Trends in Antimicrobial Resistance of *Acinetobacter baumannii* isolates from a Metropolitan Detroit Health System

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Key Words: Acinetobacter; Antimicrobial resistance; Multi-drug resistance; Nosocomial infections; Carbapenem.

Word count: Abstract: 73 Text: 1308

Running title: MDR *A. baumannii* in Detroit metropolitan
Abstract

A phenotypic and genotypic analysis of *Acinetobacter baumannii* was conducted from 2003-2008 in Detroit, MI. Incidence of *A. baumannii* increased from 1.7 to 3.7/1,000 patient days during the study period. Susceptibility to ampicillin-sulbactam and imipenem decreased from ~90% to ~40%. Genotyping revealed polyclonality, suggesting either emergence of multiple resistant strains or spread of a common genetic element. The sharp rise mandates major multidisciplinary interventions to optimize management of this multi-drug resistant pathogen.
*Acinetobacter baumannii* is an increasingly common nosocomial pathogen distributed worldwide (17, 28). Infections caused by *A. baumannii* are associated with adverse clinical outcomes, including high rates of morbidity and mortality, prolonged hospital stay, and substantial health care expenses (3, 4, 15, 19, 28). Over the past decade, increasingly resistant strains of multi-drug resistant *A. baumannii* (MDR-AB) have emerged. The prevalence of strains resistant to the usually potent and safe β-lactam antibiotics, such as ampicillin-sulbactam and carbapenems had increased substantially (5, 11, 12, 23). Infection with MDR-AB is associated with even worse outcomes compared to infections due to non-MDR-AB (9, 27). Of particular concern, strains of MDR-AB are now being encountered that are resistant to all commonly used antibiotics, including tigecycline and colistin (20).

The Detroit Medical Center (DMC) health system consist of 8 hospitals, has over 2,000 beds, and serves as a tertiary referral hospital for the metropolitan Detroit area and southeastern Michigan. *A. baumannii* has been a prominent pathogen in southeastern Michigan for several years (13). The objectives of this study were to conduct a retrospective analysis of the trends in prevalence, resistance, and clonality, among clinical isolates of *A. baumannii* at DMC, with a particular focus on the emergence of strains resistant to all available treatment options.

The DMC has a single centralized Clinical Microbiology Laboratory, which process ~500,000 samples annually. *A. baumannii* was recovered from samples by using the MicroScan® automated system, and a designated panel for *A. baumannii* was utilized to determine susceptibilities for all pre-defined antimicrobial agents. Beginning in 01/01/2008, all isolates that were resistant to ampicillin-sulbactam and/or to imipenem,
were tested routinely against colistin and tigecycline using the E-test method (AB Biodisk®, Solna, Sweden). All bacteriologic tests were standardized and performed according to the Clinical and Laboratory Standard Institutions (CLSI) criteria (1). The breakpoints of *A. baumannii* to tigecycline were determined by using the breakpoints of the European Committee on Antimicrobial Susceptibility Testing (2), since no CLSI criteria are available. An *A. baumannii* isolate was defined as MDR-AB if it was resistant to at least 3 representatives of different antibiotic classes (including broad-spectrum penicillins and cephalosporins, β-lactam/β-lactamase inhibitor combinations, fluoroquinolones, aminoglycosides, minocycline, and tigecycline), in addition to resistance to group 2 carbapenems (imipenem or meropenem) and ampicillin-sulbactam.

A retrospective analysis and review of all *A. baumannii* clinical isolates from 01/01/2003 to 12/31/2008 was conducted. Only unique clinical patient isolates were included. The susceptibility profiles, and the hospital where the isolate was recovered, were recorded. Typing MDR-AB isolates was conducted by using Pulsed-Field Gel Electrophoresis (PFGE) and repetitive extragenic palindromic (REP)-PCR, according to established methodological protocols (6, 24-26). SPSS 17 software (Chicago, IL, USA, 2008) was used for all statistical analyses and the Chi-Square for linear trend test was used for calculating trends over the study years.

During the six year study period (2003-2008), the total number of patients with *A. baumannii* isolates in DMC institutions had increased from 566 (1.7/1,000 patient days) in 2003 to 1239 (3.7/1,000 patient days) in 2008 (p <0.001). Table I depicts the prevalence of cases during the study years, and the susceptibility profiles. The susceptibility to nearly all tested antimicrobials decreased during the study period.
Notably, susceptibility decreased for the two most potent drugs used to treat *A. baumannii*, ampicillin/sulbactam and imipenem: susceptibility decreased from 89% and 99%, respectively, in 2003, to 40% and 42% of isolates in 2008 (p <0.001 for both trends). Of the 348 (28%) MDR-AB isolates from 2008, 280 (80.4%) were non-susceptible to tigecycline, 8 (2.7%) were non-susceptible to colistin and 3 (0.86%) were non-susceptible to both tigecycline and colistin. During 2008, the MIC$_{50}$ and MIC$_{90}$ for tigecycline and colistin were 4 µg/ml and 8 µg/ml, and 0.5 µg/ml and 1 µg/ml, respectively. A notable exception to the overall trend of reduced susceptibility to antibiotics during the study period was tobramycin – susceptibility increased from 41% to 65% during the study period. One isolate was resistant to all antimicrobials, including tobramycin.

Twenty-seven MDR-AB isolates from our health system in 2007-8 were genotyped using PFGE and REP-PCR. Results obtained using the PFGE genotyping and REP-PCR methods were completely concordant. As displayed in Figures 1 and 2, characteristic of the surge in MDR-AB at our health system was polyclonal in nature. More than 80% of the isolates analyzed clustered into 2 major clones as displayed in figure 1; 10 (38%) belonged to cluster I and 12 to cluster II (46%). The remaining isolates belonged to three different clusters: 1 to cluster III (4%), 2 to cluster IV (8%), and 1 to cluster V (4%).

This report describes a multi-institutional epidemic of MDR-AB at the DMC. The emergence of MDR-AB strains became clearly evident in 2007. MDR-AB organisms pose a treatment challenge because agents used to routinely treat *A. baumannii* (such as carbapenems and ampicillin/sulbactam) do not have in vitro activity and the only agents...
available with reliable in vitro activity are tobramycin and colistin, both of which have pharmacodynamic and toxicity-related limitations. Tobramycin was the only agent for which susceptibility rates increased during the study period. However, this aminoglycoside agent is nephrotoxic, has low tissue-penetration in ischemic tissues and its clinical role as a single therapeutic agent for the treatment of *A. baumannii* infections remains questionable (10). The utilization of tobramycin at our institutions did not change during the study period. The efficacy of colistin in treating MDR-AB infections has been mixed (8, 16, 21). Colistin is an old antimicrobial agent, which has not undergone rigorous pharmacokinetic and pharmacodynamic study. Therefore, strategies to maximize its efficacy and minimize its known renal and neurological toxicities remain unclear and need to be developed. Tigecycline, a glycylcycline, is a relatively new agent and is one of the few remaining options for the treatment of MDR-AB. It has a relatively safe therapeutic profile, but since greater than 80% of isolates at DMC in 2008 were nonsusceptible, as determined by E test (MIC$\geq$4 µg/ml), its therapeutic utility appears limited. Recent studies comparing the E-test with the broth microdilution (BMD) method, have reported major discordances in in-vitro susceptibility results, with E test often reporting much higher MICs (7, 22). Thus, clarifying the interpretation of in vitro susceptibility results to tigecycline, and determining the clinical efficacy of tigecycline in the treatment of infections caused by *A. baumannii* strains are important issues to address for DMC and other regions where MDR-AB is prevalent. In addition, establishing CLSI breakpoint definitions for tigecycline will help to establish uniformity in interpretation of susceptibility results.
The genotypic distribution of MDR-AB strains in DMC, as in other parts of the world, displayed polyclonal characteristics (14, 18). This highlights a relative discrepancy between the clinical manifestation of “classic” outbreaks, where a single clone is spread from patient-to-patient in a given location and time, and polyclonal outbreaks, where genotypically distinct strains contribute to outbreaks. This discrepancy may be attributed to the spread of a mobile genetic element among A. baumannii isolates although we could not investigate the presence of a mobile genetic element in this study (18). It should be emphasize though, that strains were collected for further molecular analysis by the microbiology laboratory upon request by infection control. These requests occurred only in the last 2 years of the study, when the surge in MDR-AB incidence was appreciated. The lack of availability of strains from our health system for genotypic comparison to more recent isolates is a weakness of the study.

Strict infection-control measures remain an important method for controlling the spread of MDR-AB in nosocomial settings, and the value of screening methods to detect asymptomatic carriage, enhanced barrier precautions and, cohorting of patients and staff in the control of spread of MDR-AB requires more investigation. In addition, antimicrobial stewardship strategies and clinical data assessing the effectiveness of various therapeutic options to treat A. baumannii infections are mandatory to improve management of infections due to MDR-AB.
References


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Table I: Prevalence and susceptibility trends of *Acinetobacter baumannii* at Detroit Medical Center, 2003-2008

<table>
<thead>
<tr>
<th>Year</th>
<th>No of Isolates</th>
<th>No/1,000 Pt.</th>
<th>Days</th>
<th>Imipenem</th>
<th>Amp/Sulbactam</th>
<th>Ceftazidime</th>
<th>Ciprofloxacin</th>
<th>TMP/SMX</th>
<th>Amikacin</th>
<th>Tobramycin</th>
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<tbody>
<tr>
<td>2003</td>
<td>566</td>
<td>1.7</td>
<td>99%</td>
<td>89%</td>
<td>36%</td>
<td>32%</td>
<td>33%</td>
<td>90%</td>
<td>41%</td>
<td></td>
</tr>
<tr>
<td>2004</td>
<td>593</td>
<td>1.7</td>
<td>97%</td>
<td>86%</td>
<td>43%</td>
<td>31%</td>
<td>31%</td>
<td>77%</td>
<td>36%</td>
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<tr>
<td>2005</td>
<td>890</td>
<td>2.8</td>
<td>99%</td>
<td>87%</td>
<td>28%</td>
<td>24%</td>
<td>26%</td>
<td>81%</td>
<td>28%</td>
<td></td>
</tr>
<tr>
<td>2006</td>
<td>751</td>
<td>2.3</td>
<td>99%</td>
<td>62%</td>
<td>26%</td>
<td>24%</td>
<td>27%</td>
<td>92%</td>
<td>56%</td>
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</tr>
<tr>
<td>2007</td>
<td>1175</td>
<td>3.6</td>
<td>65%</td>
<td>37%</td>
<td>16%</td>
<td>14%</td>
<td>17%</td>
<td>63%</td>
<td>60%</td>
<td></td>
</tr>
<tr>
<td>2008</td>
<td>1239</td>
<td>3.7</td>
<td>42%</td>
<td>40%</td>
<td>15%</td>
<td>15%</td>
<td>18%</td>
<td>33%</td>
<td>65%</td>
<td></td>
</tr>
</tbody>
</table>

*No= Number; Pt.= patient; Amp= ampicillin; TMP/SMX= trimethoprim/sulfamethoxazole*
Legend to Figure

1. Phylogenetic tree of 27 strains of MDR-AB isolated at DMC during the study period
2. Clone distribution according to REP-PCR of 25 MDR-AB strains
Figure 1:
Figure 2: