Multi-drug resistant *Pseudomonas aeruginosa* isolates from Kermanshah hospitals

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**ABSTRACT**

**Background and Objective:** The antibiotic resistance of pathogenic strains of *P. aeruginosa* is increasing and the treatment of *Pseudomonas* infections has become a medical concern. We aimed to test the antibiotic resistance among pathogenic isolates of *P. aeruginosa*.

**Materials:** In this study 96 pathogenic strains of *P. aeruginosa* were collected from Imam Khomainy and Imam Reza hospitals in Kermanshah during 2011-2012. After identification, the antibiotic susceptibility of bacteria was carried out using disc diffusion method according to the Clinical and Laboratory Standards Institute recommendations.

**Results:** The average age of patients was 34.7±22.5 year. Among isolates, 28 (29.1%) were resistant to all antibiotics, but only 10 (10.4%) were sensitive to all antibiotics. The highest sensitivity was for aminoglycosides in particular gentamicin and the lowest sensitivity was for carbenicillin and aztreonam.

**Conclusion:** The results of this study warn for the increasing of multidrug and extensively drug resistant strains in Kermanshah city. Therefore the continuous surveillance for antibiotic resistance can help to better understand the antibiotic susceptibility of this opportunistic pathogen to the current drugs. The results of this study suggest that from anti-*Pseudomonas* penicillins, piperacillin/tazobactam and from aminoglycosides, gentamicin and amikacin, are effective drugs and can be used for treatment of *Pseudomonas* infections.

**Keywords:** antibiotic resistance, *Pseudomonas aeruginosa*, Kermanshah

**Introduction**

*Pseudomonas aeruginosa* is a gram-negative, aerobic, non-fermentative, non-spore-forming, motile, catalase and oxidase positive bacterium. It produces pyocyanin pigment to make solid media green-blue color and a special odor that smells like a grape. This organism is ubiquitous in nature and frequently isolated from various hospital places, especially in area that are moist, such as ventilation pipes, toilets and pots, as well as medical supplies, nurses and other hospital staff (1, 2). *P. aeruginosa* is one of the most important opportunistic pathogen for hospital infections, in particular in burn units and causing severe infections (3, 4).
One of the major concerns for the treatment of *P. aeruginosa* infections is antibiotic resistance, since studies showed that this bacterium has increasingly been resistant to many antibiotics (5). It is naturally resistant to narrow spectrum penicillins, first and second-generation cephalosporins, sulfonamides and trimethoprim (6). The antibiotics normally used for the treatment of Pseudomonas infections are broad spectrum penicillins such as ticarcillin and piperacillin, cephalosporins such as ceftazidime and cefepime, carbapenems, aminoglycosides, fluoroquinolones and aztreonam (7, 8). However, *P. aeruginosa* strains that are resistant to one or more of these drugs is increasing worldwide. These strains acquire resistant genes mainly on extra chromosomal DNA in particular plasmid (9-11).

Study on resistant pattern and explore the emerging resistance in this opportunistic pathogen can provide a better understanding of regional epidemiology. Therefore, this study aimed to test the antibiotic resistance of hospital isolates of *P. aeruginosa* in Kermanshah.

**Materials and Methods**

During 2011 and 2012, 96 pathogenic *P. aeruginosa* isolated from patients, consisting of 50 men (52.1%) and 46 women (47.9%). The 65 (67.7%) and 31 cases (0.332%) were from Imam Khomeini and Imam Reza hospital, respectively. They were from various hospital places including burn unit, surgery and infectious wards. The samples were aseptically transferred to the Medical Microbiology Laboratory. The bacteria were then cultured on MacConkey and Muller-Hinton agar media and the grown colonies were used for differential diagnosis using specific tests for the identification of bacterial species, including catalase, oxidase, the growth characteristics in TSI, SH2, pigment and gas production, indole and methyl Red, SIM (Sulfide-Indole-Motility) and Growth at 42 °C on cetrimide agar. On the cetrimide agar the pigment production by bacteria was also determined. Following the identification of bacteria, standard disk diffusion method with MAST (UK) disks was carried out according to the CLSI (Clinical and Laboratory Standards Institute) guidelines. The susceptibility of bacteria was assessed to three major groups of antibiotics, namely aminoglycosides (amikacin, gentamicin, tobramycin and netilmicin), fluoroquinolones (ciprofloxacin and levofloxacin) and beta-lactams (piperacillin / tazobactam, ceftazidime, carbenicillin, cefepime, meropenem, imipenem and aztreonam). The accuracy of results was confirmed using standard strains of *P. aeruginosa* PTCC 1430 (Persian type culture collection) and ATCC 27853. The information of patients including age, sex and type of infections were also recorded.

**Statistical Analysis**

The data from the bacteriology results of isolates was analyzed using SPSS V16 software and Prism V4 Graph Pad.

**Results**

The majority of patients was young and middle aged (Figure 1). The average age of patients was 34.7± 22.5 years. The burn infection with 41 cases (42.7%) was the most prevalent followed by urinary tract infection 18 (18.8%), pneumonia 17
(17.7%), and others (Figure 2). Of the isolates, 28 strains (29.1%) were resistant to all antibiotics whereas only 10 strains (10.4%) were susceptible to all antibiotics. The least resistance was for aminoglycosides, especially gentamicin (Figure 3). The frequency of isolates on the basis of colony pigmentation was green pigment 35 cases (36.5%), light green 30 (31.2%) Green - Blue 20 (20.8%) and phosphorus yellow 11 (11.5%). Significant association between antibiotic resistance and the colony pigmentation was not observed (p=0.126). Between men and women in terms of antibiotic resistance, there was no significant difference (p = 0.236).

Figure 1. The frequency of patients according to their age

Figure 2. The frequency of patients in two hospitals and their source of infections
Overview of antibiotic sensitivity of isolates

Discussion

Although many studies on antibiotic resistance of *P. aeruginosa* have been done worldwide, because of the ability of this opportunistic pathogen in acquisition of new resistant traits to inactive antibiotics, continuous surveillance for local drug resistance is necessary. On the other hand, the antimicrobial susceptibility of microorganisms can be affected by the characteristics of regional pathogens. Our results showed that a high percentage of the isolates were resistant to all antibiotics that are higher than average rate in previous Iranian studies. For example, in a study during 1998 in Iran, only five percent of *P. aeruginosa* isolates were resistant to all antibiotics (11). The increase of multi-drug-resistant (MDR) and extensively drug-resistant (XDR) strains is a warning sign for treatment of *P. aeruginosa* infections.

Moreover, isolates in this study showed higher sensitivity to aminoglycosides, especially gentamicin and amikacin which is consistent with the results of other studies in Kermanshah and Urumia in Iran (7, 12). But the sensitivity of strains was lower than the results reported by other Iranian studies including in Kerman and Tehran (3, 13) that may indicate the increasing of drug resistant among isolates.

On the other hand, our result for resistant rate was higher than resistance reported from isolates in cystic fibrosis patients (14). One explanation could be that these patients have an impaired immune system therefore the low virulence strains can cause infections. To confirm this notion, a German study on comparison of resistance in isolates of cystic fibrosis and non-cystic fibrosis patients showed the strains isolated from cystic fibrosis patients had lower antibiotic-resistant (15). The resistance to
aminoglycoside in our study was also higher than the results of a study conducted in China on 570 clinical strains and the results of a study in Taiwan (16, 17). The results of our study suggest that the overall sensitivity to the third generation of cephalosporins was higher than the results of studies in other cities of Iran (3, 7, 12, 18, 19), but it was lower than the sensitivity reported from research in other countries (17). This difference may be related to factors such as antibiotic usage patterns in each region, type of infection that the bacteria isolated from, the method used and the sample size.

Our results also indicated that susceptibility to fluoroquinolones is relatively low, which is consistent with the results reported by a few studies in Iran (7, 12), but some other studies in Iran have reported higher resistant rate (3, 14). In other countries such as China and Spain the resistance to fluoroquinolones has been reported much lower than out results (16, 20).

Our results showed piperacillin/tazobactam was the most effective antibiotic for *P. aeruginosa* that is similar to the results of a study in Iran (7). However, comparing to the results of other studies the resistance of isolates to this antibiotic has been increased (14, 15, 17).

In the present study, no significant association between bacterial pigmentation with antibiotic-resistance and the type of infection was found. For a better understanding of the relationship between pigmentation and the phenotypic characterization of pathogenic strains of *P. aeruginosa*, further studies are needed.

In conclusion, in compare with the results from other studies, our results suggest that the resistant strains of *P. aeruginosa* in Kermanshah has increased probably due to factors such as genetic mutations, environmental conditions and inappropriate consumption of antibiotics. These differences are particularly marked in the case of beta-lactam and fluoroquinolones antibiotics that could be suggestive of increased antibiotic resistance in Kermanshah. What seems to be an alarming in this study is the increase of MDR and extensively drug resistant (XDR) and pandrug-resistant (PDR) isolates. Therefore, continuous monitoring of antibiotic resistance for this opportunistic pathogen can contribute to a better understanding of its susceptibility to available antibiotics in this region. In this regard, the present study suggests that piperacillin / tazobactam, gentamicin and amikacin have a good effect on *P. aeruginosa* strains.

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**References**


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