitted to long periods of hospitalization (El Chakhtoura, et al., 2018).

The choice of sites for collection was established according to the potential risk of infections in emergency routines, by selecting surfaces and objects of excessive use that are closer to the patients or that are commonly touched by professionals (Puzniak, et al., 2019; Smibert, et al., 2018). These nosocomial infections, mainly in patients hospitalized for a prolonged period and/or undergoing invasive procedures (Debnath, et al., 2018), usually occur due to the great manipulation of the equipment and the objects by the professionals and patients, providing an increased risk for the occurrence of cross-infection (Johani, et al., 2018; Monteiro, et al., 2009). In fact, most of the resistant microorganisms in our study were found in places for the exclusive use of professionals involved in procedures.

The identification of Gram-negative bacteria in the hospital environment emphasizes the importance of conducting studies that verify the presence of these bacteria in these places. Michael, David and Jhon (2014) also identified *Enterobacteriaceae* of clinical importance on surfaces present in departments of the Hospital of Brooklyn (USA) and they report the influence that these bacteria have in the health-disease process.

In the present study, almost the half of the analyzed samples presented KPC. This result is worrying, since the presence of microorganisms with resistance profile in the hospital setting can be characterized as a serious risk to the health of patients undergoing any type of care or procedures, especially invasive procedures (Lago, et al., 2010). However, this fact has become common. In Brazil, Pinto, et al. (2014). observed the predominance of KPC in clinical isolates of biological material in patients from four tertiary hospitals in Porto Alegre where this microorganism was present in 48.5% of the cases. KPC was also described by Renner, et al. (2013) in the respirator of one of the beds in a hospital of Rio Pardo-RS Valley and this equipament is used by patients, most of whom are severe and hospitalized in the ICU. KPC has great clinical influence since it may be responsible for cases of serious hospital infections with possible complications (Pinto, et al., 2014).

Rios, et al. (2020) isolated and identified 87 strains of bacteria, including *Staphylococcus aureus*, *Staphylococcus* spp. coagulase negative, *S. saprophyticus*, *Citrobacter freundii*, *Acinetobacter baumannii*, *Klebsiella* spp. in clothing worn by professionals (pockets, wrists, and waist region) at the Municipal Hospital of Teixeira de Freitas (MHTF) and the Emergency Care Unit (ECU), in the state of Bahia. All strains were resistant to one or more antimicrobials, such as nalidixic acid, meropenem, aztreonam and levofloxacina.

A cross-sectional research conducted in 2015 at a medium-sized hospital in the State of Rio Grande do Sul collected surface samples from the trays used for the administration of medicines. Among the 36 trays investigated, 15 (41.6%) showed growth of microorganisms such as *Staphylococcus* spp. coagulase negative, *Acinetobacter baumanni*, *Enterobacter agglomerans*, *Klebsiella oxytoca*, *Klebsiella ozaenae*, *Staphylococcus aureus*, *Acinetobacter lwoffii*, *Pseudomonas stutzeri* and *Pseudomonas aeruginosa*, such results corroborate with the findings described in the present study and point out the need for constant elaboration of care strategies in clinical practice in order to reduce the expressive presence of these microorganisms (de Freitas, et al., 2019).

A study conducted in 2018 at the university hospital of the Federal University of Uberlândia, MG, determined the presence of Gram-negative bacteria isolated on hospital surfaces of a Neonatal Intensive Care Unit, analyzing the resistance profile, the production of β-Lactamases of Extended Spectrum (ESBL) and AmpC, a gene responsable to the resistance to penicillins, clavulanic acid and cephalosporins. Among the 408 samples collected, 30 were positive for Gram-negative bacteria, 19 of which were multidrug resistant, 10 were ESBL producers and 19 were AmpC) producers (Marques, et al., 2019).

The second most prevalent resistance profile in this study was ESBL. These bacteria have been increasingly described in the literature, highlighting their participation in urinary infections, meningitis, septicemia, pneumonia, and bacteremia. ESBL was also present in the study conducted by Lago et al (2010). who observed that 24.8% of patients hospitalized at Hospital São Vicente de Paulo, Passo Fundo, RS, were contaminaned by these bacteria. According to Silva and Lincopan (2012), ESBLs have become the main public health problem in nosocomial infections related to the *Enterobacteriaceae* and they are currently widely disseminated worldwide, being found in the countries, such as France, Brazil, Greece, United Kingdom, the Netherlands, Hungary, Portugal, Bolivia, and others.

The resistance profile with lower prevalence was MBL. Meyer and Picoli (2011) analyzed isolates of *K. pneumoniae* given by an emergency hospital in Porto Alegre and observed only 3.4% of MBL positive samples. The same authors also affirm that the first reported case of metallo-beta-lactamases in Brazil occurred only in 2005 and with the simultaneous presence of ESBL. It is worth mentioning that the detection of this type of microorganism is important, since the treatment is complicated and may be associated with an unfavorable end to the patient.

According to the World Health Organization (WHO) patient safety is defined as the absence of damage throughout the care process. In this way, it is directly related to effective care and this topic has been increasingly frequent between studies (Bela-Anacleto, et al., 2013). In this context, the detection of these microorganisms with resistance profile is essential, since the problem of multiresistance, especially with KPC, has been highlighted as a higher risk for immunodepressed patients, such as debilitated and hospitalized patients in ICU (Puzniak, et al., 2019; Logan, et al., 2017). This identification has also a real importance in the prophylactic practices against the occurrence of infections, especially in university hospitals, since in these places there is a higher incidence of infections compared to other hospitals. This fact can be explained due to the high number of complex procedures in university hospitals, besides the long period of hospitalization and the existence of an effective interaction of patients with several health professionals, including academics and other students (Agarwal, et al., 2018). The identification of certain bacteria groups, as in the present research, is also essential for the therapeutic process, since the ability of these bacteria to resist to antibacterial action is high due to their possibility to adquire mechanisms of resistance that can be transfer between microorganisms (Taylor-Robinson, 2019).

It is important to highlight that patients undergoing different procedures are more vulnerable to infections, in this way any surface that can come into direct or indirect contact with patients must undergo sterilization or disinfection procedures correctly and appropriately (Rutala, et al., 2019). The presence of resistant bacteria in hospitals represents a serious risk to the health and recovery of patients those require care at these locals, it is necessary that health care professionals be aware of the possible sources of contamination in this environment to establish infection control measures in order to decrease these infections and improve patient survival time.

5. Conclusion

Lastly, in this study *Enterobacteriaceae* with resistance profile were identified, especially in the hemodialysis center and the medical clinic, and the main places contaminated were restricted to hospital professionals. Professionals responsible for attendance inside the hospital environment need to practice infection control measures through the organization of the physical space and equipments as well as through the guidance on cleaning and hand hygiene for visitors, family, and other professionals, because the direct and indirect contact of these groups with the patients by objects and equipments makes possible the microorganism transmission in cases where correct measures of prevention are not taken.

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