

OCCURRENCE OF BACTERIA BELONGING TO THE GENUS *ENTEROCOCCUS* AND *STAPHYLOCOCCUS* ON INANIMATE SURFACES OF SELECTED HOSPITAL FACILITIES AND THEIR NOSOCOMIAL SIGNIFICANCE

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SUMMARY

Objectives: This work aimed to determine the representation and resistance of bacteria belonging to the genus *Staphylococcus* and *Enterococcus* on inanimate surfaces of two selected workplaces of the University Hospital of L. Pasteur in Košice (UHLP) and to investigate their importance in the hospital environment. The men's ward of the Department of Internal Medicine (DIM) and the Department of Anaesthesiology and Intensive Care (DAIC) were chosen.

Methods: Using sterile sampling kits, a total of 182 swabs were collected from the inanimate surfaces of both UHLP workplaces. The swabs were then transported to a microbiological laboratory and inoculated onto sterile culture media (blood agar containing 5% ram erythrocytes). After culturing (24–48 hours, in a thermostat at constant temperature 37 °C), bacterial colonies were identified by mass spectrometry on a MALDI TOF MS. Bacteria belonging to the genera *Staphylococcus* and *Enterococcus* were subsequently separated from the spectrum of identified bacteria. Nosocomial significant strains of *staphylococci* (*Staphylococcus epidermidis*, *Staphylococcus haemolyticus*, *Staphylococcus aureus*) and all isolated *enterococci* were subjected to susceptibility testing for selected antibiotics using the disk diffusion method – E-tests.

Results: Several members of the genus *Staphylococcus* were identified from the inanimate surfaces of both workplaces. These were mainly coagulase-negative strains – *Staphylococcus epidermidis* (45), *Staphylococcus capitis* (34), *Staphylococcus haemolyticus* (20), *Staphylococcus hominis* (45), *Staphylococcus pasteurii* (2), *Staphylococcus saprophyticus* (1), *Staphylococcus simulans* (3), and *Staphylococcus warneri* (4). *Staphylococcus aureus* strains were also identified (2). Nosocomial significant isolates were tested for susceptibility to the antibiotics cefoxitin (FOX) and oxacillin (OXA). Two members of the genus *Enterococcus* – *Enterococcus faecium* (7) and *Enterococcus faecalis* (8) were isolated. All strains were subject to vancomycin susceptibility testing using the disk method.

Conclusion:

Key words: nosocomial, inanimate surfaces, Enterococcus, Staphylococcus

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INTRODUCTION

Despite the progress of modern medicine, nosocomial infections are still an up-to-date issue and are often discussed because of their threat to hospitalized patients and the medical staff. The fact that the resistance of nosocomial pathogens is constantly growing forces us to address this global problem. Antibiotics play a central role in the growth of resistance, and reckless and often unindicated administration has caused the emergence of multi-resistant and pan-resistant hospital strains. Infections caused by a

nosocomial strain of bacteria endanger the patient directly and are often the cause of death, especially in patients with comorbidities. Furthermore, treatment of these infections prolongs the length of hospital stay, is expensive, and often leads to other complications.

Inanimate surfaces in the hospital environment are one of the most important determinants of nosocomial infections. In up to one-third of cases of nosocomial infections, inanimate surfaces are a source of infection. Colonization of these areas by nosocomial pathogens plays a central role in transmitting the infection to patients. In this respect, it is desirable and important to know

what types of bacteria are found on inanimate surfaces in hospital facilities and their antibiotic resistance level (1).

Staphylococcus epidermidis and *Staphylococcus haemolyticus* are among the most common bacteria in the hospital environment and are increasingly causing nosocomial infections. Genetic studies show that *Staphylococcus epidermidis* strains isolated from the hospital environment differ from community-occurring strains mainly in the terms of biofilm-forming ability, antibiotic resistance, and the presence of mobile DNA elements (2).

The importance of nosocomial isolates of *Staphylococcus epidermidis*, which currently show high genome flexibility, lies in the fact that they are considered reservoirs for the development and spread of mechanisms and signs of resistance in the hospital environment. Gene analysis has shown that the gene responsible for methicillin resistance (*mecA*) is mobile and horizontally transferable to other staphylococcal strains. In a hospital setting, this is an important finding that can cause the formation of methicillin-resistant *Staphylococcus aureus* (MRSA) isolates. That is why we can no longer claim coagulase-negative staphylococci to be non-pathogenic bacteria in the hospital environment. Immuno-compromised and critically ill patients are most often affected by the infection. They are mainly involved in the development of bloodstream infections, often in patients with artificial valve replacements. The penetration of bacteria into the body most often occurs during invasive procedures such as insertion of peripheral and central venous catheters or during various endoscopies (3).

The main reason for the good survival of *enterococci* in the hospital environment is their intrinsic resistance to commonly used antibiotics and their ability to acquire resistance to other available antibiotics, either through mutation or uptake of foreign genetic material by plasmids or transposons. Vancomycin-resistant *enterococci* (VRE) are increasingly becoming problematic strains. Of particular concern is that it is complicated to control their occurrence once present in a hospital facility. In addition, vancomycin (VAN) resistance genes may be transmitted to nosocomial strains of *Staphylococcus aureus*, which further contributes to resistance growth and endangers hospitalized patients. They are most often the cause of urinary tract infections, endocarditis, and intra-abdominal infections (4).

MATERIALS AND METHODS

The Ethics Committee approved the research of L. Pasteur University Hospital on November 28, 2019 (No. 2019/EK/11060).

A total of 182 swabs were collected from the Department of Internal Medicine (DIM) and the Department of Anaesthesiology and Intensive Care (DAIC) using sterile collection kits. The most frequent collection points were bed handles, floors, personal patient tables, door handles, X-ray and ECG devices, etc. In addition, part of the samples was taken from specific inanimate surfaces related to the department's nature, e.g., infusion pumps, ventilators, etc. The samples were placed in appropriate sterile containers, labelled, and quickly transported to the microbiological laboratory in cooperation with the hospital hygienist. In the laboratory, all collected swabs were inoculated on sterile culture media (blood agar containing 5% ram erythrocytes) and cultured in a thermostat at constant temperature 37 °C under aerobic conditions for a total of 24–48 hours. After the cultivation period, the

growth of bacteria on individual blood agar was evaluated separately for each swab – absent bacterial colonies (soils remained sterile), 1 bacterial colony – pure bacterial culture, 2 or more bacterial colonies – mixed bacterial culture. The mixed bacterial cultures were inoculated again on additional sterile blood agars and re-cultured under the same laboratory conditions for 24–48 hours to isolate and obtain pure bacterial cultures. Pure bacterial cultures were identified on a MALDI TOF MS. The preparation, identification of the sample, and subsequent evaluation of the identification always followed the exact procedure of the instrument manufacturer. The sample preparation method was performed according to the instructions of the German manufacturer BrukerDaltonics. The selected bacterial colony was applied to a target plate using a sterile bacteriological loop and allowed to dry sufficiently at room temperature for 5–10 minutes. Each sample was applied in a duplicate manner to increase the success of the identification. After drying, the sample was added dropwise with 1 µl of the matrix (cinnamic acid). It was allowed to dry again at room temperature for 5–10 minutes.

The target with the prepared bacterial samples was placed in a MALDI instrument. After successful calibration, the process of identifying bacteria using the MALDI Biotyper 3.0 software system was started.

Nosocomial significant *staphylococci* and all types of *enterococci* were excluded from the group of successfully identified bacteria and tested for susceptibility to selected types of antibiotics. Isolates of *Staphylococcus aureus*, *Staphylococcus epidermidis*, and *Staphylococcus haemolyticus* were tested for sensitivity to oxacillin (OXA) and ceftioxin (FOX). Sensitivity to OXA also means sensitivity to methicillin (the same group of antibiotics). VAN susceptibility has been determined in gram-positive cocci of the genus *Enterococcus* (*faecalis*, *faecium*) and *Staphylococcus aureus* strains.

Pearson's chi-square test was the most frequently used statistical method for data processing used to determine the difference between two examined files within one monitored characteristic, e.g., occurrence of resistance to OXA and FOX between two groups of pathogens.

RESULTS

A total of 182 swabs were collected from both workplaces (DIM 102, DAIC 80 smears). After inoculation on blood agar, all swabs were also placed in a liquid medium – broth – for 24 hours. Positive cultivation was recorded in 162 cases (on 162 Petri dishes or broths). In 20 cases, the cultivation media remained sterile after inoculation. The results of cultivation in both departments confirmed 10.1% of sterile plates (20 sterile plates) (Fig. 1). At the DIM, positive culture was recorded in 99 of 102 smears (97.05%). Positivity at DAIC was recorded in 63 samples out of 80 smears (78.75%).

A total of 382 pure bacterial colonies were isolated and subsequently identified by MALDI TOF MS. Successfully we identified 377 bacterial cultures, representing 98.7%. We detected 43 different species of bacteria.

The identified bacteria (377) were divided into four basic groups according to Gram staining (Fig. 2) – gram-positive, gram-negative cocci, and gram-positive, gram-negative rods.

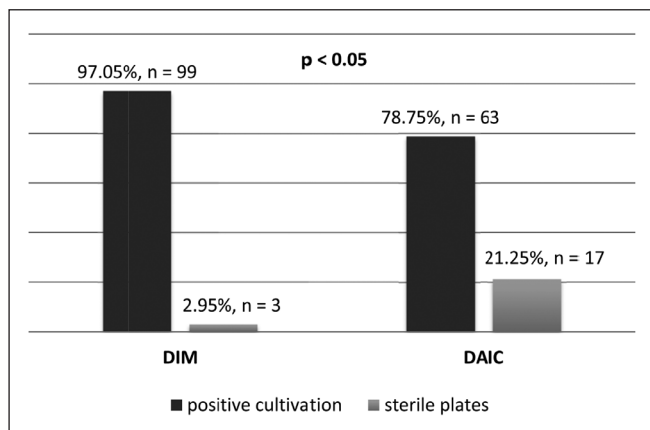


Fig. 1. Comparison of number of sterile plates and positive cultivations on both wards DIM ($n = 102$), DAIC ($n = 80$).

Statistically evaluated by chi-square test

DIM – Department of Internal Medicine; DAIC – Department of Anaesthesiology and Intensive Care

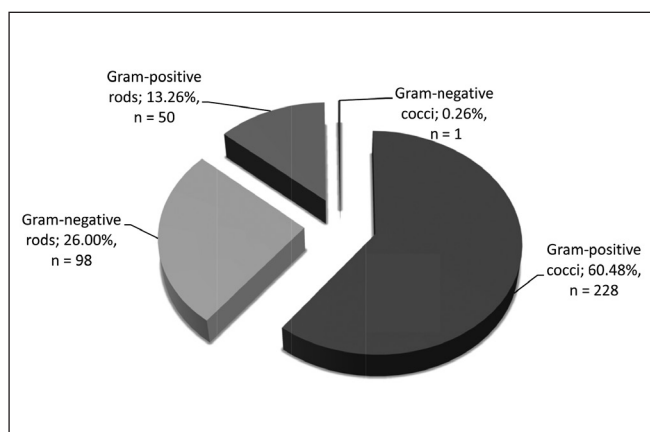


Fig. 2. Distribution of identified bacteria according to Gram staining. ($N = 377$)

Gram-positive cocci (especially from the genus *Staphylococcus* and *Micrococcus*) were the most common on the wards – 228 (60.5%), followed by gram-negative rods – 98 (26.0%), the third most common were gram-positive rods – 50 (13.3%). On the contrary, gram-negative cocci occurred very sporadically. Only one species was captured – *Neisseria subflava*, which was isolated from DAIC.

Of the total number of identified bacteria (377), 156 strains belonged to the genus *Staphylococcus*. In addition, 15 enterococcal strains were also isolated and identified (Table 1). The most common strains were *Staphylococcus epidermidis* and *Staphylococcus hominis*. Genus *Enterococcus* was more frequent at DAIC department (9 strains) comparing to DIM (6 strains). Genus *Staphylococcus* had higher prevalence (84 strains) at DIM versus 72 strains at DAIC (Table 1).

Oxacillin-resistant strains of *Staphylococcus epidermidis* ($n=45$) were the most common in both compartments (Table 2, Fig. 3). At both departments OXA resistant were 36 strain (80%), OXA sensitive were 20%. The table and graph also shows that in total 26 (57.8%) strains were resistant to FOX and 19 (42.2%) were FOX sensitive. Using the Chi-square test, it was possible

Table 1. Frequency of selected staphylococcal and enterococcal strains at both departments ($N = 156$)

Identified strain	DIM	DAIC	Overall
<i>Enterococcus faecalis</i>	4	4	8
<i>Enterococcus faecium</i>	2	5	7
Total <i>Enterococcus</i>	6	9	15
<i>Staphylococcus aureus</i>	2	0	2
<i>Staphylococcus capitis</i>	19	15	34
<i>Staphylococcus epidermidis</i>	15	30	45
<i>Staphylococcus haemolyticus</i>	13	7	20
<i>Staphylococcus hominis</i>	26	19	45
<i>Staphylococcus pasteurii</i>	2	0	2
<i>Staphylococcus saprophyticus</i>	1	0	1
<i>Staphylococcus simulans</i>	3	0	3
<i>Staphylococcus warneri</i>	3	1	4
Total <i>Staphylococcus</i>	84	72	156

DIM – Department of Internal Medicine; DAIC – Department of Anaesthesiology and Intensive Care

Table 2. Comparison of resistance and sensitivity of *Staphylococcus epidermidis* to oxacillin and cefoxitin – DIM vs. DAIC

Identified <i>S. epidermidis</i>	DIM n (%)	DAIC n (%)	Total R/S n (%)	p-value
OXA sensitive	7 (77.8)	2 (22.2)	9 (20.0)	0.0015**
OXA resistant	8 (22.2)	28 (77.8)	36 (80.0)	
Total DIM/DAIC	15 (33.3)	30 (66.7)	45 (100.0)	
FOX sensitive	11 (57.9)	8 (42.1)	19 (42.2)	0.0028**
FOX resistant	4 (15.4)	22 (84.6)	26 (57.8)	
Total DIM/DAIC	15 (33.3)	30 (66.7)	45 (100.0)	

R – resistance; S – sensitivity; OXA – oxacillin; FOX – cefoxitin; DIM – Department of Internal Medicine; DAIC – Department of Anaesthesiology and Intensive Care; ** $p < 0.01$

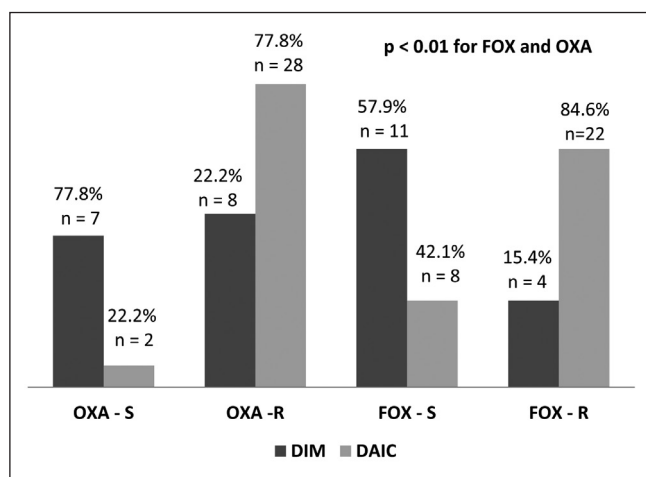


Fig. 3. Resistance of *Staphylococcus epidermidis* to oxacillin and cefoxitin.

OXA – oxacillin; FOX – cefoxitin; DIM – Department of Internal Medicine; DAIC – Department of Anaesthesiology and Intensive Care

Table 3. Comparison of resistance and sensitivity of *Enterococci* to vancomycin – DIM vs. DAIC

Identified <i>Enterococcus faecalis</i>	Department of Internal Medicine n (%)	Department of Anaesthesiology and Intensive Care n (%)	Total n (%)	p-value
VAN sensitive	2 (50.0)	2 (50.0)	4 (50.0)	n.s.
VAN resistant	2 (50.0)	2 (50.0)	4 (50.0)	
Total DIM/DAIC	4 (50.0)	4 (50.0)	8 (100)	
VAN sensitive	1 (100.0)	0 (0.0)	1 (100.0)	n.s.
VAN resistant	1 (16.7)	5 (83.3)	6 (57.8)	
Total DIM/DAIC	2 (28.6)	5 (71.4)	7 (100)	

VAN – vancomycin; n.s. – not statistically significant

Table 4. Comparison of resistance and sensitivity of *Staphylococcus haemolyticus* to oxacillin and ceftioxin – DIM vs. DAIC

Identified <i>S. haemolyticus</i>	Department of Internal Medicine n (%)	Department of Anaesthesiology and Intensive Care n (%)	Total R/S n (%)	p-value
OXA sensitive	6 (60.0)	4 (40.0)	10 (50.0)	n.s.
OXA resistant	7 (70.0)	3 (30.0)	10 (50.0)	
Total DIM/DAIC	13 (65.0)	7 (35.0)	20 (100.0)	
FOX sensitive	7 (63.6)	4 (36.4)	11 (55.0)	n.s.
FOX resistant	6 (66.7)	3 (33.3)	9 (45.0)	
Total DIM/DAIC	13 (65.0)	7 (35.0)	20 (100.0)	

R – resistance; S – sensitivity; OXA – oxacillin; FOX – ceftioxin; n.s. – not statistically significant

to demonstrate that there is a statistically significant difference between oxacillin- and ceftioxin-resistant strains of *Staphylococcus epidermidis* and susceptible strains between DAIC and DIM. Oxacillin resistant strains of *Staphylococcus epidermidis* were statistically higher ($p < 0.01$) at ward DAIC (77.8%) comparing to DIM (22.2%). Ceftioxin resistant strains of *Staphylococcus epidermidis* were also significantly higher at DAIC comparing to DIM (84.6% vs. 15.4%; $p < 0.01$). *Staphylococcus epidermidis* strains were resistant to oxacillin and thus belonged to methicillin-resistant (MRSH).

Vancomycin susceptibility was assessed in all isolated strains of *Enterococcus faecalis* and *Enterococcus faecium*. Eight strains of *Enterococcus faecalis* were also isolated; 4 isolated strains (50%) were resistant to vancomycin (2 from DAIC, 2 from DIM), 4 strains were sensitive to vancomycin. In the case of *Enterococcus faecium*, 7 strains were isolated, 6 of which showed resistance to vancomycin (5 strains were from DAIC, 1 strain from DIM). Despite the fact that *Enterococcus faecium* resistance to VAN was higher in DAIC, Fisher's test did not confirm statistical significance due to extremely small amount of examination (Table 3).

Total of 20 *Staphylococcus haemolyticus* (Table 4) strains were identified (7 from DAIC, 13 from DIM). Ten strains (50.0%) were resistant to oxacillin and thus belonged to methicillin-resistant. In addition, 9 strains (45.0%) were resistant to ceftioxin. The resistance of *Staphylococcus haemolyticus* to both OXA and FOX was higher at DIM but we did not confirm statistical significance due to small amount of isolated strains.

Staphylococcus aureus – 2 strains (Table 1) were isolated from DIM. This pathogen was not present at DAIC. Both isolated strains

were sensitive to oxacillin and ceftioxin.

DISCUSSION

Despite the rapid evolution of modern medicine, nosocomial infections are still current and often discussed problem. They pose a threat not only to hospitalized patients but also to hospital staff. The ever-increasing resistance of nosocomial pathogens, which is often associated with unindicated antibiotic administration, has reached enormous proportions today. Multi-resistant or even pan-resistant strains of nosocomial bacteria are becoming a problem in the hospital environment. The literature shows that inanimate surfaces are the source of up to 1/3 of all nosocomial infections (5).

Many other studies have shown that inanimate hospital surfaces are an essential factor in the process of spreading important nosocomial pathogens to patients. These are, in particular, coagulase-negative staphylococci, *Staphylococcus aureus*, including MRSA, enterococci, gram-negative rods (especially *Enterobacteriaceae*), which can be easily isolated from inanimate surfaces in the vicinity of colonized or infected patients. These microorganisms can survive in the hospital environment ranging from hours to days (in some cases up to months). Their dissemination in the hospital setting is facilitated by healthcare professionals as well as patients (6).

For this reason, we focused on detecting the presence of nosocomial isolates of the genus *Enterococcus* and *Staphylococcus* on various inanimate surfaces of two selected departments DIM and DIAC, University Hospital of L. Pasteur, Košice (UHLP). The reason for choosing these workplaces is the assumption of

the highest incidence of highly resistant, nosocomial pathogens. Moreover, these departments report the highest incidence of nosocomial infections among clinics and departments of UHLP and the highest administration rate of different antibiotics.

Identification of bacteria isolated from swabs from inanimate surfaces was performed by an innovative microbiological method based on mass spectrometry, using a MALDI TOF MS instrument. A total of 382 pure bacterial cultures were identified and isolated from the inanimate surfaces of both compartments. Using MALDI TOF MS, 377 bacterial strains were successfully identified, representing identification success rate of 98.7%. Furthermore, Wang et al. report the success of identifying isolated bacterial strains by the MALDI method at the level of 95.5% (7).

Bizzini et al. isolated and cultured successfully 97% of all bacteria using the MALDI method (8).

Gram-positive cocci most often occurred on the inanimate surfaces of both wards. These were mainly non-pathogenic strains of *Micrococcus luteus* and coagulase-negative staphylococci, particularly *Staphylococcus epidermidis* and *Staphylococcus haemolyticus*. Gram-positive cocci accounted for up to 60.5% (n=228) of all successfully identified bacteria.

Similarly, Akbari et al. report that the most common bacteria on inanimate hospital surfaces are coagulase-negative staphylococci (*Staphylococcus epidermidis*, *Staphylococcus haemolyticus*). Most nosocomial infections caused by coagulase-negative staphylococci are known for their ability to survive on inanimate, especially dry surfaces due to their ability to form a biofilm (5).

Similar results were obtained by the research of Róžańska et al., which focused on detecting bacterial pathogens on inanimate surfaces of several hospitals in Poland using the MALDI TOF MS method (9). In all hospitals covered by the research, coagulase-negative staphylococci were identified (especially *Staphylococcus epidermidis*, *Staphylococcus haemolyticus*, *Staphylococcus hominis*, *Staphylococcus pettenkoferi*, *Staphylococcus simulans*, *Staphylococcus warneri*).

In summary, coagulase-negative staphylococci (*Staphylococcus epidermidis*, *Staphylococcus haemolyticus*, *Staphylococcus hominis*, and *Staphylococcus capitis*) were also the most common in both studied UHLP departments.

Other gram-positive bacteria belonging to the nosocomial group identified by Róžańska et al. are coagulase-positive staphylococci, *Staphylococcus aureus*, and enterococci. *Enterococcus faecalis* and *Enterococcus faecium* accounted for about 4.6% of the total number of bacteria isolated. The authors state that *Enterococcus faecium* showed more frequent resistance to vancomycin than *Enterococcus faecalis*, in concordance with our research (9).

The importance of nosocomial isolates of *Staphylococcus epidermidis*, which currently show high genome flexibility, lies in the fact that they are considered as reservoirs for the development and spread of mechanisms and signs of resistance in the hospital environment. Gene analysis has shown that the gene responsible for methicillin resistance is mobile and horizontally transferable to other staphylococcal strains. This gene encodes the synthesis of the penicillin-binding protein PBP2a. In a hospital setting, this is an important finding that can cause MRSA isolates. That is why we can no longer say that coagulase-negative staphylococci belong to non-pathogenic bacteria in the hospital environment. Immunocompromised and critically ill patients are most often affected by these pathogens. They are mainly involved in the

development of bloodstream infections, often in patients with artificial valve replacements. The penetration of bacteria into the body most often occurs during invasive procedures such as insertion of peripheral and central venous catheters, or during various endoscopies (10).

Götz et al. state that most nosocomial strains of coagulase-negative staphylococci isolated from inanimate hospital surfaces are resistant to methicillin (11). From the group of coagulase-negative staphylococci, strains of *Staphylococcus epidermidis*, which showed resistance to methicillin (MRSE), were the most common in both departments in our study – 36 strains out of 45 (80%). Most MRSE isolates (n=28) were from DAIC. The increased incidence of MRSE isolates at DAIC is probably due to critically ill patients having several invasive inputs (peripheral and central venous catheters, permanent urinary catheters, nasogastric tubes, chest drainage, etc.).

It is well known that most nosocomial strains of *Staphylococcus haemolyticus* do not have significant virulence attributes. Some enzymes, cytolytins, or surfactants are indicated in the literature as factors that may contribute to virulence but none of them has been identified as determining virulence factors, yet. Nevertheless, *Staphylococcus haemolyticus* is the second, most frequently isolated pathogen from patients, especially in patients with central nervous system infections, wound infections and bloodstream infections (12).

We managed to isolate a total of 20 strains of *Staphylococcus haemolyticus* from the inanimate surfaces of both departments. In contrast to *Staphylococcus epidermidis*, most *Staphylococcus haemolyticus* isolates were from DIM – 15 (75%), 5 strains (25%) were found on DAIC surfaces. Of the 15 strains detected on inanimate surfaces at DIM, 9 showed resistance to methicillin (60%). According to the results of Dziri et al., up to 55% of *Staphylococcus haemolyticus* isolates isolated from inanimate hospital surfaces are resistant to methicillin (13). They also describe the relatively frequent occurrence of *Bacillus* and *Corynebacterium* species on inanimate hospital surfaces. On the contrary, the results of the study show that the occurrence of *Staphylococcus aureus* on the inanimate surfaces of the examined wards is relatively rare (only 2 findings at DIM). Dziri et al. report that *Staphylococcus aureus* isolates account for 15.7% of all pathogens isolated from inanimate hospital surfaces (13).

The results of Carlos et al. point to the relatively frequent occurrence of enterococci on inanimate surfaces of hospital facilities. Their resistance to environmental conditions allows them to survive in these areas for several weeks. In particular, VRE isolates are appearing more and more frequently (14).

Lalami et al. in their research on the inanimate surfaces of intensive care units, described the frequent occurrence of vancomycin-resistant enterococci, up to 49% of the total number of enterococcal strains identified (15).

In the results of our study, 7 vancomycin-resistant strains of enterococci (2 strains of *Enterococcus faecalis*, 5 strains of *Enterococcus faecium*) out of 11 were identified at DAIC, which represents 63.6%.

Khan et al. found in their research that most VRE recovered from inanimate surfaces are *Enterococcus faecium* strains (16).

VRE strains that were isolated from both examined UHLP sites were mainly *Enterococcus faecium* isolates (n=6). Four VRE isolates were identified as *Enterococcus faecalis*.

It is generally reported that up to 80% of *Enterococcus faecium* strains isolated from the hospital environment are resistant to vancomycin. These are most often intensive care units and departments of anaesthesiology and intensive care medicine (17). All 5 strains (100%) of *Enterococcus faecium* that were isolated from inanimate DAIC surfaces were resistant to vancomycin.

Resistance of nosocomial isolates of *Enterococcus faecalis* to vancomycin is less common compared to *Enterococcus faecium* strains. However, several studies point to the ever-increasing incidence of VRE *Enterococcus faecalis* in the hospital setting, which is mainly transferred by plasmids (18). Al-Sa'ady et al. describe in their publication that 35.7% of isolated *Enterococcus faecalis* strains obtained from patient samples show resistance to vancomycin (19).

CONCLUSION

Nosocomial infections are currently a real threat not only to all hospitalized patients but also to healthcare professionals. Infections acquired in hospital settings complicate the course of hospitalization, prolong the patient's stay in a medical facility, increase the economic costs of care, and often cause patient's death.

The resistance of nosocomial pathogens has become enormous. The occurrence of multi-resistant or even pan-resistant nosocomial strains in hospitals is no longer rare. Improperly empirically administered antibiotics or their administration in unindicated cases often result in the development of nosocomial infections. Inanimate surfaces are considered a reservoir of nosocomial pathogens and are currently thought to be responsible for up to 1/3 of all nosocomial infections.

A total of 182 smears were taken from both workplaces during our research (DIM 102, DAIC 80 smears). After successful cultivation, 212 pure bacterial cultures were isolated from DIM and 170 pure bacterial cultures from DAIC. All isolated pure bacterial cultures were subsequently identified on a MALDI TOF MS; 377 of a total of 382 bacterial strains were successfully identified, representing 98.7%.

In conclusion, it is important to note that nosocomial infections are natural part of every medical facility in the world. Elimination of the risk of nosocomial infection is based mainly on thorough disinfection of all inanimate surfaces and the administration of empirically correct antibiotics in indicated cases. In case that nosocomial infection develops, it is important to report this fact thoroughly to the hospital hygiene service to ensure adequate patient isolation and to prevent the further spread of this infection.

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Conflict of Interests

None declared

REFERENCES

1. European Centre for Disease Prevention and Control. Healthcare-associated infections acquired in intensive care units. Annual epidemiological report for 2017 [Internet]. Stockholm: ECDC; 2019 [cited 2022 Feb 2]. Available from: https://www.ecdc.europa.eu/sites/default/files/documents/AER_for_2017-HAI.pdf.
2. Chabi R, Momtaz H. Virulence factors and antibiotic resistance properties of the staphylococcus epidermidis strains isolated from hospital infections in Ahvaz, Iran. Trop Med Health. 2019 Dec 5;47:56. doi: 10.1186/s41182-019-0180-7.
3. Nguyen TH, Park MD, Otto M. Host response to staphylococcus epidermidis colonization and infections. Front Cell Infect Microbiol. 2017 Mar 21;7:90. doi: 10.3389/fcimb.2017.00090.
4. Cetinkaya Y, Falk P, Mayhall CG. Vancomycin-resistant enterococci. Clin Microbiol Rev. 2000 Oct;13(4):686-707.
5. Akbari R, Fattahi Bafghi M, Fazeli H. Nosocomial infections pathogens isolated from hospital personnel, hospital environment and devices. J Med Bacteriol. 2018;7(1/2):22-30.
6. La Fauci V, Riso R, Facciola A, Merlina V, Squeri R. Surveillance of microbiological contamination and correct use of protective lead garments. Ann Ig. 2016 Sep-Oct;28(5):360-6.
7. Wang L, Zhou KH, Chen W, Yu Y, Feng SF. Epidemiology and risk factors for nosocomial infection in the respiratory intensive care unit of a teaching hospital in China: A prospective surveillance during 2013 and 2015. BMC Infect Dis. 2019 Feb 12;19(1):145. doi: 10.1186/s12879-019-3772-2.
8. Bizzini A, Durussel C, Bille J, Greub G, Prod'homme G. Performance of matrix-assisted laser desorption ionization-time of flight mass spectrometry for identification of bacterial strains routinely isolated in a clinical microbiology laboratory. J Clin Microbiol. 2010 May;48(5):1549-54.
9. Różańska A, Romaniszyn D, Chmielarczyk A, Bulanda M. Bacteria contamination of touch surfaces in Polish hospital wards. Med Pr. 2017 Jun 27;68(4):459-67.
10. Dortet L, Glaser P, Kassis-Chikhani N, Girlich D, Ichai P, Boudon M, et al. Long-lasting successful dissemination of resistance to oxazolidinones in MDR staphylococcus epidermidis clinical isolates in a tertiary care hospital in France. J Antimicrob Chemother. 2018 Jan 1;73(1):41-51.
11. Götz F, Perconti S, Popella P, Werner R, Schlag M. Epidermin and gallidermin: Staphylococcal lantibiotics. Int J Med Microbiol. 2014 Jan;304(1):63-71.
12. Ruzauskas M, Siugzdiniene R, Klimiene I, Virgailis M, Mockeliunas R, Vaskeviciute L, et al. Prevalence of methicillin-resistant Staphylococcus haemolyticus in companion animals: a cross-sectional study. Ann Clin Microbiol Antimicrob. 2014 Nov 28;13:56. doi: 10.1186/s12941-014-0056-y.
13. Dziri R, Klibi N, Lozano C, Ben Said L, Bellaaj R, Tenorio C, et al. High prevalence of Staphylococcus haemolyticus and Staphylococcus saprophyticus in environmental samples of a Tunisian hospital. Diagn Microbiol Infect Dis. 2016 Jun;85(2):136-40.
14. Correa-Martinez CL, Tönnies H, Froböse NJ, Mellmann A, Kampmeier S. Transmission of vancomycin-resistant enterococci in the hospital setting: uncovering the patient-environment interplay. Microorganisms. 2020 Jan 31;8(2):203. doi: 10.3390/microorganisms8020203.
15. El Ouali Lalami A, Touijer H, El-Akhal F, Ettayebi M, Benchemsi N, Maniar S, et al. Microbiological monitoring of environment surfaces in a hospital in Fez city, Morocco. J Mater Environ Sci. 2016;7(1):123-30.
16. Khan HA, Ahmad A, Mehboob R. Nosocomial infections and their control strategies. Asian Pac J Trop Biomed. 2015;5(7):509-14.
17. Fiore E, Van Tyne D, Gilmore MS. Pathogenicity of enterococci. Microbiol Spectr. 2019 Jul;7(4):10.1128/microbiolspec.GPP3-0053-2018. doi: 10.1128/microbiolspec.GPP3-0053-2018.
18. Davis E, Hicks L, Ali I, Salzman E, Wang J, Snitkin E, et al. Epidemiology of vancomycin-resistant Enterococcus faecium and Enterococcus faecalis colonization in nursing facilities. Open Forum Infect Dis. 2020 Jan 6;7(1):ofz553. doi: 10.1093/ofid/ofz553.
19. Al Sa'ady AT, et al. Detection of vancomycin resistance in multidrug-resistant Enterococcus faecalis isolated from burn infections. 2019. In Drug Invent Today. 2019;11(11):2984-9.

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