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Isolation, Identification, and Antibiotic Susceptibility Patterns of *Pseudomonas aeruginosa* Strains from Various Clinical Samples in a University Hospital in Northern Cyprus

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BACKGROUND/AIMS
Pseudomonas aeruginosa is a severe opportunistic bacterium responsible for frequently lethal nosocomial infections. It is highly resistant to commonly used antibiotics, creating a real challenge in the treatment of nosocomial infections.

MATERIALS and METHODS
A total of 152 samples were collected from various departments of the hospital, which included wound, blood, urine, cerebrospinal fluid (CSF), sputum, aspiration fluid, and ear, nose, and throat swab samples. The susceptibility pattern of bacteria was determined by using the BD Phoenix 100 automated microbiology system. *P. aeruginosa* was also identified according to its cultural, microscopic, morphological, and biochemical characteristics.

RESULTS
Samples were identified by using BD Phoenix 100 and conventional biochemical tests are used for confirmation. The results of antimicrobial susceptibility testing indicated that *P. aeruginosa* was sensitive to piperacillin–tazobactam (92.5%), meropenem (87.2%), colistin (86.7%), ticarcillin–clavulanate (86.6%), amikacin (81.5%), imipenem (80.8%), norfloxacin (79.5%), cefepime (78.4%), ceftazidime (76.0%), gentamicin (76.0%), levofloxacin (73.5%), and ciprofloxacin (73.2%). On the other hand, the resistance rates of amoxicillin, ampicillin–sulbactam, cefuroxime, nitrofurantoin, cefoxitin, trimethoprim/sulfamethoxazole, and ceftriaxone were found to be 100%, 98.7%, 97.7%, 97.7%, 94.7%, 94.7%, and 93.8% respectively. The prevalence of antibiotic susceptibility of *P. aeruginosa* in Cyprus is more in males (62.5%) than in females (37.5%).

CONCLUSION
Knowledge about local antimicrobial resistance patterns of *P. aeruginosa* is essential for the guidance of empirical therapy by physicians, medical microbiologists, and public health officials. Antimicrobial surveillance should be done periodically to detect current resistance in our local area.

Keywords: Antibiotic susceptibility, Northern Cyprus, pseudomonas aeruginosa, resistance

INTRODUCTION

Pseudomonas aeruginosa is a gram-negative, bacillus, and non-spore forming bacterium. It is widely distributed in nature including soil, water, and various types of vegetation throughout the world (1, 2). It causes community-acquired and nosocomial infections such as pneumonia, urinary tract infections, and bacteremia. The infections can be particularly important in patients who are immunocompromized, such as neutropenic or cancer patients (3, 4). Nowadays, the rates of morbidity and mortality have been increased because of multidrug-resistant *P. aeruginosa* strains (5).

P. aeruginosa has an intrinsic and acquired resistance against many antibiotics. In addition, it can also gain resistance due to abusive or misuse of commonly used antibiotics (6). The microorganism possesses a natural resistance to antibiotics

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including aminoglycosides, cephalosporins, fluoroquinolones, and penicillins (7). This organism is the most common etiological agent of pneumonia, urinary tract infections, and in the bloodstream (8).

These bacteria have combination resistance mechanisms according to their cephalosporins production, having efflux pumps, low intrinsic outer membrane permeability, and target mutations. Because of this situation, in combination with the misuse of drugs, hospital infections of multidrug-resistant *P. aeruginosa* cause serious problems (9). Clinical isolates of gram-negative rods including *P. aeruginosa* have been shown to produce an enzyme called inducible beta-lactamases, which is the mechanism of bacterial resistance to beta-lactam antibiotics. These enzymes are plasmid-mediated (4). *P. aeruginosa* is isolated from various body fluids including sputum, urine, wounds, eye, ear, and blood. A narrow class of antibiotics is effective against *P. aeruginosa,* including the carboxypenicillins, quinolones, cephalosporins, and aminoglycosides (9, 10). As the antibiotic resistance profiles of *P. aeruginosa* can change in years, prevalence studies must be carried out regularly. The aim of this study was to determine the antibiotic susceptibility of *P. aeruginosa* from clinical samples and to contribute the application of appropriate empiric therapy in the Near East University in Northern Cyprus.

MATERIALS and METHODS

Collection of Samples

This study was carried out at our microbiology laboratory between June 2014 and October 2015. A total of 152 samples were collected from the different units of the hospital. These samples included urine, blood, nasal swab, sputum, aspiration fluids, intravascular catheters, CSF, and wound. All the samples were first processed to get a pure culture by subculturing using general and selective media as blood and eosin methylene blue agar. The demographic information was obtained from the hospital systems. The sensitivity pattern of the bacteria was determined by using an automated microbiology system (Becton-Dickinson, USA) according to the standards of Clinical & Laboratory Standards Institute. Ethics committee approval was not taken due to in vitro design of the study. This study does not include human participants. Thus, no consent form was required.

Statistical Analysis

Statistical Package for the Social Sciences version 22.0 (SPSS IBM Corp.; Armonk, NY, USA) was used for the analysis of the data.

RESULTS

A total of 152 records were identified as *P. aeruginosa* from different specimens and were mostly isolated from urine samples in this study (Figure 1). In our study, 62.5% of the subjects were males and 37.5% were females. Their ages ranged from 10 to 67 years and are presented in Figure 2. When the age distribution was considered, *P. aeruginosa* infections were more frequently seen in the ages between 10 and 20 and over 60. The antimicrobial susceptibility test revealed that *P. aeruginosa* was highly sensitive to most of the antibiotics tested. Piperacillin–tazobactam was the most effective drug against all *P. aeruginosa* (92.5% sensitivity) followed by meropenem (87.2%), colistin (86.7%), ticarcillin/clavulanate (86.6%), amikacin (81.5%), and imipenem (80.8%). On the other hand, *P. aeruginosa* had 98.7% resistance to ampicillin/sulbactam antibiotic and the other high resistance rates were for cefoxitin (94.7%), trimethoprim/sulfamethoxazole (94.7%), and ceftriaxone (93.8%) (Table 1).

DISCUSSION

According to the scientific studies, multidrug-resistant strains for both the hospitals and community constitute a serious problem, especially infections, induced by *P. aeruginosa* across the world (4). *P. aeruginosa* is a nosocomial bacterium, which is present in disinfectants, respiratory equipment, sinks, taps, and mops in the hospital by forming a biofilm (1). A total of 152 records were identified as *P. aeruginosa* from different specimens and were mostly isolated from urine samples in this study (Figure 1). Regarding gender, 62.5% of the subjects were males whereas 37.5% were females. It is reported that infections caused by *P. aeruginosa* are more common in males than females. This is comparable with other studies that detected 61% of pseudomonas infections in males (11).

The ages of the patients ranged from 10 to 67 years (Figure 2). When the age distribution was considered, *P. aeruginosa* infections were more frequently seen in the ages between 10–20 and over 60 years. Our results are likely similar to other studies that

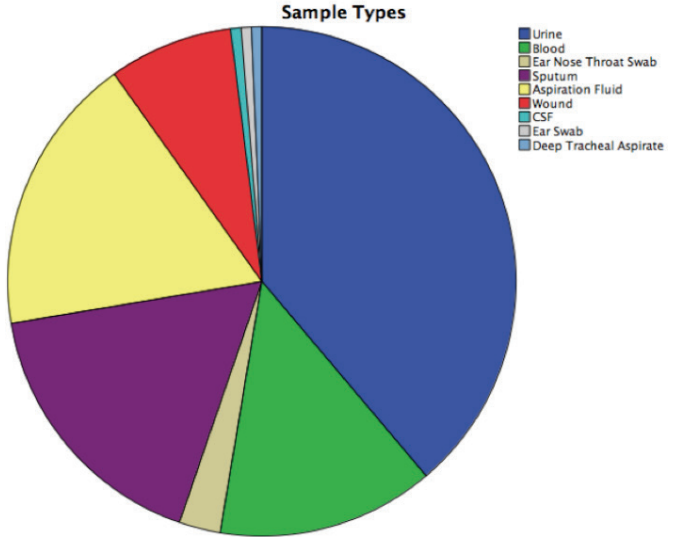

Figure 1. Distribution of *P. aeruginosa* among different samples

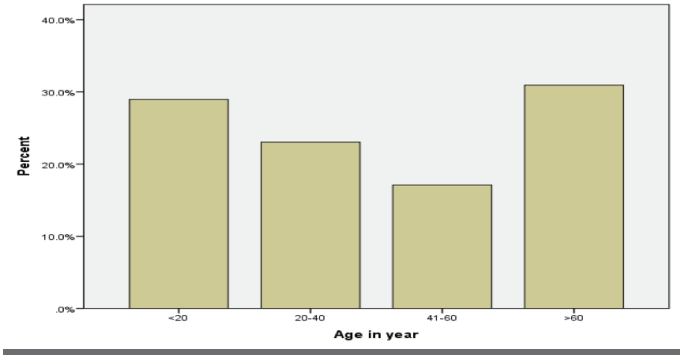

Figure 2. *P. aeruginosa* isolates from different age groups

TABLE I. Antimicrobial susceptibility pattern of *Pseudomonas aeruginosa*

Drugs	Total	Sensitive Count (%)	Intermediate Count (%)	Resistant Count (%)
Amikacin	151	123 (81.5)	8 (5.3)	20 (13.2)
Ampicillin/Sulbactam	151	2 (1.3)	-	149 (98.7)
Aztreonam	152	76 (50.0)	18 (11.8)	58 (38.2)
Cefepime	148	116 (78.4)	19 (12.8)	13 (8.8)
Cefoxitin	151	8 (5.3)	-	143 (94.7)
Ceftazidime	150	114 (76.0)	5 (3.3)	31 (20.7)
Ceftriaxone	113	7 (6.2)	-	106 (93.8)
Ciprofloxacin	149	109 (73.2)	5 (3.4)	35 (23.5)
Colistin	113	98 (86.7)	-	15 (13.3)
Gentamicin	146	111 (76.0)	8 (5.5)	27 (18.5)
Levofloxacin	113	83 (73.5)	4 (3.5)	26 (23.0)
Imipenem	151	122 (80.8)	7 (4.6)	22 (14.6)
Meropenem	149	130 (87.2)	6 (4.0)	13 (8.7)
Piperacillin/Tazobactam	148	135 (92.5)	-	17 (11.5)
Ticarcillin/ Clavulanate	144	64 (86.6)	-	46 (42.6)
Trimethoprim/ Sulfamethoxazole	150	7 (4.7)	1 (.7)	142 (94.7)

found 43.5% *P. aeruginosa* infection in the age range of 1–15 years. Studies reported that incidence of *P. aeruginosa* increased with age and was higher in males than females (11, 12). Ak et al. (6) reported that the highest resistance was observed for levofloxacin (13%) whereas the lowest resistance was observed for amikacin (3%) in Malatya. Aykan and Çiftçi (13) investigated the antibiotic resistance of *P. aeruginosa* isolates over 11 years (2003–2013) in Turkey. They reported that carbapenem resistance like imipenem resistance increased between 2007 and 2009; otherwise, these changes were not statistically significant for imipenem or meropenem. Resistance to aminoglycoside antibiotics such as amikacin, gentamicin, netilmicin, and tobramycin was decreased, but these changes were not found statistically significant. These study groups concluded that the antibiotic resistance in *P. aeruginosa* has started to decrease in Turkey.

In our study, the antimicrobial susceptibility test revealed that *P. aeruginosa* was highly sensitive to most of the antibiotics tested. Piperacillin/tazobactam was the most effective drug against all *P. aeruginosa* strains (92.5% sensitivity) followed by meropenem (87.2%), colistin (86.7%), ticarcillin/clavulanate (86.6%), amikacin (81.5%), and imipenem (80.8%). On the other hand, *P. aeruginosa* had 98.7% resistance to the antibiotic ampicillin/sulbactam and the other high resistance rates were observed for cefoxitin (94.7%), trimethoprim/sulfamethoxazole (94.7%), and ceftriaxone (93.8%) (Table 1). Ruh et al. (14) studied the antibiotic resistance of *P. aeruginosa* between 2010 and 2014 in Northern Cyprus and reported that the resistance rate for aztreonam was 42.9% and susceptibility rate for levofloxacin was 79.8%. According to our results, the resistance rate for aztreonam (38.2%) and the susceptibility rate for levofloxacin (73.5%) decreased in a one year period (2014–2015) in Northern Cyprus. In the present study, aminoglycoside (gentamicin 18.5%; amikacin 13.2%), fluoroquinolone (ciprofloxacin 23.5%; levofloxacin 23%), and ceftazidime (20.7%) resistance among *P. aeruginosa* isolates were found to be higher with the rates reported in the antimicrobial resistance report of Southern Cyprus between 2012 and 2015 (15).

The present study shows that the clinical isolates of *P. aeruginosa* are resistant to commonly used antibiotics and achieve more resistance to the newest antibiotics. In Cyprus, piperacillin/tazobactam, and meropenem are the most effective agents against *P. aeruginosa* whereas ampicillin/sulbactam, cefoxitin, and trimethoprim/sulfamethoxazole have the highest resistance. The knowledge of local antimicrobial resistance patterns is necessary to guide empirical therapy applied by practicing physicians, medical microbiologists, and public health officials.

Ethics Committee Approval: Ethics committee approval was not taken due to in vitro design of the study.

Informed Consent: N/A.

Author Contributions: Concept: M.G., K.S.; Design: K.S., A.A.; Supervision: K.S.; Resource: K.S., E.G.; Materials: E.G., M.G.; Data Collection and/or Processing: E.G., M.G., A.A.; Analysis and/or Interpretation: A.A., E.G., K.S.; Literature Search: E.G., M.G.; Writing; E.G., M.G.; Critical Reviews: K.S., M.G., A.S., E.G.

Conflict of Interest: The authors have no conflicts of interest to declare.

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