An evaluation of frequencies and types of bacterial pathogens in personnel gowns, before and after washing with sodium hypochlorite 5%, in Vali-e-Asr Hospital, Birjand, Iran

Asma Maleki, Marzieh Shamshirgaran, Hamed Aramjoo, Morteza HajiHosseini, Hakimeh Malaki Moghadam, Majid Zare Bidaki

1 MSc student of Hematology, Faculty of Paramedicine, Tehran University of Medical Sciences, Tehran, Iran
2 BSc in Laboratory Science Technology, Faculty of Paramedicine, Birjand University of Medical Sciences, Birjand, Iran
3 PhD candidate of Epidemiology, Alberta University, Edmonton, Canada
4 MSc in Biostatistics, Clinical Research Development Unit of Vali-e-Asr Hospital, Birjand, Iran
5 Associate Professor in Microbiology, Infectious Disease Research Center, Birjand University of Medical Sciences, Birjand, Iran

Received: April 26, 2019 Revised: October 14, 2019 Accepted: October 14, 2019

Abstract

Introduction: Microbial contamination of hospital gowns is the main reason for the dissemination of nosocomial infections. Bacteria are the cause of most of these contaminations. The present study aimed at determining the frequencies and types of bacterial pathogens on the surface of personnel gowns, before and after washing with sodium hypochlorite 5% in Vali-e-Asr Hospital, Birjand, Iran.

Methods: In the current study, the sampling was carried out in two steps, before and after washing with sodium hypochlorite. Sampling was performed with a sterile swap in the transport medium. Samples were incubated for 3 h and were cultured on blood agar and eosin methylene blue agar. After overnight incubation and colonization, the identification was conducted using macroscopic, microscopic, rapid enzymatic, and biochemical tests.

Results: The most common bacterium before washing was Pseudomonas aeruginosa and the least common was Enterobacter cloacae. It was revealed that sodium hypochlorite exert the most profound effect on Pseudomonas aeruginosa, whereas the least effect was detected on Staphylococcus aureus and Staphylococcus epidermidis.

Conclusions: The type of bacteria involved in nosocomial infections could be different among hospitals. Sodium hypochlorite 5% is recommended for washing hospital gowns to reduce bacterial contamination.

Key words: Disinfection, Nosocomial infections, Sodium hypochlorite

Introduction

Infections that are present neither at the time of admission nor during the incubation period; however, they occur during 48-72 hours after hospitalization or during 10-30 days after discharge from the hospital are classified as nosocomial infections (1, 2). Hospital is where microorganisms
can be transmitted from one patient to another in different ways. In such environments, some pathogenic or opportunistic microorganisms can be carried by care personnel or transmitted from surfaces that are in direct contact with patients.

Various studies carried out at national and international levels reported that microbial contamination of medical equipment in hospitals may transmit pathogens to patients (3-7). According to studies conducted by World Health Organization in 14 countries, the prevalence of nosocomial infections was 8.7% in North America, 5% in Europe, and 40% in Asia, Latin America, and Africa (8, 9). The high costs of nosocomial infections are imposed upon the treatment systems and patients. Hospital pathogens are usually more pathogenic and drastically differ from non-nosocomial pathogens.

These bacteria have specific qualities (e.g., multiple resistance to antibiotics caused by antibiotic-resistant genes exchanges in hospital) which might be due to exposure to different types of antibiotics and disinfectants used in hospitals. Mechanisms that cause bacteria to resist antibiotics include antibiotic degradation, prevention of antibiotic penetration due to purine deficiency, or intracellular antibiotics excretion (e.g., efflux pumps) (10).

Although nosocomial infections can affect different body parts, most reports are from urinary tract infections, lower respiratory tract infections, and surgical wounds (2) from past to present methods, such as using various disinfectants have been common in hospitals to prevent nosocomial infections. However, because of the inappropriate use of them, the desired result has not been achieved (11). Nonetheless, Yousefi Mashouf et al. used sodium hypochlorite as a disinfectant for cleaning slit lamp in ophthalmology departments, as well as chlorhexidine 1% as a suitable disinfectant in hospitals (6, 12).

Nosocomial infections certainly have adverse effects on the recovery period and impose extra expenses on patients and hospitals; therefore, the role of disinfectants and antiseptics are of great importance. The aim of the present study was to determine the frequencies and types of pathogens on the surface of personnel gowns, before and after washing with sodium hypochlorite 5% in Vali-e-Asr Hospital, Birjand, Iran.

**Methods**

This descriptive and analytical study was approved by the Ethics Committee and Deputy of Research and Technology of Birjand University of Medical Sciences (Ethics code: ir.bums.rec.1396.182), Birjand, Iran. In addition, the current research was a field study. A simple random sample method was selected based on the conditions of Vali-e-Asr Hospital in Birjand. Sampling was performed by a wet sterile swab in 2018. The sampling was conducted in two stages, before and after washing with sodium hypochlorite. A total of 132 gowns were randomly selected, containing a total of 528 microbial cultures, including two samples from two different parts (1.cuffs 2. front and bottom of the gown) during two sampling stages (before and after washing). Samples were obtained from the surface of both parts of the gown using a wet swab smeared with Tryptic Soy Broth (TSB).

Then each swab was placed in a test tube containing 0.5ccs of TSB liquid medium. After sampling, the tubes containing the samples were immediately transferred to a medical diagnostic laboratory, then the inoculated culture tubes were placed in a 37°C incubator for 4 h. Samples were cultured on blood agar and eosin methylene blue agar. Then plates were examined after 24 h. In the presence of bacterial colonies on the surface of each culture medium, microscopic, and biochemical features of the separated microorganism were evaluated for bacterial identification.

In order to identify the isolated bacteria, we evaluated the isolates based on some macroscopic, microscopic, and biochemical criteria. The morphology of the colonies, Gram staining, microscopic observation of bacterial shape and arrangement, motility, type of hemolysis, production of hydrogen sulfide, urease, oxidase and catalase tests, and finally growth on differential, specific, and enriched media (e.g., MacConkey agar, DNase agar, and Chocolate agar) were the main criteria which were used.

To identify gram-positive and catalase-positive cocci, the following tests were implemented: oxidase, catalase, urease, coagulase tests, resistance to bacitracin, and mannitol fermentation. Moreover, to identify gram-positive and catalase-negative cocci, the following tests were applied as well: sensitivity to vancomycin, Bacitracin, optochin, colony morphology, beta hemolysis test, and growing in bile esculine agar test. The obtained results were analyzed in SPSS software (version 16) using the McNemar’s test for comparing data. A p-value of less than 0.05 was considered statistically significant.

**Results**

The results of the present study showed that
Table 1: Effect of sodium hypochlorite on different types of bacteria

| Name of the bacterium | Frequency of bacteria before washing N(%) | Frequency of bacteria after washing N(%) | P-value |
|-----------------------|------------------------------------------|-----------------------------------------|---------|
| Pseudomonas aeruginosa | 90(34.09)                                 | 74(29.66)                               |         |
| Escherichia coli       | 43(16.28)                                 | 13(9.21)                                |         |
| Staphylococcus aureus  | 32(12.12)                                 | 45(31.91)                               |         |
| Staphylococcus epidermidis | 24(9.09)                       | 49(34.75)                               |         |
| Citrobacter freundii   | 18(6.81)                                  | 2(1.41)                                 | <0.001  |
| Klebsiella pneumoniae  | 16(6.06)                                  | 1(0.07)                                 |         |
| Gram-positive bacilli  | 13(4.92)                                  | 23(16.31)                               |         |
| Candida albicans       | 9(3.4)                                    | 1(0.7)                                  |         |
| Acinetobacter          | 7(2.65)                                   | 0(0.0)                                  |         |
| Enterobacter cloacae   | 2(0.75)                                   | 0(0.0)                                  |         |
| Negatives              | 10(3.78)                                  | 0(0.0)                                  |         |
| total                  | 264                                       | 141                                     |         |

out of 132 pre-washed cases, 128 cases were at least contaminated with one microorganism (97.96%). The number of contaminated cases were reduced to 46.59% using sodium hypochlorite (Table 1). The three bacterial species, including Pseudomonas aeruginosa, Escherichia coli, and Staphylococcus aureus accounted for 62% out of 264 bacteria, which were found from the gowns before washing (62.09%, 16.28%, and 12.12%, respectively). Although the remaining 38% of the detected bacteria were related to other types of bacteria, no bacterium type had 10% share (Table 1).

The obtained findings showed that washing with sodium hypochlorite significantly reduced the number of positive cultures to 141 (53%) (P<0.001). The McNemar’s test results also revealed that the sodium hypochlorite did not exert equal effects on different types of bacteria. Even the number of Staphylococcus aureus, Staphylococcus epidermidis, and gram-positive bacilli had been increased. Moreover, Sodium hypochlorite 5% showed a reduction in other bacteria types. Accordingly, the most lethal effect of sodium hypochlorite 5% was observed on Pseudomonas aeruginosa and Escherichia coli.

**Discussion**

Nosocomial infections as a significant concern of the medical community are known to be one of the most critical risk factors in hospitals. Infection control is one of the essential issues associated with the health of staff, patients, and their companions. Many studies have been conducted on nosocomial infection rates and methods of disinfecting hospital surface/equipment at national and international levels.

Our results showed that almost all of the pre-washed gowns were contaminated (96.97%). The highest amount belonged to Pseudomonas aeruginosa (34.09%) and the lowest to Enterobacter cloacae (0.75%). The results of a study carried out by Akbari et al. (2002-2008) indicated that almost 100% of hospital gowns were positive when they were examined at least once or three times, and the most isolated types were Enterobacter species (i.e., 49.27% of isolated bacteria), and the most contaminated gowns belonged to emergency departments and outpatient operating rooms (7). The results of this study were in line with the findings of the present study in terms of the number of positive gowns; however, the isolated bacteria percentage and types are different that is probably due to differences in sampling methods and selected hospital wards.

In another study conducted by Valian et al. (2013) in the School of Dentistry of Shahid Beheshti University of Medical Sciences in Tehran, Iran, the students’ gowns in the therapeutic ward of the school were examined for the type and extent of aerobic bacterial contaminations. Their results indicated that on average 86.7% of the samples had increased post-work contaminations; the most contamination was from gram-negative and Staphylococcus aureus (13). In addition, the aforementioned research was different from the current study in terms of the type of pathogens. Furthermore, in a study conducted by Yousefi Mashouf et al. (2006) in Teaching hospitals of Hamadan showed that the most common bacteria were Escherichia coli, Staphylococcus epidermidis, Micrococcus, Bacillus subtilis and Pseudomonas aeruginosa, respectively (14). Their results confirmed the findings of our study that Pseudomonas aeruginosa is a major pathogen in the hospital staff’s gowns after washing. According to
the available studies, the differences in pathogen types indicate that the pathogenic microorganisms vary from hospital to hospital and from one city to another.

Results of the current study revealed that the contamination rate before washing with sodium hypochlorite was 96.97% and after washing with this disinfectant was reduced to 46.59%. Based on the obtained findings, it can be concluded that sodium hypochlorite had an acceptable disinfection effect. It had the most positive impact on *Pseudomonas aeruginosa*, and the most detrimental effect on *Staphylococcus aureus* and *Staphylococcus epidermidis*. Kohler, AT et al. (2018) carried out a study on the potency of sodium hypochlorite on several *gram-negative* bacteria, including *Pseudomonas aeruginosa*. In this study, *Pseudomonas aeruginosa* showed little sensitivity to sodium hypochlorite (P=0.0025), and no significant difference in sensitivity between different strains of *Pseudomonas aeruginosa* was demonstrated (P>0.05) (15).

Another study by Lineback, CB et al. (2018) was carried out on the effectiveness of sodium hypochlorite and hydrogen peroxide on *Staphylococcus aureus* (ATCC-6538) and *Pseudomonas aeruginosa* (ATCC-15442). The results signified that sodium hypochlorite and the hydrogen peroxide have relatively significant antibacterial effects, compared to ammonium chloride-based disinfectants on *Pseudomonas aeruginosa* and *Staphylococcus aureus* (16). On the other hand, Yousefi Mashouf reported that the effect of sodium hypochlorite was relatively high and moderate on *Staphylococcus epidermidis* and *Pseudomonas aeruginosa*, respectively (14). Almatroudi et al. investigated the effect of hypochlorite on *Staphylococcus aureus* biofilms. They found that hypochlorite concentration and incubation time were among the practical factors for removing *Staphylococcus aureus* (17).

In another study performed by Ujimine, sodium hypochlorite concentration was mentioned as one of the critical factors in removing *Staphylococcus aureus* and the sufficient levels were reported from 30 to 120ppm (18). Chlorine and its derivatives, such as sodium hypochlorite, are widely used to disinfect hospital surfaces and clothing. This disinfectant will demonstrate the best results if the standard tips, including proper dilution, enough time to contact the surfaces to perform disinfection mechanisms, are taken into account. Following some practical instructions, such as collecting gowns from different sections, disinfecting surfaces regularly and timely, drying the clothes, and setting shorter time for washing hospital personnel's clothes can have a significant impact on reducing infections and this is a massive success for both the patient and the hospital.

As it was evident in the obtained results, sodium hypochlorite has a significant effect on *Pseudomonas aeruginosa*. *Pseudomonas aeruginosa* is one of the most resistant bacteria in nosocomial infections (7) and always has severe consequences for patients, as well as it can cause problems for physicians in the process of healing. As a result, sodium hypochlorite was observed to be a good option to disinfect hospital clothing. It should be noted that *Staphylococcus epidermidis* is a microbe and can be transmitted at any stage of the washing process. Therefore, the mortality rates and medical expenditures would be mitigated using new methods of disinfection and modern washing technology.

**Conclusions**

According to the results of the present study, sodium hypochlorite had a favorable effect on hospital disinfection and the most influence on *Pseudomonas aeruginosa* as one of the leading causes of nosocomial infections.

**Acknowledgments**

Authors would like to extend their gratitude to the staff of different departments of Vali_e_Asr Hospital, in particular the sterilization department for their sincere cooperation in carrying out the present study. In addition, the deepest appreciations go to M. Hosseinzadeh for contributing to the current project.

**Funding**

The present research was the result of an approved project and a student research grant (No. 4525) by the Deputy of Research and Technology of Birjand University of Medical Sciences, Birjand, Iran.

**Conflict of Interest**

The authors declared no conflict of interests.

**References**

1. Khosravi B, Razavi AH. Nosocomial infection. EBNESINA. 2010; 13(1-2):43-51. [Persian]
2. Ghasvandian S. Nosocomial infection in the intensive care unit. Hayat. 2002; 8(1):27-34. [Persian]
3. Jinadatha C, Villamaria FC, Coppin JD, Dale CR, Williams MD, Whitworth R, et al. Interaction of
healthcare worker hands and portable medical equipment: a sequence analysis to show potential transmission opportunities. BMC Infect Dis. 2017; 17(1):800. PMID: 29281998 DOI: 10.1186/s12879-017-2895-6

4. Embil JM, Zhanel GG, Plourde PJ, Hoban D. Scissors: a potential source of nosocomial infection. Infect Control Hosp Epidemiol. 2002; 23(3):147-51. PMID: 11918121 DOI: 10.1086/502026

5. Bernard L, Kereveur A, Durand D, Goton J, Goldstein F, Mainardi JL, et al. Bacterial contamination of hospital physicians' stethoscopes. Infect Control Hosp Epidemiol. 1999; 20(9):626-8. PMID: 10501265 DOI: 10.1086/501686

6. Yousefimashouf R, Esmaeeli R, Ramazani A. The evaluation of bacterial contamination of ophthalmic biomicroscopy apparatus (Slit-lamp) in eye centers of Farshcian Hospital in Hamadan and assessment of the current disinfectants. Pajouhan Sci J. 2014; 12(2):18-27. [Persian]

7. Akbari M, Davoodzadeh M, Tarahi M, Rouzbahani H, Bayat A, Radsari E. Study of different kinds of pathogens isolated from gowns of Shohadaye-Ashayer hospital personnel's during 2002-2003 time period. Pajoh. 2005; 7(25):11-6. [Persian]

8. Ghashghaei A, Behzadifar M, Azari S, Farhadi Z, Bragazzi NL, Behzadifar M, et al. Prevalence of nosocomial infections in Iran: a systematic review and meta-analysis. Med J Islam Repub Iran. 2018; 32:48. PMID: 30159299 DOI: 10.14196/mjiri.3248

9. Bagheri Nejad SB, Allegranzi B, Syed SB, Ellis B, Pittet D. Health-care-associated infection in Africa: a systematic review. Bull World Health Organ. 2011; 89(10):757-65. PMID: 22084514 DOI: 10.2471/BLT.11.086179

10. Xia J, Gao J, Tang W. Nosocomial infection and its molecular mechanisms of antibiotic resistance. Biosci Trends. 2016; 10(1):14-21. PMID: 26877142 DOI: 10.5582/bst.2016.01020

11. Dettenkofer M, Block C. Hospital disinfection: efficacy and safety issues. Curr Opin Infect Dis. 2005; 18(4):320-5. PMID: 15985828

12. Yousefimashouf R, Fallah M, Heidarbarghi Z. Efficacy of the disinfectants and antiseptics used in hospitals. Yafte. 2006; 8(1):45-54. [Persian]

13. Valian A, Moravej Salehi E, Namdari M, Ramezanpour F. Evaluation of the aerobic bacterial contamination of students gowns in restorative and periodontics departments of dental school of Shahid Beheshti University of Medical Sciences in 2013. J Mashhad Dent Sch. 2015; 39(2):181-90. [Persian]

14. Yousefi Mashouf R, Nazari M, Samarghandi M, Shams M. Evaluation of efficacy of the current disinfectants on staphylococcus epidermidis and Pseudomonas aeruginosa isolated from hospitals of Hamadan. J Tabib Shargh. 2006; 8(4):287-96. [Persian]

15. Köhler AT, Rodloff AC, Labahn M, Reinhardt M, Truyen U, Speck S. Efficacy of sodium hypochlorite against multidrug-resistant Gram-negative bacteria. J Hosp Infect. 2018; 100(3):e40-6. PMID: 30026008 DOI: 10.1016/j.jhin.2018.07.017

16. Lineback CB, Nkemngong CA, Wu ST, Li X, Teska PJ, Oliver HF. Hydrogen peroxide and sodium hypochlorite disinfectants are more effective against Staphylococcus aureus and Pseudomonas aeruginosa biofilms than quaternary ammonium compounds. Antimicrob Resist Infect Control. 2018; 7(1):154. PMID: 30568790 DOI: 10.1186/s13756-018-0447-5

17. Almatroudi A, Gosbell IB, Hu H, Jensen SO, Espedido B, Tahir S, et al. Staphylococcus aureus dry-surface biofilms are not killed by sodium hypochlorite: implications for infection control. J Hosp Infect. 2016; 93(3):263-70. PMID: 27140421 DOI: 10.1016/j.jhin.2016.03.020

18. Ujimine S, Tone S, Saito M, Yamada S. Intracellular morphological changes in Staphylococcus aureus induced by treatment with sodium hypochlorite. Med Mol Morphol. 2017; 50(3):178-84. PMID: 28516285 DOI: 10.1007/s00795-017-0159-6