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Identification of bacteria in ambulances: healthcare risks to patient safety

Identificación de bacterias en ambulancias: riesgos sanitarios para la seguridad del paciente

Identificação de bactérias em ambulâncias: riscos assistenciais à segurança do paciente

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Abstract

The aim was to identify microorganisms through the microbial cultivation of bacteria inside ambulances in two municipalities in Baixada Fluminense, in the State of Rio de Janeiro, and to test the sensitivity of these microorganisms to antimicrobials. Samples were obtained from nine specific points inside 6 ambulances. They were inoculated onto salt and blood mannitol agar and were analyzed using matrix-assisted laser desorption and ionization techniques. The sensitivity of seven samples was tested using the disk diffusion method on Muller-Hinton agar. Of the fifty-two samples collected, thirty-one (59.6%) were positive for some genus of bacteria, two (3.80%) were not recognized by MALDI and nineteen (36.6%) were negative. Infection prevention and control practices need to be reinforced, as contamination can result in possible complications for patients.

Descriptors: Emergency Medical Services; Bacteria; Infection Control; Pre-Hospital Emergency Care; Disinfection.

Resumén

El objetivo fue identificar microorganismos a través del cultivo microbiano de bacterias dentro de ambulancias en dos municipios de la Baixada Fluminense, en el Estado de Río de Janeiro, y probar la sensibilidad de esos microorganismos a los antimicrobianos. Las muestras se obtuvieron de nueve puntos específicos dentro de 6 ambulancias. Se inocularon en agar manitol con sal y sangre y se analizaron mediante técnicas de ionización y desorción láser asistida por matriz. La sensibilidad de siete muestras se probó utilizando el método de difusión en disco en agar Muller-Hinton. De las cincuenta y dos muestras recolectadas, treinta y una (59,6%) resultaron positivas para algún género de bacteria, dos (3,80%) no fueron reconocidas por MALDI y diecinueve (36,6%) fueron negativas. Es necesario reforzar las prácticas de prevención y control de infecciones, ya que la contaminación puede provocar posibles complicaciones para los pacientes.

Descriptores: Servicios Médicos de Emergencia; Bacteria; Control de Infección; Atención de Emergencia Prehospitalaria; Desinfección.

Resumo

Objetivou-se identificar microrganismos por meio do cultivo microbiano de bactérias do interior de ambulâncias em dois municípios da Baixada Fluminense, no Estado do Rio de Janeiro, e testar a sensibilidade desses microrganismos a antimicrobianos. Amostras foram obtidas de nove pontos específicos do interior de 6 ambulâncias. Elas foram inoculadas no ágar manitol salgado e de sangue, e foram analisadas por meio de técnica de ionização e dessorção a laser assistida por matriz. Foi testada a sensibilidade de sete amostras com o método de difusão em disco em ágar Muller-Hinton. Das cinquenta e duas amostras coletadas, trinta e uma (59,6%) positivaram para algum gênero de bactéria, duas (3,80%) não foram reconhecidas pelo MALDI e dezenove (36,6%) negativaram. É preciso que sejam reforçadas as práticas de prevenção e controle de infecções, dado que a contaminação pode resultar em possíveis complicações para os pacientes.

Descritores: Serviços de Emergência Médica; Bactérias; Controle de Infecções; Atendimento de Emergência Pré-Hospitalar; Desinfecção.

Introduction

Ambulance vehicles are an integral part of Emergency Medical Services (EMS). Emergency ambulances aim to help critically ill patients and prevent the development of potentially fatal complications in cases of serious injury and can respond to thousands of cases per year, after each admission, each vehicle must be cleaned and decontaminated to be ready for use the next time. mission, as they represent a potential source of infection for patients, patients' relatives, and professionals during transport to a healthcare facility or a transfer[1].

During Mobile Pre-Hospital Care (APHM), teams need to provide care to a variety of patients in different environments. They need to be ready to respond to calls, leave the patient safely in a hospital emergency department, and, in that short space of time, also clean and decontaminate all equipment. In addition to these aspects, APHM has increased the complexity of their care, including, proportionally, the number of invasive procedures, which, as they are carried out outside the hospital environment, increase the risk of infections. Another point to be highlighted is the scarcity of information about patients' infection status. Therefore, microbiological assessment of ambulance vehicles is an essential step in infection control that must be considered to develop valuable risk reduction interventions[2,3].

Microorganisms present in healthcare facilities, which include bacteria, fungi, and viruses, are responsible for 1.7 million infections associated with medical services in the United States alone. In developed countries, the rate of Healthcare-Associated Infections reaches 7% per year, while in developing countries, such as Brazil, this number reaches around 14%[3,4].

Although there is disposable equipment, such as Personal Protective Equipment (PPE), and standard precautions - defined under the Manual of Best Practices for Hospital Hygiene and Cleaning, as actions that aim to reduce the risk of transmission of pathogens through blood or from other sources, whether known or unknown, and should always be used in the care of all patients, such as basic handwashing, alcohol-based hand hygiene, PPE, transmission-based precautions, indications of transmission-based precautions -, In ambulances, potentially pathogenic and pathogenic bacteria can still be detected from contamination by blood, secretions, and other infected biological materials[5].

A study in Brazil showed that the services observed in basic and advanced vehicles of the SAMU (Mobile Emergency Care Service), in 87 (83.7%) of the services in the basic vehicle and 131 (100.0%) services in the advanced vehicles, the team did not perform hand hygiene between procedures; and 58 (58.6%) of the visits in the basic vehicle and 101 (77.1%) in the advanced vehicle, there was no change of gloves between procedures, results that confirm the exposure of patients to the risk of infection[6].

Causes for low hand hygiene adherence in emergency departments include urgent clinical situations, insufficient time, and ambiguity about when to perform hand hygiene. The use of gloves has also been associated with poor hand hygiene in emergency and trauma situations. Recent studies indicate that underreporting of cases of exposure by patients and professionals, the lack of safety equipment, the lack of PPE, and the lack of training to use this equipment are contributing factors to the increase in infections in the pre-hospital environment[7,8].

The contamination of equipment and surfaces by potentially pathogenic microorganisms inside an ambulance poses a great risk to patients, as they are often transported in serious conditions. Bacteria can survive for up to months on dry inanimate surfaces, and their ability to form biofilms provides greater survival and resistance to physical and chemical agents used to eliminate them[9-11].

A survey carried out in 16 European countries evaluated 48 isolation facilities and described the results of transport capabilities, focusing on land ambulances regarding legal aspects, technical and infrastructure aspects, and operational procedures, thus, concerning operational aspects, it was found that 34/47 (72.3%) had protocols for disinfecting ambulances and 30/43 (69.8%) had protocols for disinfecting equipment[12].

The objectives of the present study were to identify microorganisms through the microbial cultivation of bacteria inside ambulances in two municipalities in Baixada Fluminense, in the State of Rio de Janeiro, and to test the sensitivity of these microorganisms to antimicrobials.

Methodology

This is a descriptive cross-sectional study, where fifty-two swabs collected from samples inside six ambulances were analyzed, one basic and two advanced from each municipality. The ambulances were named Advanced, Basic 1, and Basic 2, belonging to the Mobile Emergency Care Service of a municipality in Baixada Fluminense in the State of Rio de Janeiro, and the swabs were collected in December 2019. Of these fifty-two samples, seventeen of them were collected from Advanced type ambulances and thirty-five of them, from Basic 1 and 2 types of ambulances. The ambulances had an average of ten professionals on the team, with small variations from one to the other.

For this study, Mobile Units of the Advanced Life Support type (Type D) were selected to collect the samples, which have a nurse, doctor, and driver, and together, these professionals are responsible for care involving procedures of greater complexity and they can be carried out in homes, public roads, hospital institutions and during patient transport. The Basic Life Support (Type B), which has a driver and a technician or nursing assistant, this vehicle is intended for the inter-hospital transport of patients with a known risk to their lives and for pre-hospital care for those at risk. unknown, unclassified, with potential need for medical intervention on site and/or during transportation to the destination service[13].

The professionals working in the vehicles included in this research stated that they did not follow any type of infection control protocol when carrying out cleaning and disinfection processes, carrying them out in a non-standardized manner, which stated that the ambulances

were disinfected using chlorine on the floor and 70% alcohol on the equipment, without using a specific technique.

The Basic type ambulances had, respectively, twenty-eight and thirty-five occurrences, and the Advanced Support ambulances had approximately two occurrences per week. Basic type ambulances had seven incidents per day and Advanced Support ambulances had an average of three to seven. It is worth mentioning that the samples were collected before any ambulance disinfection procedure, however, it was expected that the ambulances would be clean and disinfected so that the vehicle would be ready to respond to an incident.

The samples were collected by two nurses (authors of the research), who received guidance from a professional microbiologist (author of the research) on how to collect the samples. Samples were collected from instruments and places, inside six authorized ambulances, that showed some type of dirt; the places and instruments were: tourniquet, doorknob, oximeter, sphygmomanometer, floor, handrail, O2 cylinder, stretcher, and furniture.

A sterile swab was soaked in saline and then used to collect each sample and inserted into Stuart transport medium for further processing in the laboratory. The samples were transported in Styrofoam boxes immediately after collection. Transport lasted between 30 and 45 minutes after collection, depending on the city. There was no storage between collection and delivery to the laboratory[14].

The samples were inoculated in Blood Agar, a nutrient-rich culture medium that favors the growth of a wide range of microorganisms, and in Salt Mannitol Agar, a selective and differential medium used for the cultivation of Staphylococcus spp. The samples were incubated in the oven at a temperature of 35±2ºC and, after 24h to 48h, their growth was observed. The incubation time was between 24h and 48h, due to the variation in the growth time of the bacteria, which varies within this period of hours. It is important to emphasize that this result did not impact the results of the research, as it is known that some bacteria can take longer than 24 hours to grow in culture media[14].

The isolated colonies were analyzed using an ionization and desorption technique, known as MALDI, which allows the analysis of biomolecules and large organic molecules. All samples were placed on a matrix plate and bombarded with a laser that evaporated them; a system ionized and aspirated all the volatilized material, which reached detectors, which recorded the time in which the substance reached the detector and its quantity. Each microorganism has a characteristic spectrum that is analyzed by software, which allows, in a few minutes, the recognition of the bacteria present in each sample. The process was redone once again with all samples, to analyze the possible increase in scores. Retaking the samples did not compromise the results of this research, since the entire method used for identification was the same[15].

After identifying the bacteria, the sensitivity of 7 samples to the antibiotics Ampicillin 10 µg, Ciprofloxacin 5 µg, Clindamycin 2 µg, Chloramphenicol 30 µg, Erythromycin 15 µg, Gentamicin 10 µg, Linezolid 30 µg, Rifampicin 5 µg, Tetracycline 30 µg, 30µg by disk diffusion method on Muller-Hinton agar.

To perform the sensitivity test, the antimicrobial discs were dispensed onto an agar plate after applying the bacterial inoculum with approximately 1 to 2 x 10 CFU/mL. Plates were incubated for 16 to 24 hours in room air before results were determined. The diameters of the bacterial growth inhibition halos around each disc were measured in millimeters and interpreted by comparing the value of the inhibition halo with the criteria published by CLSI (Clinical and Laboratory Standards Institute)[16], the S. Aureus strain (ATCC 25923) was used as a control. In this way, bacterial samples were categorized as sensitive, resistant, or intermediate. Of the thirty-one positive samples, only 7 of them had their sensitivity tested, due to the COVID-19 pandemic, which made it impossible to continue testing.

The management of the public institution, based in the municipality of Baixada Fluminense, in the State of Rio de Janeiro, authorized the collection and microbiological laboratory analysis without any type of analysis of human waste. The analyses were carried out in a hospital infection laboratory at a federal university located in the city of Rio de Janeiro.

Results

Of the fifty-two samples collected, thirty-one (59.6%) of them tested positive for some bacterial genus, two (3.8%) were not recognized by MALDI and nineteen (36.6%) did not show any results. Bacteria were identified at the level of probable genus (Score: 1,700 to 1,999) or species (Score greater than or equal to 2,000) on different surfaces. Of the thirty-one positive samples, 64.5% were from Advanced-type ambulances and 35.4% were from Basic 1 and 2 type ambulances.

In the Advanced type ambulances, seventeen samples were collected, of which bacteria were detected in eleven (64.7%). Thirty samples were collected in Basic 1 and 2 ambulances, and some types of bacteria were identified in fourteen (40.0%) of them. Despite having less time of use and relatively fewer occurrences than Basic type ambulances, Advanced type ambulances had a higher rate of bacterial identification.

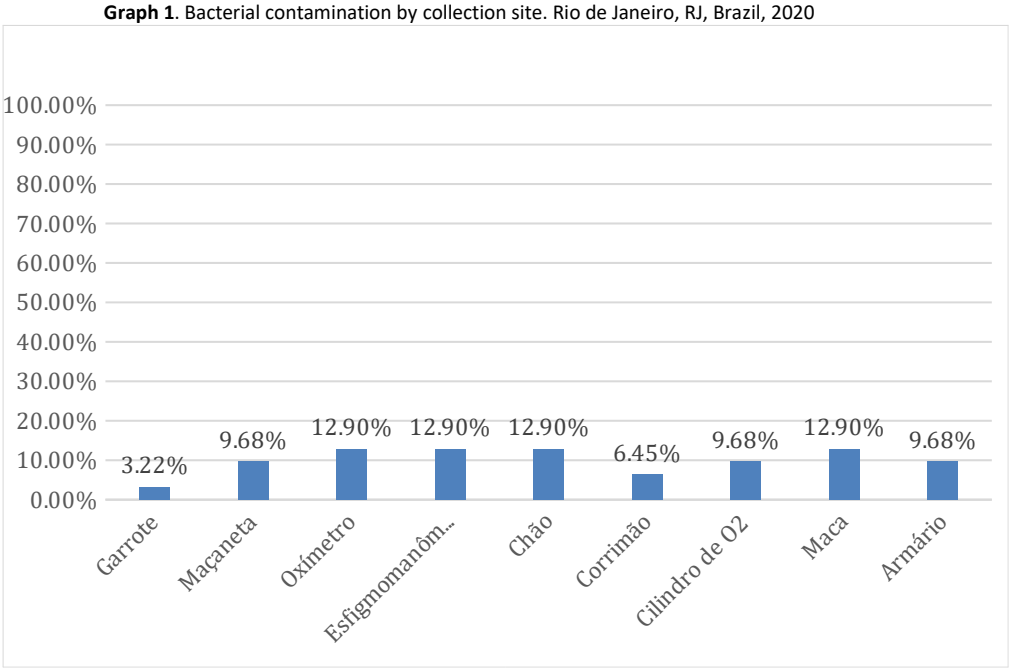
According to Table 1, in fourteen (56.0%) positive samples, Staphylococcus species were found, commonly present on human skin, such as S. saprophyticus, S. hominis, and S. warneri. Other species of bacteria were observed, such as those belonging to the genus Exiguobacterium, Aerococcus (viridans) and Bacillus (marisflavi, oceanisediminis, pumilus and megaterium). Two samples identified bacteria of the genus Enterobacter sp. and Enterococcus sp. The presence of S. aureus was identified in none of them.

As shown in Graph 1, the collection sites with the highest detection of bacteria inside the ambulances were the oximeter, the stretcher, and the sphygmomanometer with 4 (12.9%) positive samples each. The doorknob, O2 cylinder, and cabinet each had 3 (9.6%) positive samples. The place with the lowest contamination rate was the garrote.

Table 1. Distribution of bacteria identification using the MALDI method, according to the type of ambulance. Rio de Janeiro, RJ, Brazil, 2020

Identified bacteria	Basic 1 N=10	Basic 2 N=10	Advanced N=11
Staphylococcus saprophyticus	-	-	6(54.5%)
Staphylococcus sp	-	2(20.0%)	1(9.1%)
Enterococcus sp	-	-	1(9.1%)
Staphylococcus xylosus	1(10.0%)	-	-
Bacillus marisflavi	2(20.0%)	1(10.0%)	-
Bacillus sp	1(10.0%)	-	-
Exiguobacterium sp	1(10.0%)	-	-
Siccibactercolletis/bacillus marisflavi	1(10.0%)	-	-
Siccibactercolletis	-	1(10.0%)	-
Bacillus pumilus/ pseudomonas stutzeri	-	1(10.0%)	-
Bacillus pumilus	-	1(10.0%)	1(9.1%)
Enterobacter sp	-	1(10.0%)	-
Bacillus oceanisediminis	-	-	1(9.1%)
Staphylococcus hominis	1(10.0%)	2(20.0%)	1(9.1%)
Staphylococcus warneri	1(10.0%)	-	-
Bacillus megaterium	1(10.0%)	-	-
Aerococcusviridans/ bacillus megaterium	-	1(10.0%)	-

Graph 1. Bacterial contamination by collection site. Rio de Janeiro, RJ, Brazil, 2020



The antimicrobial susceptibility test showed that the Enterococcus sample was resistant to Tetracycline and has intermediate resistance to Rifampicin, Erythromycin, and Ciprofloxacin, and is sensitive to Vancomycin. S. saprophyticus was resistant to Chloramphenicol and Erythromycin; S. xylosus was resistant to Tetracycline.

Regarding the relationship between the frequency of occurrences and the detection of bacteria, Advanced type ambulances, with an average of 2 occurrences per week, presented the highest contamination rate with 8 (88.9%) of 9 collection sites with the detection of bacteria, including those considered most pathogenic. Next, Basic type ambulances with 7 (70.0%) of ten contaminated sites, with an average of forty-nine occurrences per week. The remainder of the ambulances analyzed had 3 contaminated locations, with an average of twenty-eight to forty-nine occurrences per week and were of the Advanced and Basic types. Advanced-type ambulances had a disinfection rate of 47.0% and Basic type ambulances had a disinfection rate of 48.5%.

Table 2. Quantity and frequency of antimicrobial resistance in samples collected from the study. Rio de Janeiro, RJ, Brazil, 2020

	Number and (%) of samples in the Multidrug Resistance category				
		Sample susceptibility			
Antimicrobials	**Total**	**Sensitive**	**Resistant**	**Intermediate**	**Not determined**
	N=70	**N=48**	**N=4**	**N=3**	**N=15**
Ampicilina 10 µg	7(10.0%)	1(2.1%)	-	-	6(40.0%)
Ciprofloxacina 5µg	7(10.0%)	6(12.5%)	-	1(33.3%)	-
Clindamicina 2µg	7(10.0%)	5(10.4%)	-	-	2(13.3%)
Cloranfenicol 30 µg	7(10.0%)	6(12.5%)	1(25.0%)	-	-
Eritromicina 15 µg	7(10.0%)	5(10.4%)	1(25.0%)	1(33.3%)	-
Gentamicina 10 µg	7(10.0%)	6(12.5%)	-	-	1(6.6%)
Linezolida 30 µg	7(10.0%)	7(14.6%)	-	-	-
Rifampicina 5 µg	7(10.0%)	6(12.5%)	-	1(33.3%)	-
Tetraciclina 30µg	7(10.0%)	5(10.4%)	2(50.0%)	-	-
Vancomicina 30µg	7(10.0%)	1(2.1%)	-	-	5(33.3%)

Chart 1. Sensitivity of samples to antibiotics. Rio de Janeiro, RJ, Brazil, 2020

Identification (sample)	CLI	CLO	TET	ERI	RIF	GEN	LNZ	CIP	VAN	AMP
Enterococcus sp (5)	ND	S	**R**	**I**	**I**	ND	S	**I**	S	S
Staphylococcus saprophyticus (2)	S	S	S	S	S	S	S	S	ND	ND
Staphylococcus xylosus (10)	S	**R**	S	**R**	S	S	S	S	ND	ND
Staphylococcus warneri (36)	S	S	**R**	S	S	S	S	S	ND	ND
S. hominis (43)	S	S	S	S	S	S	S	S	ND	ND
S. hominis (49)	ND	S	S	S	S	S	S	S	ND	ND
S. hominis (50)	S	S	S	S	S	S	S	S	ND	ND

Notes: S: Sensitive, R: Resistant, I: Intermediate, ND: Not determined.

Discussion

The results of this research are in line with the idea of another study that states that the surfaces of ambulances that are surrounding and directly in contact with the patient are often exposed to microorganisms. Likewise, they agree that microorganisms are not only detected inside ambulances but also in the equipment used by teams[1].

According to a study, it was found that the presence of pathogens is most often associated with areas of high contact with the patient and/or contact with the emergency medical team, such as stretchers (mattresses and handrails), door handles, areas preparation of APHM workers and steering wheels, which corroborates the findings of the present study. The floor of the ambulances analyzed was one of the most contaminated collection sites (4 samples) and in this location, a genus was identified that could be significantly pathogenic[3].

Researchers noted that the stretcher, specifically the headboard, was the place that showed the most contamination by Staphylococcus, as in the present research, in which of the 4 samples collected from the stretchers, 3 of them belonged to this genus. Other objects that had a similar degree of contamination were the oximeter and the sphygmomanometer[1].

O_2 cylinders are assistance instruments that deserve a lot of attention from the APHM team, as they are the only medical equipment that needs to be refilled and for this reason, if they are contaminated, they can spread

microorganisms to large areas[12]. The results of this research showed that the O_2 cylinder was one of the most contaminated collection sites inside ambulances.

According to research findings, bacteria of the genus Staphylococcus were also predominantly found. The saprophyticus species was the most identified of the genus in this research and in recent years it has been found in studies involving cases of acute pyelonephritis, septicemia, nephrolithiasis and endocarditis[17].

Other species of bacteria were observed, such as those belonging to the genus Exiguobacterium, Aerococcus (viridans), which is already considered a pathogen of the urinary tract in humans, and Bacillus (marisflavi, oceanisediminis, pumilus and megaterium). Strains of B. pumilus associated with foodborne illnesses and from clinical settings have been isolated and several strains of B. pumilus, such as strain F4791/87 and F4552/87, have been recognized to produce toxins that are harmful to humans. Two isolates of B. pumilus have been identified that cause severe sepsis in newborns[18].

In one of the samples (collected from the floor of the Advanced type of ambulance), the presence of Enterococcus sp. was evidenced, a commensal of the human digestive system, but depending on the species, if found in the environment, it can cause endocarditis, bacteremia, tract infections urinary, intra-abdominal and wounds. It is estimated that, currently, the species Enterococcus faecalis is responsible for most infections in humans. The pathogen exhibits intrinsic resistance to some classes of antibiotics such as β-lactams, lincosamides, trimethoprim-sulfamethoxazole, fluoroquinolones, and low concentrations of aminoglycosides, making it difficult to choose effective therapeutic strategies. This microorganism has a high capacity to survive in adverse environments, which facilitates its ability to cause diseases in a nosocomial environment[19,20].

The bacteria can also acquire resistance to high concentrations of glycopeptides (vancomycin and teicoplanin), tetracyclines, erythromycin, rifampicin, chloramphenicol, and nitrofurantoin, as found in the antibiogram. The antibiogram result detected resistance of the Enterococcus sp. Tetracycline. Tetracycline is an antibiotic active against gram-positive and gram-negative aerobic bacteria, with a widely used bacteriostatic effect. Inhibits bacterial protein synthesis by binding to the 30S subunit of the bacterial ribosome, preventing aminoacyl-tRNA from accessing the receptor site in the ribosomal mRNA complex[20-22].

Furthermore, the Enterococcus sample in this study was sensitive to Vancomycin. This is a significant result, as studies indicate that contact with patients colonized by Vancomycin-Resistant Enterococcus (VRE) favors contamination of healthcare professionals, as does contact with contaminated surfaces in wards where patients with VRE are present. The number of procedures performed is also associated with the acquisition of VRE by professionals[23]. If it is assessed that APHM professionals generally need to perform many procedures in a short space of time, given the severity of the patients, the risk for professionals in these services can be suggested.

Therefore, as recommended by a study, it is important that all equipment that meets the skin and/or mucous membranes is subjected to a two-step cleaning and disinfection process after each use, and this hygiene must be carried out in an appropriate way to prevent the spread of microorganisms between professionals and patients. Another study showed that the number of contaminated areas inside an ambulance before cleaning and disinfection increased from 57.0% to 86.0%, demonstrating how the lack of these processes can lead to an increase in contamination inside vehicles[24].

In Denmark, guidelines mandate thorough cleaning of ambulances once a day, including tidying up the patient compartment, wiping down all surfaces, and sweeping and washing floors, and moderate/regular cleaning should be carried out after caring for each patient and cleaning. This involves cleaning the patient compartment and cleaning areas where there is manual touch using pre-soaked wipes containing a detergent or disinfectant depending on the risks of the previously treated patient. Such cleaning is very important, as researchers link the high presence of Staphylococcus with inadequate cleaning and disinfection of vehicles and assistance instruments at organized times[1,2].

In addition to the correct cleaning and disinfection of ambulances, the correct use of PPE is part of the prevention practices that must be adopted by all healthcare professionals. The proper use of this equipment guarantees protection against infectious agents that may be present in the blood, body fluids and mucous membranes of patients and it is necessary for the professional to also understand which PPE must be used in each operating scenario, which will be determined by factors such as the degree of contact with the patient and the extent of blood and bodily fluids to which the professional will be exposed[8].

Gloves, for example, for APHM professionals need to be more resistant due to the typical work routine. Latex and nitrile gloves may not be as durable as necessary and the PPE also needs to be suitable for the individual's size, comfortable, and should not hinder (or as little as possible) the professional's performance, just like protective glasses, for example, must be well adjusted to the person's face and must allow peripheral vision[8].

Therefore, it is important to create programs aimed at preventing the transmission of agents that cause infectious diseases and promoting a safe care environment to provide the necessary assistance, which includes administrative and engineering control programs, control of workplace practices, education, medical and immunization management[8].

Therefore, to improve the effectiveness of infection control programs, staff training, continuing education, real-time feedback on the effectiveness of cleaning and disinfection procedures, routine microbiological inspection of surface hygiene, and the use of fluorescent markers or assays to verify the robustness of the process. Non-manual disinfection of ambulances may also be a possibility, as it reduces human errors and ensures a more effective

elimination of pathogens, even though there is still a lack of definitive evidence in the scientific literature regarding its effectiveness. Furthermore, it should be noted that hand hygiene by professionals is an action that reduces the risk of infection and should be encouraged on an ongoing basis. Because even though it is a simple procedure, full adherence to the hand hygiene steps is still a challenge[24,25].

Conclusion

The study identified the presence of bacteria that could be harmful to the health of patients transported by ambulances, as well as to the professionals who work there. Mobile pre-hospital care is a dynamic, complex, and little-studied process, especially in the Brazilian reality, which demonstrates the need to invest in research that can contribute to better care in this environment and thus provide safe assistance. In general, due to the presence of some bacteria that are sensitive and/or resistant to the antimicrobials used against them, it is necessary to reinforce and even intervene in the hygiene and disinfection protocols of ambulances to avoid subsequent transmission.

This study highlights the importance of cleaning and disinfecting ambulances regularly as a way of ensuring safe healthcare. It also contributes to the identification of some bacteria, so that early intervention can be taken to avoid any type of contamination. In this sense, there is a need for new studies in the area to encourage the practice of health professionals.

The study's limitation is the fact that it only presented qualitative and not quantitative microbial evaluation of the samples since it is not possible to have a sterile environment. However, the number of microorganisms can be controlled at low levels. And since an antibiogram was not carried out on all samples to check whether the identified bacteria were resistant to the main antimicrobials used in treatments, due to the advent of the COVID-19 pandemic, which restricted testing time. Some of the ambulances analyzed had little circulation time and, therefore, the results may change if analyzed from a longer period of use.

Furthermore, in this study, samples from private service vehicles or other public services were not obtained, which restricted the scope.

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