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Research Article

The Quality of Chemical Surface Disinfectants Used to Reduce Microbial Contamination Inside the Emergency Departments in Benghazi Medical Center Hospital - Benghazi City - Libya

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Abstract

Work in Hospital Emergency Departments exposes both emergency ward staff and patients to certain bacteria, viruses and other infectious agents. Samples were collected from various surfaces and equipment in Hospital Emergency Departments (HEDs). In total, 60 swabs were collected at pre- and post-cleaning stages and were cultured and identified using standard microbiological procedures. The type of microorganism, proportion of positive and negative cultures (both before and after cleaning) and all bacterial isolates were tested for in vitro antibiotic disc sensitivity. The results of this study show the presence of pathogenic bacteria in samples collected from Hospital Emergency Departments (HEDs). The types of bacteria isolated from floors, doors, treatment tables, wash basins, anesthetist machines, patient beds, laryngoscopes and operation tables in the emergency department were *Micrococcus laylai* (27.30%), *Rhizobium radiobacter* and *Pseudomonas aeruginosa, Escherichia coli* (13.64%), *Stenotrophomonas maltophilia, Staphylococcus aureus, Globicatella sanguinis* and *Acinetobacter baumannii* (9.10%), *Arcanobacterium pyogenes* and *Bacillus cereus* (4.55%). From our investigation and evaluation of quality of cleaning inside the ER that was done by using questionnaire method, we found reduction in quality to 10.5% during night shift. Various factors that contributed to improper disinfection were shortage of, or poor training of the cleaning staff about the correct cleaning methods and lack of experience of staff in handling various types of chemical disinfectants. The relatively high level of contamination by bacteria in HEDs could also be due to the lack of standard infection control precautions. In terms of effectiveness of disinfectants, no single product can provide optimum performance on all surfaces. In our study, Husky, Dialkyl Quaternary ammonium compound-ACS, was found to be more effective in killing broad spectrum of organisms as compared to Konex, Didecyl dimethyl ammonium chloride- DDAC. A third disinfectant called Surfa'safe was also tested but was rendered ineffective, since no change was observed in bacterial contamination before and after its use on highly contaminated areas.

Keywords: *Microbial Contamination; Emergency Departments; Disinfection; Antimicrobial Resistance; Harmful Biological Agents*

Introduction

The role of surface contamination in the transmission of nosocomial pathogens is being recognized increasingly. For more than 100 years, the inanimate environment in operating rooms (e.g. walls, tables, floors, and equipment surfaces) has been considered a potential source of pathogens that may result in surgical site infections (SSIs). However, the role of contaminated surfaces in pathogen acquisition in this setting is generally considered negligible, as most SSIs are believed to originate from patients' or healthcare workers' flora. The characteristics of HEDs work exposes both emergency ward staff and patients to the presence of dangerous biological agents; however, the level of microbiological risk is not only related to the specifics of HEDs' functioning, but also to the manner in which medical or surgical procedures are performed by health care professionals. Medical procedures when performed in accordance with disinfection standards can ensure safety of both patients and medical personnel. The level of microbiological safety is further enhanced by ensuring the hospital environment is free of microorganisms capable of causing infection. Pathogenic microorganisms in hospital environments, such as HEDs, are located generally on the surfaces of medical devices, as well as on other surfaces that come in direct contact with patients (walls, bed frames, medical device switches, sinks, etc.) and may pose the risk of hospital acquired infections (HAIs). The most common routes of transmission of potential pathogens are direct contact with infected hospital personnel and contact between patients (cross transmission). Improper cleaning of rooms and/or improper disinfection of medical equipment contributes to the spreading of pathogens through the devices routinely used in diagnosis, treatment, and rehabilitation [1]. Bacteriological monitoring of the hospital environment enables detection and differentiation of the colonizing and infective bacterial flora, provides the basis for effective empiric antibiotic therapy, and eradication of the microorganisms from the ward environment [2]. Systematic surveillance of infections, mainly by identifying the etiological agents of HAIs, enables identification of the most commonly found microorganisms posing a threat not only to the patient, but also to the whole hospital environment [3].

Nosocomial infections are a significant cause of morbidity and mortality in hospitalized patients [4]. Every member of the health care team must know about these infections and familiarize themselves with their identification, since prevention may represent the best rationale for avoiding human suffering [5]. The Centre for Disease Control (CDC) defines Healthcare-Associate infections (HAIs) as a "localized or systemic condition resulting from an adverse reaction to the presence of an infectious agent(s) or its toxin(s). There must be no evidence that the infection was present or incubating at the time of admission to the acute care setting." HAIs may be caused by infectious agents from endogenous (body sites) or exogenous sources (patient care personnel, visitors, patient care equipment, medical devices, or the health care environment) [6]. Every year, millions of people across the world suffer from HAIs. HAIs are a wide-ranging concern in the medical field, not only because of morbidity and the possibility of lethal consequences for patients, but also because of extended hospital stays and associated high costs [7-10]. In Europe HAIs cause an extra 16 million days of hospital stay and 37,000 attributable deaths, resulting in approximately € 7 billion associated costs annually. In the USA, around 99,000 deaths were attributed to HAIs in 2002 and associated costs were approximately US$ 6.5 billion in 2004 [10,11]. A Europe-wide point prevalence survey estimated that at least 2.6 million cases of HAIs occur annually in long-term care facilities, in addition to the ECDC's earlier estimate of 1 million patients acquiring HAIs in acute-care hospitals [11]. These infections often have little or no association with the primary reason for the hospital visit, but are a result of poor or inadequate hygiene in the healthcare setting [12]. Healthcare equipment is frequently shared between hospital staff, who may have different hygiene practices. Medical devices (stethoscopes, otoscopes, and thermometers), various objects in hospital environments (telephones and computers), and high contact areas (floors, doors, emergency surfaces, doorknobs, treatment tables, washbasins, anesthetist machines, and operation tables) have been associated with transmission of HAIs [13-23]. Current scientific knowledge suggests that the disinfection of environmental surfaces in modern hospitals is paramount. Furthermore, other simple measures, such as hand hygiene of medical staff, is crucial and most important way to avoid HAIs [24]. Various studies have conducted isolated investigations on the role of stethoscopes, computer keyboards, telephones, floors, doors, emergency surfaces, doorknobs, treatment tables, wash basins, anesthetist machines, and operation table in HAIs [14-18,20,21]. At the time of this study, there is lack of research on the evaluation of all these devices together, in different units of the same hospital, where there should be a specific organizational model and risk factors [13] with regard to HAIs. This study used a random sampling strategy in the HED environment in controlled workplaces (offices) not exposed to contact with harmful biological agents. The samples were taken from hospital surfaces (floors, walls, tables) and irregular, hard to access or frequently used surfaces (treatment tables, washbasins, anesthetist machines, and operation tables, etc.) that pose a risk of transferring pathogenic microorganisms.

Aim of the Study

The aims of the present study were to (i) evaluate the contamination of floors, doors, emergency room surfaces, doorknobs, treatment tables, washbasins, anesthetist machines, patient beds, laryngoscopes and operation tables in the emergency department of Benghazi Medical Center (BMC) hospital before and after the use of a disinfecting technique (DT), (ii) to determine the kind of micro-organisms that can be isolated from these surfaces and instruments used by physicians and nurses working in the emergency department (ER) of BMC, and (iii) to assess whether or not the degree of contamination can be diminished with different cleaning techniques and solutions.

Materials and Methods

Setting: A cross-over study was conducted in Benghazi Medical Center hospital with 1,200 beds in Benghazi - Libya. A variety of hospital environments were chosen to be considered based on their varied characteristics as follows: "emergency unit" and "first aid" which has a high turnover of patients and disinfection of the environment cannot always be pursued effectively, "intensive care" which observes aseptic conditions and most patients are at high risk of developing infections, and "cardiology/hemodynamic" which are medical units and have rooms with more beds than the intensive care unit and are frequented by many visitors. Before the study began, meetings were held between hospital management and the principal researcher. This was necessary to explain the project, establish the necessary contacts and avoid any bias in conducting the study. It was considered important to avoid bias caused by doctors/nurses knowing when the investigation would be run, as this might prompt changes in hygiene. It was also decided that the sampling would be on the same day in each unit, to prevent news of the study circulating and resulting in modified hygienic practices.

To achieve the objectives of this study, a transverse prospective before-after study was designed. The questionnaires were collected from the personnel (physicians and nurses) working in the ER. Physicians and nurses participating in the study were unaware of the real purposes of the study, since the questionnaire was designed to ask questions related to the quality of the disinfectant (Table 1). There were no incentives for any person being involved in this study. The questionnaire was administered from June to August 2019. The procedure of cleaning inside the emergency department was evaluated two times every week at all times (morning, midday and night), the evaluation of cleaners and cleaning methods that involved the recovery room and the operation room in ER plus the general infrastructure of department

No.	Standard	Assessment				Notes
		0	1	2	3	
1	There is a policy and a cleanliness schedule written and announced in the Operations Department.					
2	The general view of the section is clean (floors - wall ceilings).					
3	The cleaners follow the correct methods of cleaning.					
4	The cleaning quality for the weekly operations of the operations department.					
5	The quality of administrative office cleaning, reception area and recovery room.					
6	The cleaners received vaccination against (HBV) or any other recommended vaccinations.					
7	The personal protectors are worn during work and when spills occur within operating theaters.					
8	The bathrooms in the department are clean and have their own tools.					
9	The suitable toiletries are available (contents trolley).					
10	The hygiene tools are for operations department only.					
11	There is a place to store cleaning tools.					
12	All spills (blood, body fluids, etc.) are handled correctly.					
13	Disinfectants are handled well in terms of use (concentration, storage, shelf life).					
Total						
Total summation						
The percentage = (Total ÷ 39) x = 100						

Table 1: *Questions from the questionnaire used to disguise the true purpose of the study: Comprehensive assessment of the level of cleanliness in ER.*

Disinfecting technique

Chemical disinfectants used for cleaning were Konex (Didecyl dimethyl ammonium chloride), Husky (Quaternary ammonium) and Surfa'safe, detergent foam containing DDAC.

Data collection: A cross sectional study was conducted at all Emergency departments in Benghazi Medical Center hospital from 1 July 2019 to 30 August 2019. Data was collected to evaluate the quality of cleaning in ER. The study involved taking swabs to detect prevalence of microorganisms most commonly found in the environment of ER. The surfaces studied were all floors, doors, emergency surfaces, doorknobs, treatment tables, washbasins, anesthetist machines, patient beds, laryngoscopes and operation tables in the emergency department. To evaluate the initial level of contamination, the experimental protocol required a first sample (swab H0) from the floors, doors, emergency surfaces, doorknobs, treatment tables, washbasins, anesthetist machines, patient beds, laryngoscopes and operation tables, before cleaning with the three types of disinfectants. A second sample (swab H1) was taken from all those surfaces and objects after cleaning with each of the disinfectants individually. In total, 60 swabs were collected at pre- and post-cleaning stages, which were sent to the laboratory for culture and identification using standard microbiological procedures.

Laboratory analysis: The type of microorganisms, proportion of positive and negative cultures (both before and after cleaning), percentages of colony reduction in floors, doors, emergency surfaces, doorknobs, treatment tables, washbasins, anesthetist machines, patient beds, laryngoscopes and operation tables after cleaning, and the relative frequencies of cleaning on a regular basis by professional rank were analyzed. Isolates from cultures were identified by standard methods. All bacterial isolates were tested for *in vitro* antibiotic disc sensitivity. The antibiotics used in this study were Ceftazidime, Tetracycline, Piperacillin, Ampicillin, Tobramycin, Flucloxacillin, Colistin, Cefoxitin, Ciprofloxacin, Imipenem, Aztreonam, Levofloxacin, Ciprodex, Cefepime, Cephalexin, Chloramphenicol, Trimethoprim/sulfamethoxazole, Polymyxin, Cefotaxime, Teicoplanin, Ceftriaxone, Clindamycin, Vancomycin, Meropenem, Oxacillin, Doxycycline, Ticarcillin, Penicillin, Fusidic acid, Erythromycin, Nitrofurantoin, Nalidixic acid, Cloxacillin, Augmentin, Gentamicin, and Cefazolin.

Statistical analysis: The data were analyzed by SPSS 18.

Results

Based on the swabs collected from the ER before and after cleaning by first type of disinfectant Didecyldimethylammonium chloride (Konex), figure 1 shows 79% of surfaces tested in Emergency environment were contaminated by various types of microbes, including pathogenic bacteria. The high percentage of contaminated ER surfaces indicate that the quality of cleaning is inadequate and points towards the ineffectiveness of this chemical disinfectant. In addition, findings suggest that percentage index for quality of cleaning was poor. The type of chemical material performed below the desired level and in addition, the cleaning staff was not committed to the infection control instructions provided, specifically for diluting the chemical material. The types of bacteria isolated from floors, doors, emergency surfaces, doorknobs, treatment tables, washbasins, anesthetist machines, patient beds, laryngoscopes and operation tables in the emergency department are presented in table 2 and figure 2. Figure 1 shows the distribution of all microbes both pathogenic and environmental in Emergency department.

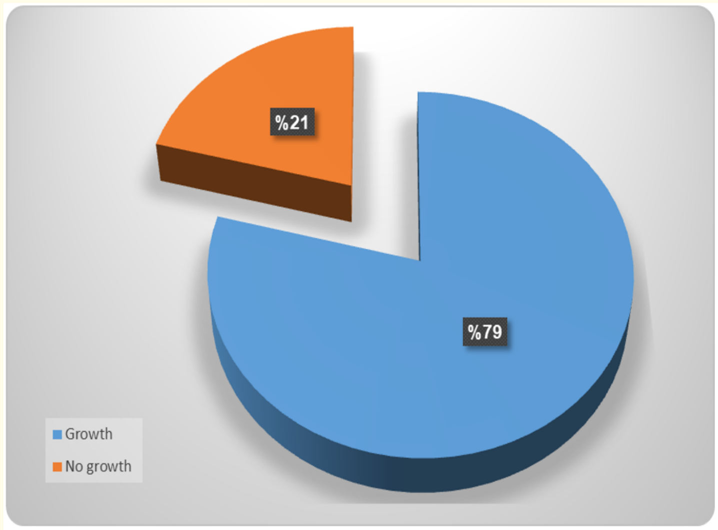

***Figure 1:** Distribution of bacterial contamination in Emergency Department environment in BMC.*

Bacterial growth	Percentage
Micrococcus lylae	27.30%
Rhizobium radiobacter	13.64%
Pseudomonas aeruginosa	13.64%
Escherichia coli	13.64%
Stenotrophomonas maltophilia	9.10%
Staphylococcus aureus	9.10%
Globicatella sanguinis	9.10%
Acinetobacter baumannii	9.10%
Arcanobacterium pyogenes	4.55%
Bacillus cereus	4.55%

***Table 2:** Prevalence of various micro-organisms in the Emergency Department at BMC*

The second type of disinfectant used on high contact surfaces including medical devices was Quaternary ammonium compound (Husky). It has the ability to kill a broad spectrum of microbes, viruses (lipid viruses and non-lipid viruses), and fungi, but is not effective in killing spurs and TB bacteria. Quaternary ammonium compound is safe for health care workers handling it. The dilution method used was 20 ml of Husky in one litre of water (1oz. per gallon of water) and once mixed, this compound has a shelf life of 14 days. Figure 3 and 4 show the level of contamination on ER surfaces before and after disinfection using Quaternary ammonium compound. In comparison with the results of the first chemical material, Didecyl dimethyl ammonium chloride, which are discussed above, figure 3 and 4 show that Quaternary ammonium is more effective in killing various pathogens that are typically found on ER surfaces.

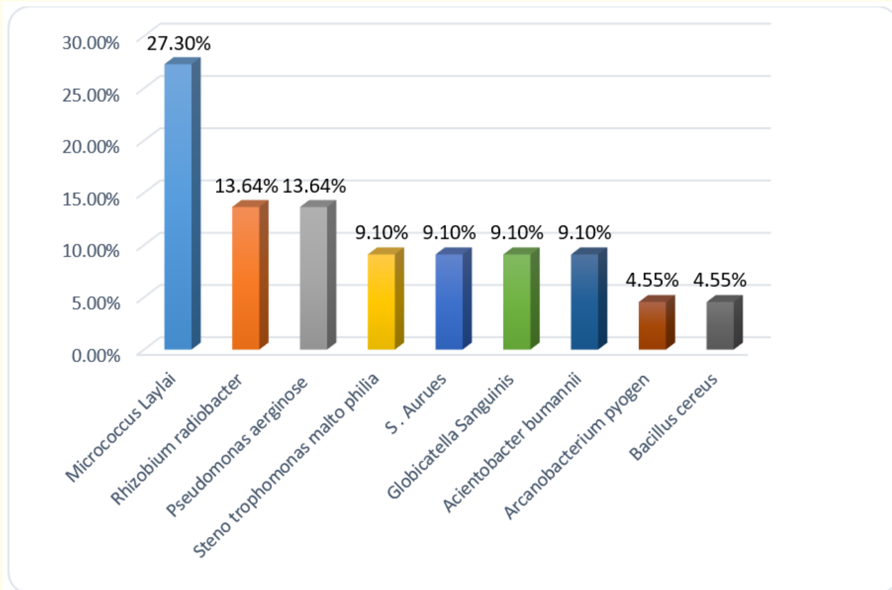

Figure 2: *Prevalence of various micro-organisms in the Emergency Department at BMC.*

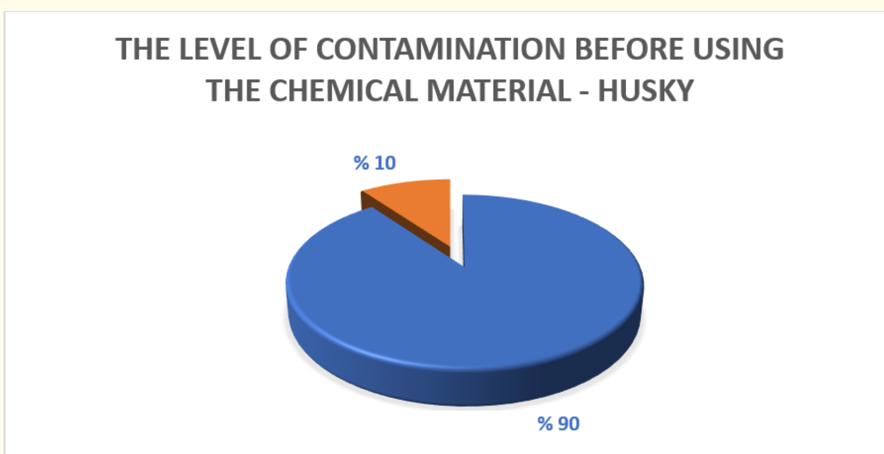

Figure 3: *The level of contamination before using the chemical material-Husky.*

The third type of disinfectant is (Surfa'safe), a ready to use chemical material that can be used on various surfaces. The active ingredient is DDAC, this ingredient has the ability to kill all types of bacteria, fungi, viruses, but cannot kill the bacterial spores. The swabs taken before cleaning with Surfa'safe indicated the level of surface contamination to be the same as before using Husky. The testing for levels of contamination after using Surfa'safe to disinfect surfaces showed the presence of all types of bacteria found prior to disinfecting.

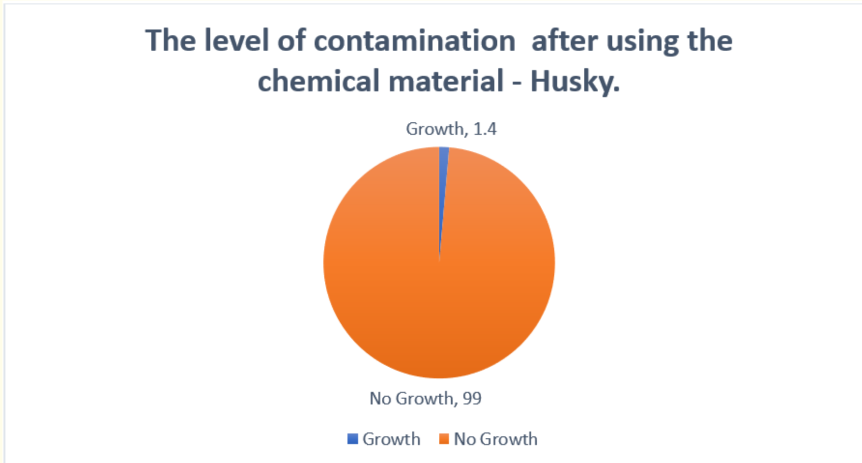

Figure 4: *The level of contamination after using the chemical material-Husky.*

These findings suggest that Surfa'safe is not effective in disinfecting surfaces with high levels of contamination.

Our study isolated two types of pathogenic bacteria, *Staphylococcus aureus* and *Acinetobacter baumannii*. Figure 5 and 6 show the prevalence of resistance in these bacteria to different generations of antibiotics, such as first, second, and third generation of cephalosporins, certain beta lactams, and some types of aminoglycosides. The level of surgical site infections with these bacteria is high inside ER departments in BMC.

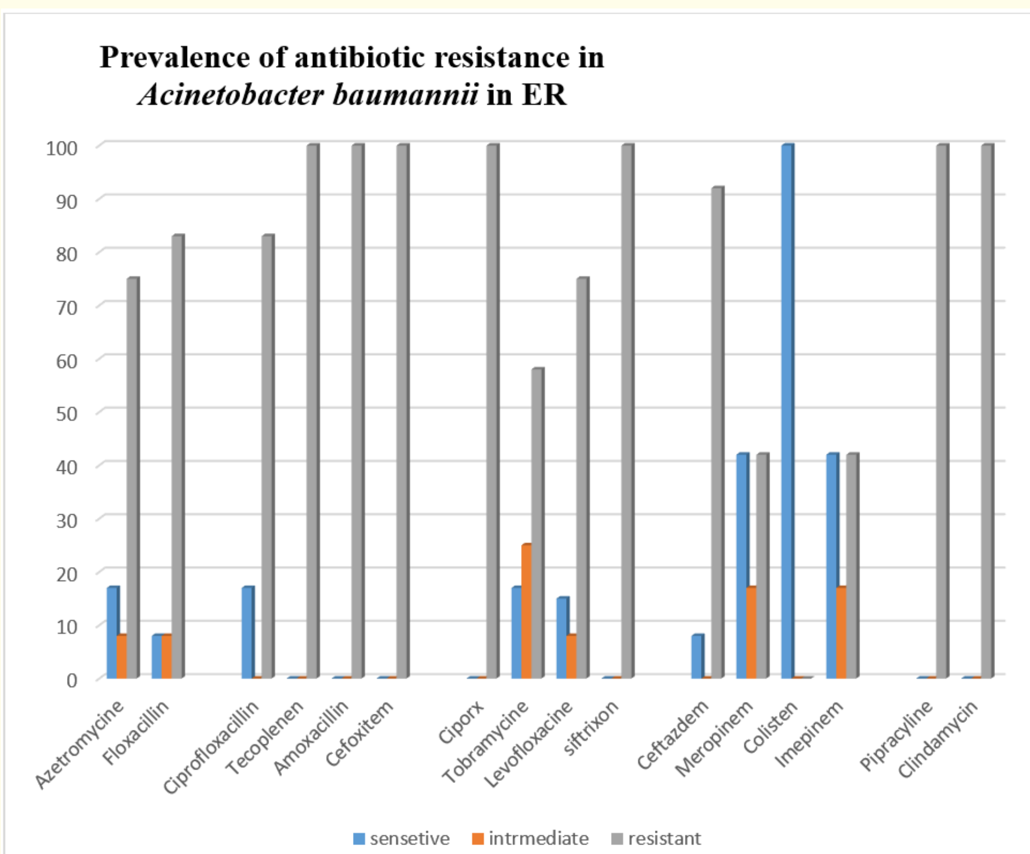

Figure 5: *Prevalence of antibiotic resistance in Acinetobacter baumannii in ER.*

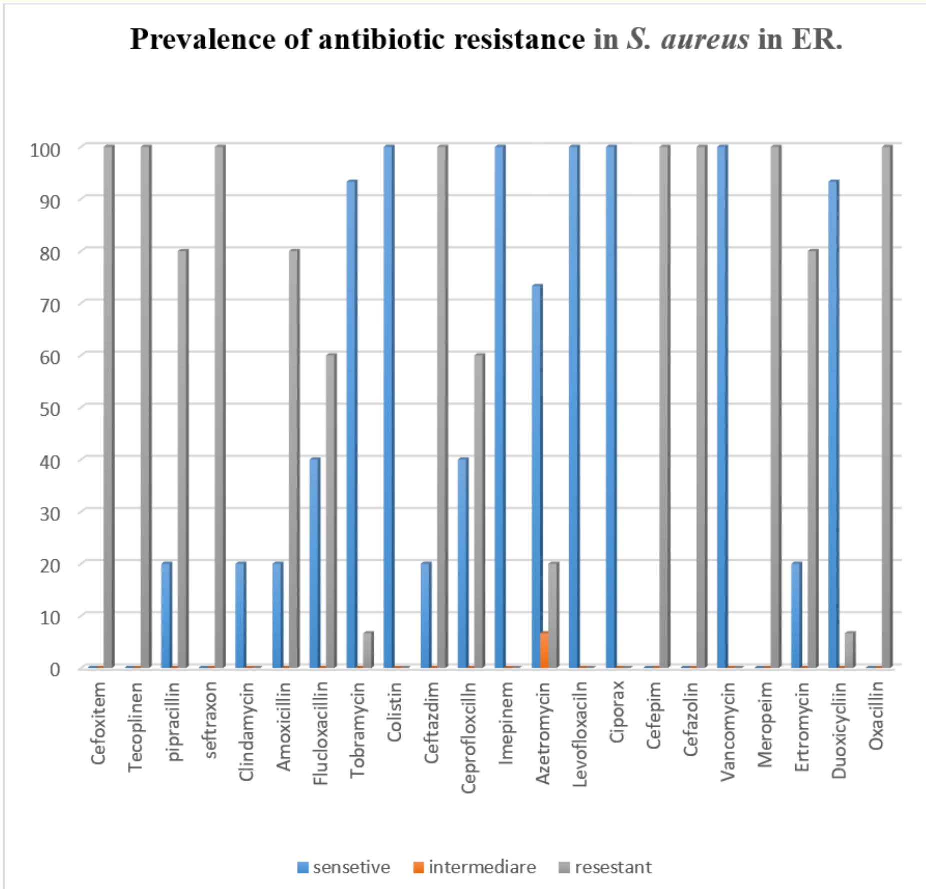

Figure 6: *Prevalence of antibiotic resistance in S. aureus in ER*

In our study we also isolated two other types of pathogenic bacteria (*E. coli* and *Pseudomonas aeruginosa*). Figure 7 and 8 show that these bacteria are also resistant to several antibiotics, such as first, second and third generation of cephalosporins, certain beta lactam antibiotics, and some types of aminoglycosides, with the exception of ciprofloxacin, amikacin, colistin, tobramycin, levofloxacin, meropenem, aztreonam, and cefotaxime. Therefore, the level of surgical site infections for these bacteria is also high inside ER departments of BMC.

Furthermore, using Husky for cleaning, keeping in consideration its dilution method and controlling the cleaning procedures/following the right cleaning procedures, together contributes towards achieving the highest standard for cleaning.

The investigation and evaluation of quality of cleaning inside ER was done with the help of a questionnaire. We found a reduction of 10.5% in the quality of cleaning during the night shift at all times. Moreover, it was found that during the night shift there was insufficient

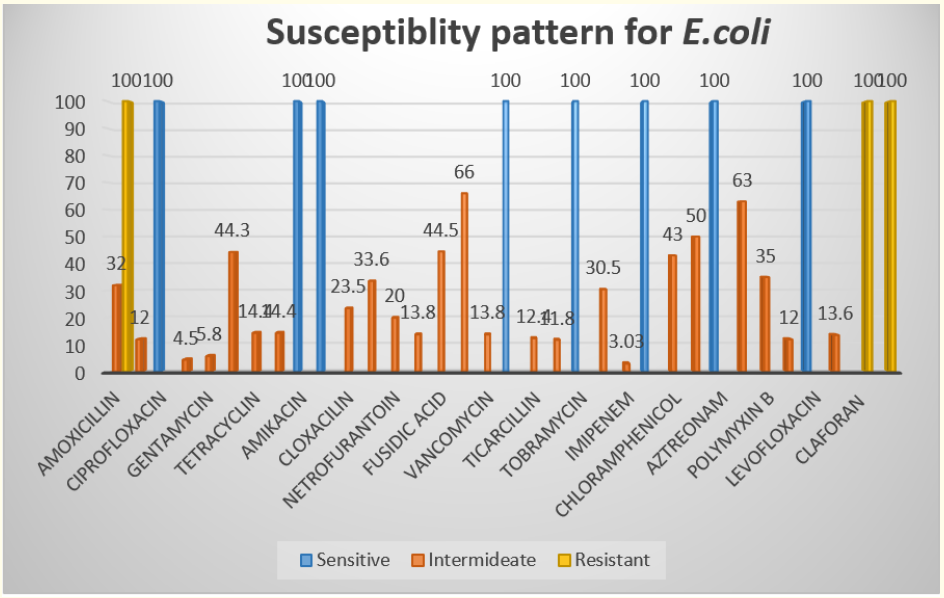

Figure 7: *Prevalence of antibiotic resistance and susceptibility in E. coli in ER.*

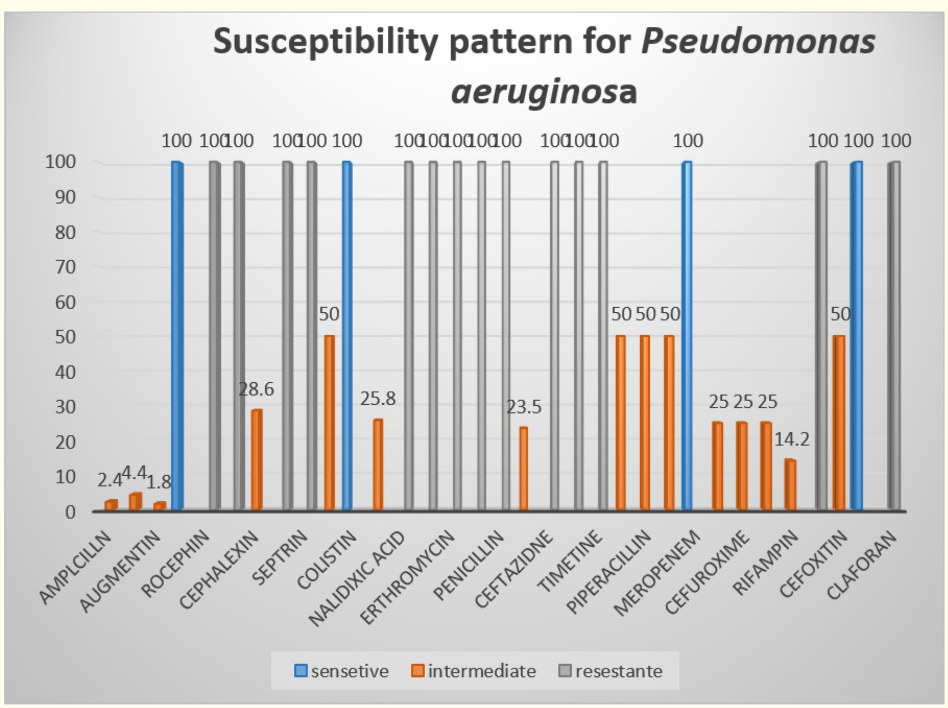

Figure 8: *Prevalence of antibiotic resistance and susceptibility in Pseudomonas aeruginosa in ER.*

staff in this critical department and the staff did not have enough knowledge of the correct cleaning procedures, besides the handling of different types of chemical disinfection. In figure 9 shows the average of the quality of cleaning in different time in ER.

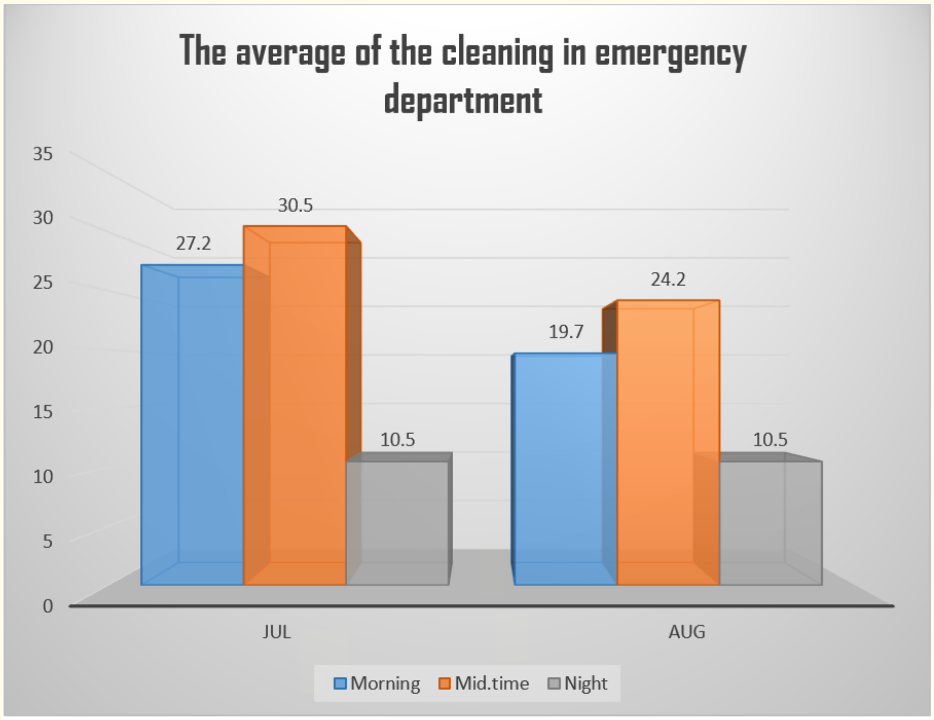

Figure 9: *The average of the quality of cleaning in different time in ER.*

Discussion

Relatively high concentrations of microorganisms detected in the hospital environment (contact surfaces and air) could be a result of outdated and underdeveloped infrastructure in hospitals, such as inefficient air conditioning and HEPA filters, or the non-implementation of modern and effective sanitary procedures by hospital infection control teams. This study, carried out in the summer of 2019, did not achieve quantitative results similar to the values observed by other researchers who conducted their tests in the winter season [25-27]. In terms of assessing the microbiological quality of samples, the results of this study are similar to other studies that found non-fermentative Gram-negative bacilli and *Stenotrophomonas maltophilia*, which are naturally resistant to several broad-spectrum antibiotics (e.g. β-lactams including all carbapenems) [28]. Bielawska-Drózd and others in 2017 showed that these bacteria may constitute to a considerable risk of HAIs due to their antimicrobial resistance to a large number of antibiotics and therefore cause treatment problems; these bacteria may be classified as potentially pathogenic agents. In the examined contact and swab samples collected from HEDs, gram-negative bacilli were more prevalent. In air samples, besides the mentioned non-fermentative gram-negative bacilli, Acinetobacter spp. and *S. maltophilia* (mainly isolated from HEDs corridors), large numbers of coagulase-negative Staphylococci with constitutive macrolide-lincosamide-streptogramin B resistance (especially in HEDs rooms) were found. In the air samples pathogenic microorganisms were not detected. Widespread usage of antibiotics, both in hospitals and ambulatory treatments, has led to the prevalence of pathogens with varied phenotypes of antimicrobial resistance i.e. alert-pathogens. Among the pathogens isolated from contact and air samples collected in HEDs, the Bielawska-Drózd study in 2017 detected multidrug-resistant *Acinetobacter baumannii* and *Burkholderia cepacia* that produce extended spectrum of beta β-lactamases. [28] Literature concerning microbiological quality assessment discusses mainly research conducted in wards that require long-term hospital stay e.g. intensive care, internal, hematology, urology, ophthalmology and surgical wards [29-35]. In this study, the bacteriological quality analysis of isolates from hospital emergency departments (not sure if s or not and the have to change everywhere) (HEDs) correlates with the profiles of potentially pathogenic species determined by other researchers in similar tests. For example, in other studies the main bacterial isolates responsible for microbiological contamination of contact surfaces in hospitals (floors, medical tables, couches, washbasins) were methicillin resistant *S. aureus* (MRSA), vancomycin resistant *Enterococcus, Clostridium difficile*, multidrug-resistant *Acinetobacter* spp., *Pseudomonas* spp., *Enterococcus* spp., and additionally Enterobacteriaceae (*E. coli*, *Enterobacter* spp., *Salmonella* spp. and *Klebsiella* spp.) [34,36-38].

According to Paluchowska., *et al.* (2012b) the largest proportion of HAIs caused by alert-pathogens is registered in intensive care wards, burns units, internal, hematological and surgical wards. The pathogens isolated most often were multi-drug resistant, non-fermentative Gram-negative *bacilli* (mainly *A. baumannii* and *P. aeruginosa*) which are recognized as the most problematic to control and eradicate [32]. Coagulase-negative *Staphylococci* and Enterococcus spp. are the key factors in HAIs and they are mostly isolated form surgical, hematological and oncology wards [33,30,34]. The results quoted above show that the diversity of microorganisms detected in this study is comparable with the standard trends. In quality testing of control samples alert-pathogens were not detected; however, potentially pathogenic strains (mainly non-fermentative Gram-negative *bacilli* like *Acinetobacter* spp. and *S. maltophilia*) were found. The relatively high level of contamination by bacteria in HEDs may attest to the ineffectiveness of implementing the standard protective precautions in the examined hospitals. In quality assessment of HEDs, the main bacterial strains found were non-fermentative Gram-negative *bacilli* and coagulase-positive *Staphylococci*; the presence of the bacteria may result from person to person transmission or introduction of pathogens from outside the hospital.

Conclusion

In conclusion, our results highlight the importance of proper disinfection of medical devices and various surfaces such as floors, doors, doorknobs, treatment tables, washbasins, anesthetist machines and operation tables in the hospital emergency department.

Multiple factors play a key role in bacterial contamination and transmission, which possibly results in number of HAIs. Not every disinfectant is effective in reducing or eliminating the bacterial load. The emergence of resistant bacteria, hospitalization of older and critical patients, high turnover of healthcare staff, lack of process organization between units and non-observation of protocols makes the standardization process difficult [10] and all these factors need to be addressed to prevent HAIs. Environmental disinfection has a central role in the primary prevention of HAIs. Despite a limited number of studies evaluating the role of surface contamination in operating rooms, there is accumulating evidence that the inanimate environment of the operating room can become contaminated with pathogens despite standard environmental cleaning. These pathogens can then be transmitted to the hands of personnel and then to patients and may result in SSIs and infection outbreaks. Contaminated surfaces can be responsible for the transmission of pathogens in the operating room setting. Further studies are necessary to quantify the role of contaminated surfaces in the transmission of pathogens and to inform the most effective environmental interventions. Given the serious consequences of SSIs, special attention should be given to the proper cleaning and disinfection of the inanimate environment in operating rooms in addition to the other established infection control measures to reduce the burden of SSIs.

Limitations of the Study:

1. Educational of ER staff to protect themselves and patients from most pathogenic microbes.
2. Reinforcement of hand hygiene to reduce the transmission of infection.
3. Monitoring of disinfectant materials and the method of dilution to ensure elimination of broad spectrum of pathogenic microbes.
4. Thorough training program for cleaning staff regarding the correct cleaning and disinfection procedures of ER environment.
5. Control of antibiotics use and implementation policy for using the antibiotics inside the Emergency department.
6. Establishing these experiments on more than one hospital and increasing the number of samples and swabs to compare the infection of nosocomial infection bacteria of hospitals inside Benghazi city and setting a way to get rid of them.

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Bibliography

1. Bielawska-Drózd Agata., *et al*. "Evaluation of the Levels and Quality of Microbial Contamination in Medical Emergency Departments in Comparison to Other Workplaces". *Polish Journal of Microbiology* 65.4 (2016): 465-469.
2. Maszkiewicz W. "Nosocomial infections in neonates (in Polish)". *Zdr* 117 (2007): 260-263.
3. Heczko PB and J Wójkowska-Mach. "Nosocomial infections (in Polish), 1st edition". PZWL, Warszawa (2009): 61.
4. Emori TG and RP Gaynes. "An Overview of Nosocomial Infections, Including the Role of the Microbiology Laboratory". *Clinical Microbiology Reviews* 6.4 (1993): 428-442.
5. Sakula A. "R T H Laennec 1781--1826 His Life and Work: a Bicentenary Appreciation". *Thorax* 36.2 (1981): 81-90.
6. Horan Teresa C., *et al*. "CDC/NHSN Surveillance Definition of Health Care-Associated Infection and Criteria for Specific Types of Infections in the Acute Care Setting". *American Journal of Infection Control* 36.5 (2008): 309-332.
7. Morales Eva., *et al*. "Hospital Costs of Nosocomial Multi-Drug Resistant Pseudomonas Aeruginosa Acquisition". *BMC Health Services Research* 12.1 (2012).
8. Ogilvie Isla., *et al*. "Burden of Community-Acquired and Nosocomial Rotavirus Gastroenteritis in the Pediatric Population of Western Europe: a Scoping Review". *BMC Infectious Diseases* 12.1 (2012).
9. Roberts Rebecca R., *et al*. "The Use of Economic Modeling to Determine the Hospital Costs Associated with Nosocomial Infections". *Clinical Infectious Diseases* 36.11 (2003): 1424-1432.
10. WHO. "Report on the Burden of Endemic Health Care-Associated Infection Worldwide (2011): 1-34.
11. Klevens R Monina., *et al*. "Estimating Health Care-Associated Infections and Deaths in U.S. Hospitals, 2002". *Public Health Reports* 122.2 (2007): 160-166.

12. ECDC. "Annual epidemiological report Reporting on 2010 surveillance data and epidemic intelligence data (2012).
13. P Manzi., *et al*. "Asepsi e disinfezione nel controllo delle infezioni". *Progettare per la Sanità* 119 (2010): 37-40.
14. Alleyne SA., *et al*. "Stethoscopes: Potential Vectors of Clostridium Difficile". *Journal of Hospital Infection* 73.2 (2009): 187-189.
15. Fenelon L., *et al*. "Contamination of Stethoscopes with MRSA and Current Disinfection Practices". *Journal of Hospital Infection* 71.4 (2009): 376-378.
16. Hartmann Bernd., *et al*. "Computer Keyboard and Mouse as a Reservoir of Pathogens in an Intensive Care Unit". *Journal of Clinical Monitoring and Computing* 18.1 (2003): 7-12.
17. G Messina., *et al*. "Indagine sulla contaminazione microbica di stetoscopi, telefoni e tastiere di computer presso una casa di cura". *Mondo Sanitario* 19.10 (2012): 1-5.
18. Messina Gabriele., *et al*. "Impact of a Disinfecting Technique on Microbial Contamination of Computer Keyboards and Telephone Handsets". *Journal of Hospital Administration* 2.4 (2013): 1.
19. Messina Gabriele., *et al*. "How Many Bacteria Live on the Keyboard of Your Computer?" *American Journal of Infection Control* 39.7 (2011): 616-618.
20. Rutala William A., *et al*. "Bacterial Contamination of Keyboards: Efficacy and Functional Impact of Disinfectants". *Infection Control and Hospital Epidemiology* 27.4 (2006): 372-377.
21. A Singh and B Purohit. "Mobile phones in hospital settings: a serious threat to infection". *Occupational Health and Safety* 81.3 (2012): 42-44.
22. I Youngster., *et al*. "The stethoscope as a vector of infectious diseases in the paediatric division". *Acta Paediatrica, International Journal of Paediatrics* 97.9 (2008): 1253-1255.
23. ME Zuliani Maluf., *et al*. "Stethoscope: A Friend or an Enemy?" *Sao Paulo Medical Journal* 120.1 (2002): 13-15.
24. S Kalenic., *et al*. "The floors, doors, emergency surfaces, door knob, treatment table, wash basin, anesthetist machines and operation table in the emergency department in the Benghazi Medical Center (BMC) hospital before and after use of a disinfecting technique". *Lijecnicki Vjesnik* 133.5-6 (2011): 155-170.
25. National Sanitary Inspectorate (NSI). Country Sanitary Conditions Report (2009).
26. National Sanitary Inspectorate (NSI). Country Sanitary Conditions Report (2010).
27. National Sanitary Inspectorate (NSI). Country Sanitary Conditions Report (2011).
28. Bielawska-Drózd Agata., *et al*. "Evaluation of the Levels and Quality of Microbial Contamination in Medical Emergency Departments in Comparison to Other Workplaces". *Polish Journal of Microbiology* 65.4 (2016): 465-469.

29. Kępa M., *et al*. "Antimicrobial susceptibility of bacterial strains isolated at Urology Ward environment (in Polish)". *Annales Academiae Medicae Stetinensis* 66 (2012): 7-15.

30. Nikaeen Mahnaz., *et al*. "Evaluation of Bio-Aerosols Concentration in the Different Wards of Three Educational Hospitals in Iran". *International Journal of Environmental Health Engineering* 1.1 (2012): 47.

31. Paluchowska P., *et al*. "[Occurrence of Alert Pathogens in Hospital Environment". Part I. ESBL-Producing Enterobacteriaceae Strains]". Medycyna Doswiadczalna i Mikrobiologia, U.S. National Library of Medicine 64.1 (2012): 45-53.

32. Paluchowska Paulina., *et al*. "Occurrence of Alert Pathogens in Hospital Environment Part II. Multidrug-ResistantNonFermentingBacilli".MedycynaDoswiadczalnaMikrobiologia,U.S.NationalLibraryofMedicine (2012).

33. Guzek A., *et al*. "*In Vitro* Susceptibility of Staphylococci and Enterococci to Vancomycin and Teicoplanin". *Neurobiology of Respiration Advances in Experimental Medicine and Biology* (2013): 125-132.

34. Hoseinzadeh Edris., *et al*. "Evaluation of Bioaerosols in Five Educational Hospitals Wards Air in Hamedan, During 2011-2012". *Jundishapur Journal of Microbiology* 6.6 (2013).

35. Seweryn M., *et al*. "Alert Microorganisms Isolated From Patients Hospitalized in Małopolskie Province in 2010-2012". Przeglad Epidemiologiczny, U.S. National Library of Medicine.

36. Kramer Axel., *et al*. "How Long Do Nosocomial Pathogens Persist on Inanimate Surfaces? A Systematic Review". *BMC Infectious Diseases* 6.1 (2006).

37. Rafiei Neda., *et al*. "Fungal and Bacterial Contamination on Indoor Surfaces of a Hospital in Mexico". *Jundishapur Journal of Microbiology* 5.3 (2012): 460-464.

38. Weber David J., *et al*. "The Role of the Surface Environment in Healthcare-Associated Infections". *Current Opinion in Infectious Diseases* 26.4 (2013): 338-344.

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