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Egg crater mattresses: a deposit of methicillin-resistant *Staphylococcus aureus*?

COLCHÕES DO TIPO CAIXA DE OVO: UM RESERVATÓRIO DE STAPHYLOCOCCUS AUREUS RESISTENTE À METICILINA?

COLCHONES DEL TIPO CAJA DE HUEVOS: ¿UN RESERVORIO DE STAPHYLOCOCCUS AUREUS RESISTENTES A LA METICILINA?

Adriano Menis Ferreira¹, Denise de Andrade², Margarete Teresa Gottardo de Almeida³, Keith Cássia Cunha⁴, Marcelo Alessandro Rigotti⁵

ABSTRACT

The purpose of this study was to evaluate the microbiological conditions of the *egg crater* mattress in hospital use to identify the presence of *Staphylococcus aureus* and its methicillin-resistance phenotype (MRSA). Petrifilm™ plates were used to collect the microbiological data from the mattresses, in pre-established positions. A total 180 plates were collected in 15 mattresses, 139 (77.2%) of which were positive for *Staphylococcus aureus*. Of the positive plates, 77 (55.4%) were collected before and 62 (44.6%) after washing the mattresses. There was a significant reduction ($p=0,023$) in Colony Forming Units (CFU); however, regarding the resistance profile, 8 (5.3%) mattresses with MRSA were identified. Results show the risk of these mattresses acting as a secondary deposit in the infection chain, especially regarding the presence of MRSA.

KEY WORDS

Beds.
Staphylococcus aureus.
Equipment contamination.
Cross infection.
Methicillin resistance.

RESUMO

O estudo teve como objetivo avaliar as condições microbiológicas de colchões *caixa de ovo* em uso hospitalar com a finalidade de identificar a presença de *Staphylococcus aureus* e seu fenótipo de resistência à meticilina (MRSA). Coletaram-se as amostras microbiológicas nos colchões por meio de placas de contato Petrifilm™ em posições pré-estabelecidas. Totalizou-se 180 placas coletadas em 15 colchões, das quais 139 (77,2%) foram positivas para *Staphylococcus aureus*. Desse total, 77 (55,4%) e 62 (44,6%) corresponderam respectivamente à coleta antes e após a lavagem dos colchões. Evidenciou-se redução significativa ($p=0,023$) das Unidades Formadoras de Colônias (UFC), entretanto com relação ao perfil de resistência foi identificado 8 (5,3%) colchões com MRSA. Diante dos resultados, pode-se inferir sobre o risco destes colchões atuarem como reservatórios secundários na cadeia de infecção, especialmente no que se refere à presença de MRSA.

DESCRIPTORES

Leitos.
Staphylococcus aureus.
Contaminação de equipamentos.
Infecção hospitalar.
Resistência a meticilina.

RESUMEN

El estudio tuvo como objetivo evaluar las condiciones del tipo *caja de huevos* en uso hospitalario con la finalidad de identificar la presencia de *Staphylococcus aureus* y su fenotipo de resistencia a la meticilina (MRSA). Se recolectaron las muestras microbiológicas en los colchones a través de placas de contacto Petrifilm™ en posiciones preestablecidas. Se totalizaron 180 placas colectadas en 15 colchones, de las cuales 139 (77,2%) fueron positivas para *Staphylococcus aureus*. De ese total, 77 (55,4%) y 62 (44,6%) correspondieron respectivamente a la recolección anterior y posterior al lavado de los colchones. Se evidenció reducción significativa ($p=0,023$) de las Unidades Formadoras de Colonias (UFC), mientras que con relación al perfil de resistencia se identificaron 8 (5,3%) colchones con MRSA. Ante tales resultados, se puede inferir el riesgo de que estos colchones actúen como reservorios secundarios en la cadena de infección, especialmente en lo que se refiere a la presencia de MRSA.

DESCRIPTORES

Lechos.
Staphylococcus aureus.
Contaminación de equipos.
Infección hospitalaria.
Resistencia a la meticilina.

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INTRODUCTION

The increase and dissemination of multiresistant microorganisms in hospital environment is a great concern and keeps challenging the controller of infection and the epidemiological practices. The frequency in which the cross contamination occurs varies from 13 to 34.6%. The situation is a concern when observing that the prevalence of hospital infections involving multiresistant bacteria has continuously increased in several countries since the decade of 1980. Multiresistant bacteria have become endemic in several hospitals, even when control measures are implemented⁽¹⁾.

Several microorganisms can survive and remain viable in dry surfaces for days, weeks and even months. For instance, strains of methicillin-resistant *Staphylococcus aureus* (MRSA) may remain viable for over 14 days in surfaces of Formica and over 6 to 9 weeks in cotton blankets^(1,2).

It is known that the patient's endogenous microbiota is a real source of infection, which may be transmitted through the health team professionals, mainly when they do not hygienize their hands⁽³⁾. Hereafter, the inanimate environment that surrounds him and the objects used in the care also contain microorganisms that may be transmitted to the patients, professionals and to other objects⁽¹⁻³⁾.

The contamination of the hospital environment by gram-positive bacteria like MRSA and vancomycin-resistant *Enterococcus* (VRE), specially in epidemic situation, has been frequently described⁽¹⁾. The proportion of hospital surfaces contaminated with MRSA has varied considerably from 1% to 27% in hospital rooms to 64% of surfaces in Burn Units with patients carrying MRSA⁽²⁾. Therefore, the environmental contamination may contribute to the transmission of microorganisms that are epidemiologically important in the genesis of hospital infections.

Although it is not possible to state that contaminated surfaces and objects may cause infections, undoubtedly there are evidences that these sources act as secondary deposits and may lead to cross infections^(1,4).

Several epidemiological studies whose microbiological samples were collected in the hospital environment have indicated that multiresistant bacteria are present in many objects and surfaces⁽¹⁾. The most frequently contaminated surfaces are: floor, bed structure, tables, clothes used by the patients, pillows and mattresses^(1,4).

Among the objects that are kept near the patient, the mattress is the one that has more contact to the body, and may also serve as deposit for organic and/or inorganic dirtiness and for microorganisms responsible for infec-

tions. According to the literature, there is no doubt that the environment and the objects close to the patient get contaminated with multiresistant bacteria. Among the objects, the mattress is a deposit element of these bacteria. However, the studies that evaluated the microbiology of the mattresses have done it through the collection of samples from the external covering, not directly from the foam mattress⁽⁵⁾.

One of the resources that is frequently used to prevent the development of pressure ulcers has been the pyramidal mattress *egg crate*, mainly in hospitalized patients whose treatment and constant monitoring lead to a long stay in bed or absolute rest, making him more likely to develop pressure ulcers. In general, these mattresses are used over the conventional hospital mattresses and are only protected by the bedclothes. Besides, this covering does not configure a barrier to the absorption of humidity and dirtiness. Therefore, its contamination by bacteria resulting, mainly, from the patient is possible. These considerations foment the possibility that these mattresses contain microorganisms, including multiresistant ones.

OBJECTIVES

This study had the objectives to: quantify the Colony-Forming Units (CFU) of *Staphylococcus aureus* present in "egg crate" mattresses used by hospitalized patients in absence, countable and uncountable state, before and after washing the mattresses and to check the frequency of the phenotype MRSA in the analyzed mattresses.

METHOD

Study location

The study was developed at a large tertiary School Hospital, situated in the interior of the state of São Paulo in the months of October and November of 2008. The authorization of the administrative parties was obtained for its execution.

Sample collection of the mattresses

Samples were collected from 15 leaden "egg crate" mattresses, with the following dimensions: 180x80x4cm in pyramidal format and made of polyurethane foam.

The mattresses were marked with paint on the surface that was in contact to the patient, that is, at the headboard, central part and feet. Each mattress was divided in three areas (superior, medium and inferior) and 2 samples were collected from each area (Figure 1).

According to the literature, there is no doubt that the environment and the objects close to the patient get contaminated with multiresistant bacteria.

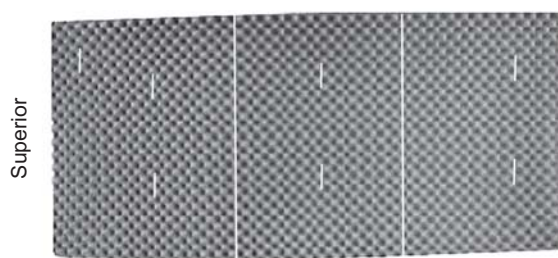


Figure 1 - Panoramic view of the areas of the "egg crate" mattress and respective collection areas - São José do Rio Preto, SP - 2008

Afterwards, each mattress was individually packed in plastic bags and sent to the laundry department. After going through the washing process (Table 1), new samples were collected from the same places previous to the process. Thus, 6 samples were collected before the mattresses were washed and 6 samples were collected after.

For the collection of microorganisms, the authors used Petrifilm™ plates (3M™, St Paul, MN, USA) model Staph Express 3M™ prepared with modified cromatogenic medium of Baird-Parker, which is selected and differential for *Staphylococcus aureus*.

Table 1 - Experimental laundry process of the mattresses - São José do Rio Preto, SP - 2008

Operation	Time (min)	Temperature (°C)	Level of water	Product	Dosage
Rinsing	1	Cold	High	-----	-----
Washing/Bleaching	10	Cold	Medium	Detergent	400ml
-----	-----	-----	-----	Sodium hypochlorite 8%	100ml
Rinsing	1	Cold	High	Water	-----
Rinsing	1	Cold	High	Water	-----
Neutralizer/Softener	3	Cold	High	Chlorine neutralizer	100ml
-----	-----	-----	-----	Softener	250ml

At the end of each collection, the plates were identified with: date, time, area (superior, medium and inferior) and moment of the collection: before washing (BW) and after washing (AW), packed in polystyrene boxes and transported to the Laboratory of Microbiology.

For the Petrifilm™ model, it was decided to adopt a sampling area of 30 cm² and incubation at 35°C during 24-48h. The reading of the Petrifilm™ plates was performed with the use of stereomicroscope (Nikon, JP) under reflected light and described in CFU. Red-violet colonies were considered as *Staphylococcus aureus*. Aimed at standardizing the reading process of the plates, the following denominations were established: (1) absence: plate without the development of colonies; (2) countable: plate with the development of until 150 colonies; (3) uncountable: plate with the development of over 150 colonies.

The susceptibility to the methicillin was verified by a sorting test for resistance to oxacillin⁽⁶⁾. Petri dishes were used, containing Muller-Hinton agar supplemented with 4% of NaCl and 6 µg of oxacillin, known as MRSA medium (Probac do Brasil®). These microorganisms were transferred into BHI broth and incubated at 37°C for 24 hours. After this period, they were inoculated in the plates and incubated at 37°C for 24 and 48 hours. The growth in the plates was considered positive for MRSA.

Statistical procedures

The exploratory analysis calculated the mean, median and standard deviation of the CFU in the two moments (before and after washing) considering the superior, medium

and inferior positions of the mattresses. In the confirmatory statistical analysis the non-parametrical Wilcoxon Matched Paris Test was used with a level of significance of $p < 0.05$.

RESULTS

Fifteen *egg crate* mattresses used by hospitalized patients in bed were microbiologically analyzed.

Frequency of positive and negative cultures before and after washing the mattresses

Among the 180 plates used for collecting the samples in the 15 mattresses, 139 (77.2%) were positive for *Staphylococcus aureus*. Among this total of positivity, 77 (55.4%) and 62 (44.6%) corresponded to those collected before and after washing the mattresses respectively. There was a reduction observed in the quantity of positive cultures in only 15 plates.

Among the 90 plates examined after the continuous use of the mattresses, 61 (67.7%) presented countable colonies, 16 (17.7%) uncountable colonies and 13 (14.4%) the absence of colonies. After washing, the situation of the plates was: 59 (65.5%) with countable colonies, 28 (31.1%) absence and 03 (3.3%) with uncountable colonies.

The distribution of the CFU according to their position (superior, medium and inferior region of the mattresses) and, to the moment of collection (before and after washing) is presented in Table 2.

Table 2 - Measures of position and variability of the *Staphylococcus aureus* Colony-formation Units (CFU) according to the position in the mattresses and the moment of collection (before or after washing) - São José do Rio Preto, SP - 2008

Collection position	Before washing			After washing			p
	Mean	Median	Standard deviation	Mean	Median	Standard deviation	
Superior	95.2	22	108.2	50.4	15	58.4	0.211
Medium	71.4	20	90.2	18.8	12	22.09	0.013
Inferior	78.06	35	99.7	23.9	10	39.7	0.016
Total	224.6	252	210.6	93.1	97	69.7	0.023

Note: (n=15)

It was observed that, in general, there was a significant reduction of the CFU, with exception only to the superior region of the mattresses ($p=0.211$). Considering the total-ity of CFU before and after washing, a statistically significant reduction was evidenced.

Among the 15 evaluated mattresses, methicillin-resistant *Staphylococcus aureus* was identified in 8 (53.3%) of them. It is important to add to this situation the fact that 05 mattresses presented MRSA before the washing process and 03 after it. Among the mattresses that presented MRSA before the washing process, 01 of them kept the same situation after this process. It is necessary to highlight that MRSA was identified in 02 mattresses that did not present strains of this nature before being washed.

DISCUSSION

The objective of this study was to evaluate the presence of *Staphylococcus aureus* and MRSA in egg crate mattresses, used by hospitalized patients.

Initially, it is important to highlight that *Staphylococcus aureus* is considered the main agent causing infections acquired in the community and in the hospital environment. This microorganism represents one of the most frequent causes of infections associated to the health care reported by the National Nosocomial Infection Surveillance System (EUA), including ventilator-associated pneumonia, surgical site infections and catheter-associated bacteremia⁽¹⁻²⁾.

The direct contact of the patients to the mattresses may cause a microbial contamination resulting from body fluids and cells that scale from the skin, and, thus the surface of the mattresses become a source of infection⁽⁷⁾. There are evidences that the MRSA may survive in an inanimate environment for several months^(4,8-9).

Mattresses are usually made of foam, whereas the cover is normally made of synthetic substances such as polyurethane, vinyl, polyethylene or polyester. These covers must be impermeable to liquids, but permeable to steam⁽¹⁰⁾. Besides, these characteristics preserve their integrity and fewer conditions to act as microbial development and culture means.

Several studies have demonstrated the contamination of MRSA in mattresses and, consequently, highlighted the

risk of cross infection⁽⁹⁻¹⁶⁾. Therefore, mattresses can be secondary deposits for MRSA and the contamination may persist even after washing them with detergent⁽¹¹⁾.

It is worth highlighting that the mattresses used in this study differ from the mattresses of the previously mentioned studies, since when referring to the presence of MRSA, this microorganism was generally collected from the cover that protects them. In the present study, the samples had their origin directly from the raw material (polyurethane) of the mattresses, since they do not have covers that may be disinfected by mechanical action of disinfection with alcohol or other antimicrobial product.

The mattresses were washed according to the protocol previously described, in the attempt of a possible reutilization, considering their microbiological safety. Thus, after the process of washing the mattresses, they were transferred to the centrifuge and, after the cycle, the collections were performed in the same locations as before washing. It is important to highlight that after the centrifugation the mattresses still remained with a certain proportion of humidity and, as they are thermo-sensitive, it was unfeasible to submit them to the process of industrial drying.

Mattresses used in an attempt to prevent pressure ulcers need cleaning and disinfection between the use of one patient and another or in weekly intervals when used by the same patient⁽¹⁰⁾.

It was observed that, in general, this process reduced the microbial load of *Staphylococcus aureus* of the mattresses, except for the superior position ($p=0.211$). This reduction probably results from the mechanical and chemical action, since the hypochlorite is an antimicrobial agent⁽¹³⁾. It is important to highlight that during the washing stage, at least four mattresses were placed in the machine, since it would be economically unfeasible to wash them individually. Therefore, it is not possible to conclude that quantitatively the number of CFU would be the same in case it was possible to wash them individually.

There is still the question about what is the infecting dose of *Staphylococcus aureus* that may represent a risk of infection? Experimental studies indicate that from 10 to 10⁶ CFU may cause infection⁽¹⁴⁾. Therefore, it is possible to infer that, considering the clinical conditions of the hospitalized patients, few CFU resulting from the environment may be transported to vulnerable sites with the possibility to cause an infection.

Another important aspect that was observed after the process of washing the mattresses was the permanence of dirtiness like blood, stains, presence of hair, among others. The foam of the mattresses acts as a favorable environment for the growth of bacteria, including *Staphylococcus aureus*, since they use substances contained in the matrix of the polyurethane as a source of carbon and nitrogen⁽¹²⁾.

The inanimate environment may represent risk of infection whenever it presents a microbial load and, thus, cause the dissemination of microorganisms, including multiresistant ones. *Staphylococcus*, including MRSA, may survive in dry environmental conditions and persist in clinical areas that are inadequately cleaned⁽¹⁴⁾.

Guidelines emphasize preventive measures like the rational use of antibiotics, monitoring of high risk patients, active surveillance and isolation of patients, methods of disinfection and sterilization, among other measures⁽⁴⁾. It is important to highlight that, as for the risk the infection associated to the inanimate environment or surfaces, there is a need for investments in terms of surveillance and control.

A study indicates that the environment near patients who are colonized or infected with gram-positive multiresistant bacteria becomes frequently contaminated and, therefore, surfaces and objects may probably act as secondary deposits for cross transmission⁽⁴⁾.

The effective participation of the inanimate environment in the transmission of MRSA is still not totally established. The environment may act as a secondary deposit for this microorganism, contaminating a variety of hospital surfaces and equipment, and surviving for long periods of time^(1-2,9,12,14-15). The demonstration of the direct transmission of MRSA from the environment to the patient is a hard task, but there are evidences that the environmental contamination with MRSA is enough to contaminate the gloves of health professionals and the use of molecular biotyping has related environmental strains to those that patients acquired in intensive care units^(1,15).

Evidences of the acquisition of MRSA from the environment by patients have been demonstrated by molecular biotyping techniques⁽¹⁵⁻¹⁶⁾. A study⁽¹⁵⁾ observed that 3 out of 26 patients of an Intensive Care Unit were colonized by MRSA, which was acquired from the environment. Another study⁽¹⁶⁾ proved that patients may acquire the same strain of MRSA from the environment at the Intensive Care Unit when they have been recently allocated in the same unit.

The main means through which patients acquire MRSA is through the hands of the health professionals, but studies have demonstrated that, even with a satisfactory compliance with the measures of hygiene of the hands, the cross contamination still occurs⁽¹⁷⁾. This fact is probably due to the possibility of contamination of the environmental surfaces and objects of care to the patient.

Studies about the contamination of the health professionals' hands support the hypothesis of transmission of

bacteria in the clinical environment⁽¹⁷⁾. In a study, 15% of the nurses who worked at an isolation unit carried in average 10,000 CFU of *Staphylococcus aureus* in their hands. In another one, *S aureus* was recovered from the hands of 20% (67 out of 328) of the ICU team, 21% from the hands of doctors (69 out of 328) and 5% (16 out of 328) from nurses who were carriers, and over 3 CFU were recovered from the hands of these professionals. Studies show that the hands or gloves of health professionals get contaminated with MRSA through the contact with hospital articles or surfaces^(1-2,17-19).

In a study, 42% of 12 nurses, who did not have direct to the patients, contaminated their gloves by touching objects in the rooms of patients who carried MRSA in the wound or urine⁽¹⁾. In another study, 31% of the volunteers who touched the bed grid and the shelf above the bed in patient rooms contaminated their hands with *Staphylococcus aureus* and among these, 35% were MRSA. When volunteers touched the same objects, in empty rooms that went through the process of terminal cleaning, 7% contaminated their hands with *Staphylococcus aureus*⁽¹⁸⁾.

As for the MRSA phenotype, it was identified in eight mattresses, being five before washing and three afterwards. However, in two mattresses this phenotype was recovered after the washing process, not before. Therefore, it is believed that either the adopted washing process is not efficient, or during this process, colonies of MRSA may have dislocated from other regions of the mattresses as they were in the deepest part of the foam. It is still necessary to consider that the mattresses were submitted to the washing process, in industrial machine and tied (Figure 2), this situation may have complicated the penetration of the cleaning products and the disinfectant. Nevertheless, this preparation was necessary because, when trying to wash them loose, they were torn, which made them unfeasible for use and for the microbiological collection. Therefore, it is evident that there are not possibilities, considering the microbiological condition of possible reutilization of these mattresses, even by the same patient.



Figure 2 - Panoramic view of the egg crate mattress prepared to be washed in an industrial machine - São José do Rio Preto, SP - 2008

In this study, the prevalence of MRSA was at 53.3%. Other studies have demonstrated high rates of contamination of surfaces close to patients, such as 22.0%⁽¹⁵⁾, 24.0%⁽⁴⁾, 27.0%⁽¹⁾, 50.9%⁽¹⁹⁾ and 54.0%⁽⁹⁾.

Despite of being considered as non-critical items and with a low risk of causing infection, *egg crate* mattresses may become critical due to the contamination by fluids, secretions and excreta and, therefore, they represent a secondary deposit of *Staphylococcus aureus* and MRSA, consequently contaminating the health professionals hands and clothes, equipment and, finally, contributing to the increase of the infection levels.

CONCLUSION

The present study provides important contributions regarding the microbiological safety of *egg crate* mattresses. It is possible to conclude that:

- There was colonization by *Staphylococcus aureus* in the mattresses before and after the washing process;
- After washing the mattresses, there was a statistically significant reduction of the colonies of *Staphylococcus aureus*, except at the superior position ($p=0.211$).
- The standardized washing process was proved to be ineffective, because even though it reduced the number of CFU, they remained in a significant number and in several mattresses the dirtiness was macroscopically evident.
- The phenotype MRSA was detected in 8 (53.3%) mattresses, 5 mattresses presented this phenotype before the washing process and 3 afterwards, and among these, 02 presented MRSA that were not identified before the washing process.

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