Original article

**Pseudomonas aeruginosa** infection in an intensive care unit

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**Abstract**

*P. aeruginosa* is a well-known cause of hospital-acquired pneumonia (HAP) in intensive care units (ICU). We conducted an epidemiologic and molecular investigation of endemic *P. aeruginosa* infection in an ICU. *P. aeruginosa* strains isolated from hospitalized patients and environmental samples in the ICU of the National Medical Center (NMC) were collected by the hospital infection laboratory of National Center for Disease Control and Public Health, October 2005 - April 2007. The antimicrobial susceptibility of the isolates in vitro was assessed by an agar disk diffusion method, as recommended by the Clinical and Laboratory Standards Institute. The antimicrobial susceptibility data were analyzed using WHONET software. Isolates resistant to cefepime, imipenem, aztreonam, ciprofloxacin, piperacillin, and gentamicin were defined as multidrug resistant (MDR).

*P. aeruginosa* was isolated in 89 specimens obtained from 53 patients with HAP. The incidence rate of MDR *P. aeruginosa* infection was 15,8/100 patient admissions per year. MDR strains were common, making up 28/89 (31,5%) of all *P. aeruginosa* isolates in this study. In March - August 2006 there was an outbreak of HAP caused by *P. aeruginosa*. During the outbreak *P. aeruginosa* was isolated from 25 patients with HAP. Using PFGE typing, it was observed that twelve *P. aeruginosa* had the same genetic pattern. Environmental investigations demonstrated the presence of *P. aeruginosa* in the ventilation equipment. In one case, MDR *P. aeruginosa* was found in the microfilter of an AV machine. The most predominant etiological factor responsible for HAP in ICU of NMC was endemic *P. aeruginosa*.

**Introduction**

Intensive care units (ICUs) are characterized by an extremely high risk of nosocomial infections (NI). NI levels in this kind of unit is several times higher than other departments.¹⁰,¹² Hospital-acquired pneumonia (HAP) is a serious problem in ICUs, leading to lengthened hospital stays, higher health-care costs. HAP is an important factor morbidity and mortality of patients in ICUs.²⁷,³⁵ *P. aeruginosa* is a well-known cause of HAP in intensive care units.¹ Respiratory tract colonization with *P. aeruginosa* may develop from endogenous sources or from exogenous sources such as contaminated equipment or other patients infected with *P. aeruginosa*.³⁵,¹¹ The situation for patients with *P. aeruginosa* infections is a problem since this microorganism is inherently highly resistant to many antibiotic classes.⁴

**Methods**

We conducted an epidemiological and molecular investigation of endemic *P. aeruginosa* infection in the ICU of Georgian National Medical Center, Tbilisi, Georgia. All patients admitted to ICU were studied prospectively. De-
tained demographic and epidemiological data were collected from each patient. Study of risk factors with MDR *P. aeruginosa* infection was done by a case-control study. An odds ratio (OR) and 95% confidence interval (CI) were calculated to evaluate the strength of any association. Frequency comparisons were performed by the chi-square test. The statistical analyses were performed using Microsoft Access software. Infections that developed 48 hours after admission into the ICU were considered ICU acquired. Ventilator-associated pneumonia (VAP) was defined according to criteria proposed by an international panel. *P. aeruginosa* strains isolated from hospitalized patients and environmental samples in a 10-bed ICU of NMC were collected by the hospital infection laboratory of Georgian National Center for Disease Control and Public Health, from October 2005 through April 2007. The antimicrobial susceptibility of the isolates in vitro was assessed by an agar disk diffusion method, as recommended by the Clinical and Laboratory Standards Institute (formerly NCCLS). The antimicrobial susceptibility data were analyzed using WHONET software. Isolates resistant to cefepime, imipenem, aztreonam, ciprofloxacin, piperacillin, and gentamicin were defined as multidrug resistant (MDR). Acquired strains were genotypically characterized by pulsed-field gel electrophoresis (PFGE).

**PFGE:** The bacteria were harvested by centrifugation and then suspended to an optical density at 600 μl in EEH buffer (100 mM EDTA, 10 mM EGTA, 10 mM HEPES, pH 8.0). The suspension was mixed with an equal volume of 1.2% SeaKem Gold agarose (SeaKem GTG; FMC Bioproducts) in EEH buffer. The plugs were chilled to 4°C for 30 min. Plugs with immobilized bacteria were incubated overnight at 37°C with EEH buffer containing 1 mg of proteinase K per ml and 1% sodium dodecyl sulfate. Plugs were washed six times at room temperature six times for 30 min in TE buffer (10 mM Tris-HCl, 1 mM EDTA, pH 8.0) with gentle mixing. One-sixth of the plug was cut and incubated with 10 U of SpeI (NEB) for 2 hours at 37°C. The plugs were placed in a 1% agarose gel in 0.5×TBE (45 mM Tris, 46 mM boric acid, 1 mM EDTA). PFGE was performed at 14°C in a CHEF-DR II system (Bio-Rad) at 200V with switch times that ranged from 5 to 15 s for 10 hours followed by switch times that ranged from 15 to 45 s for 10 hours. Gels were stained with ethidium bromide. DNA banding patterns were photographed after transillumination with UV light.

**Results**

*P. aeruginosa* was isolated from 89 infected patients (76 respiratory tract, 5 wound, 3 drainage, 1 urine, 1 blood, 1 pus 1 eye and 1 oral cavity) obtained from 53 patients with HAP. *P. aeruginosa* strains were resistant to chloramphenicol (98.9%), aztreonam (69.6%), gentamicin (87.6%), amikacin (78.7%), clindamycin (100%), ciprofloxacin (71.9%), imipenem (68.5%), piperacillin (70.8%), nitrofurantoin (100%), tetracycline (98.9%), tobramycin (79.8%), trimethoprim (97.7%), carbenicillin (85.4%), cefepime (45.0%) and amoxicillin/clavulanic acid (81.0%). MDR strains were common, making up 28/89 (31.5%) of all *P. aeruginosa* isolates in this study. Most MDR *P. aeruginosa* came from respiratory tract isolates (24/28; 82.1%). All patients with MDR *P. aeruginosa* were infected during their hospitalization. The incidence rate of MDR *P. aeruginosa* infection in ICU was 15,8/100 patient admissions per year. Almost twice as many male as female patients had HAP with MDR *P. aeruginosa*. Most of the patients were 35-50 year old (median, 43; range, 16-78). Mechanical ventilation (MV) was a significant risk factor for HAP with MDR *P. aeruginosa* (OR = 9.71; CI = 2.19–43.03). Prolonged (> 72 h) MV was the most important risk factor to be VAP with MDR *P. aeruginosa*. VAP with MDR *P. aeruginosa* developed in 43.75%; 14/32 (OR = 4.86; CI = 1.71–12.66) patients who required > 72 h of MV.

From March through August 2006 there was an outbreak of HAP caused by *P. aeruginosa*. During the outbreak *P. aeruginosa* was isolated from 25 patients diagnosed with HAP. Using PFGE typing, it was observed that twelve MDR *P. aeruginosa* isolates had the same genetic pattern. Environmental investigations demonstrated the presence of *P. aeruginosa* in the ventilation equipment. In one case, MDR *P. aeruginosa* was found in the microfilter of AV machine.

**Discussion**

Based on the study findings, the resistance of *P. aeruginosa* strains for most of antibiotics is relatively higher than in EU countries. For example, the percentage of *P. aeruginosa* strains resistant to piperacillin was high (70.8%) compared with data of studies performed in Europe (4,4–26,2%).

High resistance of *P. aeruginosa* strains to cephalosporins and, at the same time, low resistance to cefepime might be explained by the hyper-production of plasmid β-lactamases type PSE-1 and PSE-2.

In the ICU patients were infected with MDR *P. aeruginosa* by both endogenous and exogenous pathways. In most cases, infections were caused due to patients prolonged artificial ventilation (AV). All patients infected with genetically similar strains were under the artificial ventilation and for 41.7% of those patients used an AV machine from which MDR *P. aeruginosa* strains were isolated.

**Conclusions**

The most predominant etiologic factor responsible for HAP in the ICU of NMC was endemic MDR *P. aeruginosa*. Decreasing the incidence of MDR *P. aeruginosa* infection in ICU was achieved through the implementation of an effective infection control system.
References


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