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Original Article

Antimicrobial Susceptibility Pattern of Clinical Isolates of *Pseudomonas Aeruginosa* in a Pediatric Hospital

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ABSTRACT

Background and Objective: ***Pseudomonas aeruginosa*** **has become a frequent cause of nosocomial infections, particularly in intensive care units (ICUs). Many reports have documented high rates of resistance in this species to commonly-used broad-spectrum antibiotics. The aim of this study was to assess the in vitro activity of some antibiotics against** ***Pseudomonas aeruginosa*** **strains to determine the susceptibility patterns of isolates to different antibiotics.**

Materials and Methods: **A total of 233** ***Pseudomonas aeruginosa*** **isolates obtained from various clinical specimens of hospitalized children in Ali-Asghar hospital of Tehran (Iran) were considered for susceptibility test. These strains were tested against 12 different antibiotics by a disk diffusion method. Of these isolates, 33.9% were from trachea, 31.8% from urine, 6.9% from eye, 5.2% from blood, 5.1% from ear, 1.3% from cerebrospinal fluid, 1.2% from stool, and 14.6% from other sites. In addition, 48.5% of** ***P. aeruginosa*** **strains were isolated from patients in ICUs.**

Results: **The most active antimicrobials were amikacin and other active compounds were gentamicin, ceftazidime, and ciprofloxacin respectively. Isolates from ICUs were more resistant to amikacin and gentamicin as compared to those from non-ICU wards ($p<0.05$). Isolates from trachea were more resistant to amikacin, gentamicin, ciprofloxacin and ceftazidime than those from other sites ($p<0.05$).**

Conclusion: **Our study showed that amikacin was the most active agent against** ***P. aeruginosa*** **followed by gentamycin, ceftazidime, and ciprofloxacin. According to our in vitro study results, active antibiotic susceptibility testing and surveillance should be continued in order to curtail the problem of antibiotic resistance.**

Key words: ***Pseudomonas aeruginosa*****, Antibiotic, Resistance, Disk Diffusion Antimicrobial Tests**

Introduction

Pseudomonas aeruginosa remains an important cause of hospital-acquired infections, particularly among immunosuppressed patients. According to the National Nosocomial Infections Surveillance (NNIS) System, between 1992 and 1999, *P. aeruginosa* was the second most common cause of pneumonia, the fourth most common cause of urinary tract infection, and the sixth most common bloodstream isolates in intensive care units (ICUs) (1, 2).

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This organism shows intrinsic and acquired resistance to many antibiotics and previous exposure to antibiotics leads to multidrug-resistant strains. Data from NNIS and the Intensive Care Antimicrobial Resistance Epidemiology (ICARE) project on the resistance of *P. aeruginosa* between 1998 and 2002 showed that the pooled mean figures for resistance to piperacillin, ceftazidime, imipenem, and ciprofloxacin increased from previous years and represented 14.3, 10.5, 13.7 and 28.9% of ICU-associated isolates respectively. The emergence of multi-drug resistant strains has been associated with increase in secondary bacteremia and mortality and has led in some cases to longer hospital stays and increased hospitalization costs (3).

There are a limited number of antimicrobial agents with reliable activity against *P. aeruginosa* including antipseudomonal penicillins and cephalosporines, carbapenems, and fluoroquinolones, particularly ciprofloxacin. For each of these agents, emergence of resistance during therapy has been described. Therefore, therapy for *P. aeruginosa* infections should consist of antimicrobial agents selected on the basis of extended susceptibility testing (4-8). Different resistance profiles are due to the diversity of clinics and the regional variations in antibiotic protocols (9).

The aim of this study was to assess the in vitro activity of some antibiotics against *Pseudomonas aeruginosa* strains to determine the susceptibility patterns of isolates to different antibiotics.

Patients and Methods

This cross-sectional study was carried out in Ali-Asghar hospital in Tehran (Iran). The study was conducted with the approval of the institutional review board of Iran University of Medical Sciences. Informed consent was obtained from the patients' parents or guardian. Totally, 233 consecutive isolates were obtained from various clinical specimens of hospitalized children. Of these isolates, 33.9% were from trachea, 31.8% from urine, 6.9% from eye, 5.2% from blood, 5.1% from ear, 1.3% from cerebrospinal fluid, 1.2% from stool and 14.6% from other sites. In addition, 48.5% of *P. aeruginosa* strains were isolated from patients in ICUs; the remainders were from other units as follows: 9% from nephrology ward, 6.9% from hematology ward, 12.4% from infectious ward, 5.2% from internal ward, 9% from surgical ward and 9.9% from outpatient clinic.

All of the isolates were identified as *P. aeruginosa* by positive reaction to oxidase, catalase, growth at 42 ^{o}C, and production of pyocyanin on pseudomonas P agar (Difco).

Susceptibility to 12 antimicrobial agents (nalidixic acid, cefotaxime, cephalothin, trimethoprim-sulfamethoxazole, nitrofurantoin, cephalexin, amikacin, ciprofloxacin, ceftazidime, gentamicin, ceftriaxone and cefepime) (Padtan teb, Tehran, Iran) was determined by the standard disk diffusion technique in accordance with the recommendations of National Committee for Clinical laboratory standards (10). Mueller-Hinton plates were incubated for 24 h after inoculation with organisms and placement of the disks and zones of inhibition were measured.

The results of the susceptibility testing were classified into three categories, i.e susceptible (sensitive), intermediate and resistant.

Statistical analysis:

Data were collected and the statistical analysis was performed using SPSS (version 11.5; SPSS Inc., Chicago, IL, USA). Values were tested for statistical significance using chi-square test. A p-value of 0.05 or less was considered significant.

Results

Totally, 233 specimens were obtained from 233 hospitalized children (130 boys and 103 girls). Seventy seven (33%) of the patients were under 1 month, 5 (2.1%) cases were from 6 months to 1 year, 12 cases (5.2%) between 1 and 2 years, 74 cases (31.8%) between 2 and 5 years, 50 cases (21.5%) between 5 and 10 years and 15 (6.4%) cases between 10 and15 years. The highest percentage rates of resistance were found for cephalexin (100%), cephalothin (97.8%), trimethoprim-sulfamethoxazole (96.5%), nitrofurantoin (95.2%), cefotaxime (92.5%), and naldixic acid (90.77%). Amikacin was the most active compound (sensitivity: 60.8%). Other active compounds were gentamicin (49.77%), ceftazidime (34.5%) and ciprofloxacin (33%) respectively. Table 1 shows the frequency of susceptibility to active compounds including amikacin, gentamicin, ceftazidime, and ciprofloxacin by age. Organisms isolated from newborns were the most resistant isolates to amikacin and ciprofloxacin ($p<0.05$).

The frequency of susceptibility to amikacin, gentamicin, ceftazidime, and ciprofloxacin was compared between the wards. Isolates from ICUs were more resistant to amikacin and gentamicin than those from non-ICU wards ($p<0.05$) (Table 2). Isolates

from trachea were significantly more resistant than those from other sites to amikacin, gentamicin, ciprofloxacin and ceftazidime.

Table 1. Frequency of *P. aueruginosa* susceptibility to amikacin, gentamicin, ceftazidime and ciprofloxacin by age in hospitalized children

Age group	Amikacin			Gentamicin			Ceftazidime			Ciprofloxacin		
	Sensitive	Intermediate	Resistant	Sensitive	Intermediate	Resistant	Sensitive	Intermediate	Resistant	Sensitive	Intermediate	Resistant
Newborn (≤1 month)	35	5	33	25	1	47	17	8	28	1	1	23
1 m–1 y	3	1	1	3	0	2	0	0	1	0	0	2
1–2 years	9	2	1	6	1	5	4	0	4	2	0	5
2–5 years	44	11	14	42	2	26	12	2	26	16	0	19
5–10 years	31	4	8	24	2	18	13	0	17	11	1	13
10–15 years	10	0	5	8	1	4	4	1	8	3	1	2

Table 2. Frequency of P. aueruginosa susceptibility to amikacin, gentamicin, ceftazidim, and ciprofloxacin by wards in hospitalized children

Wards	Amikacin			Gentamicin			Ceftazidime			Ciprofloxacin		
	Sensitive	Intermediate	Resistant	Sensitive	Intermediate	Resistant	Sensitive	Intermediate	Resistant	Sensitive	Intermediate	Resistant
ICU	46	12	47	40	1	64	20	8	45	7	1	35
Non-ICU	86	11	15	68	6	38	30	3	39	26	2	29

Discussion

Pseudomonas aeruginosa has become a frequent cause of nosocomial infections, particularly in intensive care units (ICUs). Many reports have documented high rates of resistance in this species to commonly used broad-spectrum antibiotics. Because of the widespread use of antibiotics, especially in developing countries, the resistance profile of microorganisms is changing. *Pseudomonas aeruginosa* is naturally resistant to ß-lactams, quinolones, chloramphenicole and tetracyclines, mainly because of the very low permeability of their cell wall, production of cephalosporinase, active efflux, and poor affinity for the target (DNA gyrase) (11-15). Thus, treatment of Pseudomonas infections with many available drugs would be difficult due to this resistance. Consequently, local surveillance with antibiograms should be implemented to guide the current use of antibiotics.

Intensive care patients create an environment for infection because of the debilitating effect of a prolonged hospitalization and the application of medical equipment (9). The majority of strains tested at this center came from ICUs (NICU and PICU). Ergin et al (9) showed that the rate of *Pseudomonas* infections in ICU was higher than in non-ICU in their hospital; also they found that tracheal aspirate was the most important source of *Pseudomonas* sp. in internal wards of ICU ($p<0.05$). Our results regarding the higher rate of Pseudomonas infections in ICU and tracheal specimens were in concordance with Ergin et al results (9). In Raja et al (16) study, the rates of antimicrobial resistance of isolates were 6.73% to amikacin, 12.9% to gentamicin, 10.1% to netilmicin, 10.9% to ceftazidime, 11.3% to ciprofloxacin, 9.9% to imipenem, 10.8% to piperacillin, 9.4% to piperacillin-tazobactam and 0% to polymyxin B. Out of the 505 isolates, 29 (5.74%) were found to be multidrug-resistant; these were most commonly isolated from respiratory tract specimens of patients in surgical units, followed by respiratory tract specimens in patients in

medical units. Our study also showed that amikacin was the most active agent against *P. aeruginosa* and respiratory tract specimens were significantly more resistant than those from other sites. An Iranian study by Haddadi et al showed that the susceptibility rates among isolated *P. aeruginosa* were 75% for imipenem and 39% for ciprofloxacin (17). Nikbin et al reported that the resistance rates of *P. aeruginosa* isolates to 13 different antibiotics were as follow: ceftizoxime (99%), lomefloxacin (94.3%), ceftazidime (59.6%), ticarcillin (50%), ceftriaxone (44.3%), cefoperazone (37.5%), tobramycin (34.6%), piperacillin and gentamicin (33.7%), carbenicillin (25%), amikacin (22%), ciprofloxacin (15.4%) and imipenem (2.9%) (18). Aminoglycosides are frequently used as part of combination regimens for treatment of pseudomonal infections but are generally not recommended as single drugs (4,19). In this study, amikacin and gentamicin were found as effective agents. This fact reflects the importance of controlling the use of these antimicrobials in the hospital units for preventing the emergence of aminoglycosides-resistant strains.

Conclusion

This study examined the resistance profile of *P. aeruginosa* in Ali-Asghar hospital in Tehran and found that the level of resistance to antibiotics was high. Our study also showed that amikacin was the most active agent against *P. aeruginosa* followed by gentamycin, ceftazidime and ciprofloxacin. According to our in vitro study results, active antibiotic susceptibility testing and surveillance should be continued in order to curtail the problem of antibiotic resistance.

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