Original article

The Prevalence of Antibiotic-Resistant Bacterial Nasal Carriage at a Saudi University Hospital

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Summary

Objectives: To identify the nasal carriage of common and novel nosocomial pathogens that may occur in the patients of an Intensive Care Unit (ICU) of a Saudi university hospital.

Methods: Nasal swabs taken from 95 randomly selected patients were used as clinical specimens, which were plated onto blood, MacConkey, and chocolate agars. Thereafter, these pathogens were grown, they were identified using Matrix Assisted Laser Desorption Ionization-

Time of Flight mass spectrometry (MALDI-TOF MS) and tested for antimicrobial-susceptibility using an automated VITEK2 system.

Results: *Pseudomonas aeruginosa* was the most prevalent species, accounting for 16 (16.84%) isolates. Gentamicin was the most effective antibiotic against Multidrug-Resistant (MDR) *Pseudomonas aeruginosa, Acinetobacter baumannii,* and *Klebsiella pneumonia*. Trimethroprim-sulfamethoxazole A was the least effective antibiotic against *K. pneumonia*.

Conclusions: The AMR bacteria isolated from the nasal swabs showed different resistance patterns compared to those in other studies in Saudi Arabia. This data could assist healthcare practitioners to prescribe the most effective and appropriate antibiotics, and it also provides information upon which infection control plans, which are critical for outbreak prevention, can be based.

Highlights: • *Pseudomonas aeruginosa* was the most prevalent species. • Gentamicin is the most active antimicrobial compounds against ICU bacteria. • The AMR nosocomial bacteria showed different resistance and sensitivity patterns.

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Introduction

The intensive care units (ICUs) of King Abdulaziz University Hospital comprise the Medical Intensive Care Unit (MICU), the Surgical Intensive Care Unit (SICU), the Pediatric Intensive Care Unit (PICU), and the Neonatal Intensive Care Unit (NICU), which are collectively known as "the epicenter of infections".[1] This is because the patient communities, which they serve (neonates, pregnant women, and immunocompromised patients), have reduced immunity. These patients are particularly vulnerable to infection. The ICU population has one of the highest rates of nosocomial infection (20–30% of all admissions).[2] This has a huge impact on morbidity, mortality, and hospital costs. Nosocomial infections are mainly caused by multidrug-resistant (MDR) pathogens, which are considered a major risk to public health. Prevention of nosocomial infection, timely diagnosis, and appro-

priate patient and infection management are key factors for improving nosocomial infection control and patient outcomes. The most common nosocomial infections in the ICU are surgical site infections, ventilator-associated pneumonia, central-line associated bloodstream infection, and catheter-related urinary tract infection.[3]

Infection control measures are an essential daily practice, because many patients admitted to the ICU are colonized and/or infected with MDR pathogens. In addition to these measures, a quick and accurate diagnostic tool is required for efficient infection control practice in hospitals. In Saudi Arabia, the most common nosocomial pathogens include *E. coli, K. pneumoniae, P. aeruginosa,*[4] and *A. baumannii,*[5] which are antimicrobial-resistant (AMR) bacteria associated with outbreaks and high mortality rates. The objectives of this study were to identify common and novel nosocomial pathogens in the ICUs and to assess the pre-

valence of AMR bacteria isolated from patients admitted to the ICUs at the King Abdulaziz University, Jeddah, Saudi Arabia.

Materials and Methods

This study was a single center cross-sectional epidemiological pilot study. It was approved by the Unit of Biomedical Ethics Research Committee, Faculty of Medicine, King Abdulaziz University and conducted in accordance with the Helsinki Declaration of 1975 as revised in 2000.

Collection of Strains
Isolates were collected from patients with respiratory infections admitted to the ICUs at King Abdulaziz University Hospital, a large 900 bed tertiary care hospital in Jeddah, from 25 February until 15 August 2016. The ICUs included the following: the MICU, SICU, PCICU, in addition to Pediatric Cardiac Unit (PICU), and Isolation Unit (ISO). Nasal swabs were used as clinical specimens.

Species and Antibiotic Susceptibility-Based Identification
Bacteria were plated onto enriched selective media (5% blood, MacConkey, and chocolate agars, made by Saudi Prepared Media Laboratory Company, Saudi Arabia) to identify their species. MacConkey agar was incubated at 37°C for 24 h, blood agar was incubated at 37oC in 5% CO_2 for 24 h, and chocolate agar was incubated at 35oC in 5% CO_2 for 24 h. Then, bacteria were identified using MALDI-TOF MS (bioMerieux, Marcy l' Etoile, France). Next, bacteria underwent antimicrobial susceptibility testing (AST) using a VITEK2 system (bioMerieux, Marcy l' Etoile, France). Finally, the results of these analyses were written up as a report, which was sent to the infection control unit of the hospital and related departments.

Results

This study examined nasal specimens from 95 randomly selected patients with a mean age of 41.14 ± 30.14. Most participants were male (69.47%) and non-Saudi (68.42%). As shown in Table 1, the majority of cases were from the MICU (45.26%).

Identification of Nosocomial Pathogens
Of the 95 isolates, *P. aeruginosa* was the most prevalent species, as it was present in 16 (16.84%) specimens, followed by *A. baumannii* (14.72%), *K. pneumoniae* (13.68%), and extended-spectrum β-lactamase (ESBL)-

Table 1	Demographic characteristics of the study sample
Variable	**Total N = 95**
Age	41.14 ± 30.14
Gender *Male* *Female*	66 (69.47%) 29 (30.53%)
Nationality *Saudi* *Non-Saudi*	30 (31.58%) 65 (68.42%)

producing *K. pneumoniae* (10.53%) (Tables 2 and 3). Gentamicin was the most effective antibiotic for MDR *P. aeruginosa, A. baumannii,* and *K pneumoniae*. Trimethroprim-sulfamethoxazole A was the drug of choice for treating ESBL-positive *K. pneumonia*. Ciprofloxacin was equivalent to gentamicin in MDR *P. aeruginosa* cases (Table 2). However, trimethroprim-sulfamethoxazole A was the least effective antibiotic in MDR *P. aeruginosa* and *K. pneumoniae*, while ampicillin was the most effective for ESBL-positive *K. pneumoniae*. *A. baumannii* developed resistance to a variety of antibiotics (Table 3).

Pattern of Antimicrobial Susceptibility/Resistance
In Table 4, the antimicrobial susceptibilities and resistance of strains identified in this study have been shown. The antimicrobial compound that was most active against nasal carriage bacteria bacteria was gentamicin (43.16%), followed by ciprofloxacin (29.47%), trimethroprim-sulfamethoxazole (26.32%), and piperacillin-tazobactam (24.21%). Gentamicin was considered the first line of treatment (Group A) while ciprofloxacin and trimethroprim- sulfamethoxazole were considered the second line of treatment (Group B). Trimethroprim- sulfamethoxazole was highly effective as a first or second line of treatment. Of the total number of isolates in this study (N = 95), 22.1% (n = 21) were gentamicin resistant, 20.0% were piperacillin-tazobactam resistant, 18.95% were ceftazidime resistant, and 18.95% were resistant to ciprofloxacin. Notably, the most antimicrobial resistance was to Group A, except piperacillin-tazobactam (Group B).

Discussion

Nosocomial infection usually occurs 48 hours after hospital admission.[3] Microorganisms that cause this type of infection are commonly AMR. It is believed that antimicrobial resistance in nosocomial pathogens is acquired by their evolution and transmission through

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| Table 2 | | Susceptibility pattern (frequency and percentage) of 95 strains isolated from intensive care unit patients | | | | | | | |

Strain	Total (N = 95)**	Gentamicin	Ceftazidime	Trimethroprim-sulfametho-xazole A	Ciproflo-xacin	Piperacillin-tazobactam	Trimethroprim-sulfametho-xazole B	Ciproflo-xacin	Colistin
MDR *Pseudomonas aeruginosa*	16 (16.84%)	8 (50.00%)	3 (18.75%)	3 (18.75%)	8 (50.00%)	4 (25.00%)	6 (37.50%)	3 (18.75%)	2 (12.50%)
Acinetobacter baumannii	14 (14.74%)	6 (42.86%)	2 (14.29%)	3 (21.43%)	0	3 (21.43%)	2 (14.29%)	3 (21.43%)	3 (21.43%)
Klebsiella pneumonia	13 (13.68%)	7 (53.85%)	2 (15.38%)	2 (15.38%)	1 (7.69%)	3 (23.08%)	1 (7.69%)	5 (38.46%)	0
ESBL-positive *Klebsiella pneumoniae*	10 (10.53%)	1 (10.00%)	0	4 (40.00%)	0	0	1 (10.00%)	1 (10.00%)	1 (10.00%)

ESBL: Extended-spectrum β-lactamase; MDR: multidrug-resistant
*all percentages are raw percentages except N, which is a column percentage
**Each patient with specific strain may susceptible to one antibiotic or more. These analyses are based on the most resistance and the most susceptible antibiotic only. If a patient with specific strain is the most susceptible to certain antibiotic, it is not necessary to be very resistance to all other antibiotics.

| Table 3 | | Resistance pattern (frequency and percentage) of 95 strains isolated from intensive care unit patients | | | | | |

Strain	Total (N = 95)**	Gentamicin	Ceftazidime	Trimethroprim-sulfamethoxazole A	Ciprofloxacin	Ampicillin	Piperacillin-Tazobactam
MDR *Pseudomonas aeruginosa*	16(16.84%)	3(18.75%)	1(6.25%)	4(25.00%)	2(12.50%)	1(6.25%)	2(12.50%)
Acinetobacter baumannii	14(14.74%)	3(21.43%)	3(21.43%)	2(14.29%)	2(14.29%)	3(21.43%)	3(21.43%)
Klebsiella pneumoniae	13(13.68%)	1(7.69%)	2(15.38%)	4(30.77%)	2(15.38%)	1(7.69%)	2(15.38%)
ESBL-positive *Klebsiella pneumoniae*	10(10.53%)	1(10.00%)	1(10.00%)	1(10.00%)	1(10.00%)	3(30.00%)	1(10.00%)

ESBL: Extended-spectrum β-lactamase; MDR: multidrug-resistant
*all percentages are raw percentages except N, which is a column percentage.
**Each patient with specific strain may resistance to one antibiotic or more. These analyses are based on the most resistance and the most susceptible antibiotic only. If a patient with specific strain is the most resistance to certain antibiotic, it is not necessary to be very susceptible to all other antibiotics.

direct contact with hospital environments or procedures.[3] This study, the bacteria, which unexpectedly showed new patterns of antibiotics resistance. This could be due to the fact that such bacteria are under the most evolutionary pressure to evolve resistance to antimicrobial agents as a consequence of antibiotics misuse, a profound malpractice throughout the country, which is related to drug resistance and abuse.[6,7] In addition, it may be an indication of antibiotics overuse, insufficient dose, practitioners' failure to follow instructions for use, and/or its interaction with other medications.

A group of bacteria commonly found to be AMR includes MDR Gram negative bacteria, penicillin-resistant pneumococci (PRP), vancomycin-resistant Enterococci (VRE), and methicillin-resistant *Staphylococcus aureus* (MRSA).[3] These bacteria have acquired antimicrobial resistance as a result of inappropriate antibiotic therapy, which is a risk factor for morbidity and mortality in patients.[3] In this study, MDR *P. aeruginosa* was the most prevalent, probably because it is considered one of the most common nosocomial bacteria isolated from immunocompromised patients[8] suffering from respiratory tract infection.[9] In addition,

Table 4	Frequency and percentage of the most effective and least effective antibiotics		
Status	**Variable (Antibiotic)**	**Group**	**Total N = 95**
Most Effective	Gentamicin	A	41 (43.16%)
	Ciprofloxacin	B	28 (29.47%)
	Trimethroprim-sulfamethoxazole	B	25 (26.32%)
	Piperacillin-tazobactam	B	23 (24.21%)
	Trimethroprim-sulfamethoxazole	A	21 (22.11%)
	Colistin	C	16 (16.84%)
	Ciprofloxacin	A	15 (15.79%)
	Ceftazidime	A	12 (12.63%)
Least Effective	Gentamicin	A	21 (22.11%)
	Piperacillin-tazobactam	B	19 (20.0%)
	Ceftazidime	A	18 (18.95%)
	Ciprofloxacin	A	18 (18.95%)
	Trimethroprim-sulfamethoxazole	A	17 (17.89%)
	Ampicillin	A	16 (16.84%)

Note: Gentamicin was the most common antibiotic used. However, it was the most effective and should be used in the first line of treatment (group A) in some patients (41), and some others patients who used the same antibiotic as a first line (group A) developed resistance and it was the least effective in 21 participants. For Ciprofloxacin, it was the most effective as a second line of treatment (group B) in 28 patients or as a first line in 15 patients. However, some other patients who used it a first line (Group A) developed a resistance and it was least effective in 18 of them.

A. baumannii is one of the major pathogens causing nosocomial infections in ICU patients, especially septicemia, pneumonia, and urinary tract infection following hospitalization.[5,10]

Gentamicin, ciprofloxacin, and piperacillin-tazobactam antibiotics were chosen as they are commonly used to treat most nosocomial infections.[11] Due to the misuse of these antibiotics, nosocomial bacteria have developed various patterns in their resistance and sensitivity to antimicrobial agents.[12] The present study identified the most common nosocomial bacteria isolated at ICU units at the King Abdulaziz University Hospital in Jeddah. This list of bacteria were partly aligned with those isolated at an adult ICU at a tertiary care hospital in Riyadh,[13] which showed that the most frequently isolated bacteria were *A. baumannii*, followed by *P. aeruginosa, E. coli, K. pnemoniae, Stenotrophomonas maltophilia,* and *Enterobacter* respectively.[13]

In the present study, all strains were collected in one center, meaning that findings may not be generalizable to other medical centers. The random collection of samples may be a limitation, as it may have neglected some pathogens and/or over or underestimated prevalence a specific strain of bacteria than another. The sample did not include patients who were colonized or infected at the time of hospital admission, and such samples were not kept for the purposes of identification. In addition, specimens were only nasal swabs for each patient, regardless of whether they were colonized at additional anatomical sites. Thus, the results of this study must be interpreted with caution and can be used as an indicator for outbreak. However, this study is more of a pilot, and a larger research project using a sample size calculation is being planned. In the future, we will use whole genome sequencing to provide comprehensive information about the diagnosis of nosocomial pathogens. This will allow a tracking study of infectious disease outbreaks. In addition, these two molecular technologies can catalog microbes present in a certain environment, such as the ICU, and how these microbes are transmitted from place to place or person to person. This will allow hospitals and healthcare centers to predict and prevent outbreaks, saving lives, time, and costs.

Conflict of interest statement
The authors declare no conflict of interest.

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Περίληψη

Τίτλος

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References

1. Brusselaers N, Vogelaers D, Blot S. The rising problem of antimicrobial resistance in the intensive care unit. *Annals of Intensive Care* 2011;1:47-.
2. Hanberger H, Garcia-Rodriguez J, Gobernado M, et al. Antibiotic susceptibility among aerobic gram-negative bacilli in intensive care units in 5 european countries. *JAMA* 1999;281:67-71.
3. Trubiano JA, Padiglione AA. Nosocomial infections in the intensive care unit. *Anaesthesia & Intensive Care Medicine* 2015;16:598-602.
4. Zowawi HM. Antimicrobial resistance in Saudi Arabia. An urgent call for an immediate action. *Saudi medical journal* 2016;37:935-40.
5. Almasaudi SB. Acinetobacter spp. as nosocomial pathogens: Epidemiology and resistance features. *Saudi Journal of Biological Sciences* 2016.
6. Aljadhey H, Assiri GA, Mahmoud MA, Al-Aqeel S, Murray M. Self-medication in Central Saudi Arabia: Community pharmacy consumers' perspectives. *Saudi medical journal* 2015;36:328-34.
7. Al-Mohamadi A, Badr A, Bin Mahfouz L, Samargandi D, Al Ahdal A. Dispensing medications without prescription at Saudi community pharmacy: Extent and perception. *Saudi Pharmaceutical Journal* 2013;21:13-8.
8. Tassios PT, Gennimata V, Spaliara-Kalogeropoulou L, et al. Multiresistant Pseudomonas aeruginosa serogroup O:11 outbreak in an intensive care unit. Clinical Microbiology and Infection 1997;3:621-8.
9. Ahmed S, Al-Harbi MN. Antibiotic susceptibility pattern of isolates of Pseudomonas aeruginosa in a Saudi Arabian Hospital. *Bangladesh Journal of Medical Science* 2014;13:45.
10. Bergogne-Bérézin E, Towner KJ. Acinetobacter spp. as nosocomial pathogens: microbiological, clinical, and epidemiological features. *Clinical Microbiology Reviews* 1996;9:148-65.
11. Izadpanah M, Khalili H. Antibiotic regimens for treatment of infections due to multidrug-resistant Gram-negative pathogens: An evidence-based literature review. *J Res Pharm Pract* 2015;4:105-14.
12. Hsueh PR, Chen WH, Luh KT. Relationships between antimicrobial use and antimicrobial resistance in Gram-negative bacteria causing nosocomial infections from 1991-2003 at a university hospital in Taiwan. *International journal of antimicrobial agents* 2005;26: 463-72.
13. Al Johani SM, Akhter J, Balkhy H, El-Saed A, Younan M, Memish Z. Prevalence of antimicrobial resistance among gram-negative isolates in an adult intensive care unit at a tertiary care center in Saudi Arabia. *Annals of Saudi Medicine* 2010;30:364-9.