USA600 (ST45) Methicillin-Resistant *Staphylococcus aureus* Bloodstream Infections in Urban Detroit

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Methicillin-resistant *Staphylococcus aureus* (MRSA) has emerged as a major source of invasive infections, implicated in 18,000 deaths annually (9). Mortality rates of 20 to 30% have been reported in patients with MRSA bloodstream infection (BSI) with a recent study, spanning 15 years, reporting a mortality rate of approximately 28% (11, 12, 18). Recently, we reported 60% mortality in a small number of MRSA BSI infections caused by the USA600 strain type, suggesting this strain may have unique virulence characteristics (4). USA600 or ST45, first reported as an epidemic strain spreading throughout Germany and the Netherlands in the last decade, has not previously been associated with serious infection (20-22). Given our preliminary findings, we investigated a series of consecutive cases of USA600 MRSA BSI to describe patient, treatment and strain related characteristics of these infections.

Pulsed-field gel electrophoresis (PFGE) was performed on 420 consecutive MRSA bloodstream isolates and 16 patients with USA600 MRSA BSI were identified between 7/2005 and 7/2008 at a 900 bed tertiary care hospital in Detroit, Michigan (Figure 1). During the study period, 65% of all *S. aureus* infections were MRSA. The source of the BSI was identified by chart review using a combination of clinical and laboratory findings and other diagnostic tests according to CDC definitions (6). Epidemiologic classification was conducted based on presence or absence of healthcare risk factors and determination of whether the acquisition was community or hospital onset, as previously described (10). The Acute Physiology and Chronic Health Evaluation (APACHE) II score was calculated for each patient at onset (10). Thirty-day mortality was defined as mortality occurring in the 30 days following collection of the index culture. Microbiologic failure was defined as growth of MRSA from blood culture > 10 days from collection of
index culture while still on therapy. Clinical failure was defined as a composite of 30-day mortality and/or microbiologic failure. Using similar definitions, the clinical failure rate for MRSA BSI overall at our institution during the study period was 23% (3).

Each USA600 isolate underwent PFGE, SCCmec and agr typing, and testing for PVL as previously described (3, 8). In vitro susceptibility testing was performed according to standards set by the Clinical and Laboratory Standards Institute (8). Vancomycin MIC was determined by Etest (bio-Merieux, Durham, NC) and manual broth microdilution (BMD) (8). Vancomycin MBCs were determined using previously established methods (8, 14) and vancomycin tolerance was defined as an MBC/MIC ratio of at least 1:32 after 24 hrs incubation. Isolates were tested for heterogeneous vancomycin-intermediate S. aureus (hVISA) phenotype using the macrodilution Etest (MET) method (bio-Merieux, Durham, NC) as previously described (23). Isolates positive for hVISA by this method underwent population analysis as previously described (5). Each method was performed in duplicate to confirm findings.

Clinical characteristics, therapy, outcomes and vancomycin susceptibility results are reported in Table 1. Thirty-day mortality, microbiologic failure and clinical failure rates were 50%, 31% and 75% respectively. Patients were mostly female (69%), had a mean age of 64 (±19) years and APACHE-II score of 20 (±7) points at presentation. Comorbid diseases included; diabetes (63%), cardiovascular (69%), kidney (19%), hemodialysis (31%), liver (6%), neurologic (32%), chronic obstructive lung (25%), malignancy (6%), immunosuppression (25%) and HIV (6%). Other conditions present at baseline were acute renal failure (44%), previous hospital admission (56%), surgery within 30 days...
(19%), nursing home residence (38%) and intravenous drug abuse (0%). Antimicrobial exposure in the previous 90 days included; any (56%), vancomycin (25%), fluoroquinolone (25%), beta-lactam (19%), cephalosporin (13%), linezolid (13%), trimethoprim/sulfamethoxazole (13%), aminoglycoside (13%) and macrolide (13%). Epidemiologic classification of the infection was community (6%), healthcare-associated community-onset (75%) or hospital onset (19%).

None of the USA600 isolates were susceptible to clindamycin or erythromycin, 60% were susceptible to trimethoprim/sulfamethoxazole (TMP/SMX) and 75% susceptible to gentamicin. All isolates were susceptible in vitro to vancomycin by BMD, whereas one isolate was intermediate by Etest. A majority of isolates had an MBC ≥ 32 µg/ml (53%) and were tolerant to vancomycin (60%). Fifty percent of USA600 MRSA isolates tested positive for hVISA by the MET method. Four of these 8 isolates demonstrated the hVISA phenotype by population analysis. Molecular analysis revealed 15 strains were SCCmec type II and agr I, and one isolate was SCCmec type IVa and agr I. All isolates were PVL negative.

This initial report describes a series of bloodstream infections caused by USA600 MRSA. Although USA600 MRSA BSI was uncommon, we found a high rate of mortality and clinical failure, relative to previously reported outcomes in MRSA BSI (11, 12, 17). This sample was not adequate to evaluate the effect of different antimicrobial strategies or the contribution of vancomycin serum trough concentrations. However, most patients were treated with vancomycin, a drug to which half of the isolates were heteroresistant.
by MET, which could partially explain the poor outcomes. A previous case series reported hVISA mortality of approximately 40%, occurring mainly in two PFGE strain types, neither of which was USA600 (7). Another hospital in the Detroit area, reported a mortality rate of 33% in hVISA BSI, which was not significantly different than the non-hVISA BSIs (13). This observation was also shown in a small series of infective endocarditis, which reported a mortality rate of 42% in hVISA compared to 35% in non-hVISA (2). In our study, 63% of the hVISA patients died within 30 days compared to 38% of the non-hVISA patients. This suggests that other unique factors may be involved with USA600 BSIs. We found discordance in testing by MET and population analysis, which is consistent with a recent evaluation demonstrating only 64% of isolates positive by MET confirming by population analysis (15). The optimal method to test for hVISA phenotype is unclear, although population analysis is considered the ‘gold standard’.

The first reports of vancomycin-resistant S. aureus (VRSA) and 8 of 10 known cases were from patients in the Detroit area. Recently, hVISA in USA600 MRSA was reported in children from Detroit where 2 out of 3 USA600 MRSA infections were hVISA (1).

Understanding the emergence of potentially novel strains in Detroit may have important implications for other geographic areas. In this series, USA600 comprised less than 5% of all MRSA BSI. However, experience in Europe and Canada demonstrates this strain has profound ability for widespread dissemination (16, 20, 21). USA600 is clonally related to the Berlin strain of MRSA (ST45), which spread throughout Germany (21), the Netherlands (20) and Ontario (16) over the last decade. Despite this, colonization with USA600 MRSA remains low in the USA (19), and reported infections with USA600
remain relatively infrequent. In order to fully understand the impact of USA600 MRSA both within and outside the Detroit area, these findings need confirmation in a larger comparative evaluation in both BSI and possibly other types of infection.

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References


Figure 1. Pulsed-field Gel Electrophoresis Patterns for the USA600 MRSA Strains
### Table 1. Characteristics and Outcomes of USA600 MRSA Bloodstream Infections

| PI # | PI | Source of Bloodstream Infection | APACHEII | Treatment (Day)** | DOB (d) | V MIC ETEST (µg/ml) | V MIC BMD (µg/ml) | V MBC (µg/ml) | V Tolerant | MET hVISA | POP hVISA | Outcome/Comments |
|------|----|---------------------------------|----------|-------------------|---------|--------------------|------------------|--------------|------------|-----------|-----------|------------|-----------------|
| 1    | 56 | M Pneumonia                      | 16       | V (3-18)          | 1       | 2                  | 0.5              | 0.5          | No         | Yes       | Yes       | Died (Day 45) |
| 2    | 60 | F Endocarditis                    | 24       | V (1-11)          | 5       | 2                  | 1                | 2            | No         | Yes       | Yes       | Died (Day 11) |
| 3    | 86 | M Pneumonia                      | 22       | V+T (1-6), V (6-8) | 1       | 1.5               | 0.5              | ≥32          | Yes        | No        | NT        | Died (Day 6)  |
| 4    | 75 | F Infected Graft                  | 20       | V+G (1-6), V (6-42)| 4       | 1.5               | 0.5              | ≥32          | Yes        | Yes       | No        | Died (Day 100) Graft removed Day 6 |
| 5    | 41 | F Unknown                         | N/A      | Dead on Arrival to ER | N/A | 1.5               | 0.5              | 16           | Yes        | No        | Yes       | Died (Day 1)  |
| 6    | 69 | F Genitourinary                   | 32       | V (2-3)           | 1       | 2                  | 0.5              | ≥32          | Yes        | Yes       | No        | Withdrew Care Died (Day 8) |
| 7    | 63 | F Pneumonia                       | 38       | V+T (1), C (1-2)  | 1       | 1.5               | 0.5              | ≥32          | Yes        | No        | NT        | Died (Day 2)  |
| 8    | 77 | F Skin/Wound                      | 18       | V (1-6)           | 1       | 2                  | 0.5              | 0.5          | No         | No        | NT        | Died (Day 6)  |
| 9    | 54 | M Endocarditis                    | 23       | V (1-10)          | 13      | 2                  | 0.5              | ≥32          | Yes        | Yes       | No        | Microbiologic Failure Died (Day 13) |
| 10   | 44 | F Catheter                        | 20       | L+G (5-1) D (5-7) V (7-38) | 1       | 1.5               | 0.5              | 0.5          | No         | No        | NT        | Success       |
| 11   | 98 | F Genitourinary                   | 14       | L (1-3), V (3-38) | 25      | 1.5               | 0.5              | 0.5          | No         | No        | NT        | Microbiologic Failure Died (Day 62) |
| 12   | 55 | F Skin/Wound                      | 14       | V (2-15), TS (4-7) | 11      | 1.5               | 0.5              | ≥32          | Yes        | Yes       | Yes       | Microbiologic Failure |
| 13   | 65 | M Osteomyelitis                   | 19       | V (1-4), D (4-22), R (6-26), G (10-14), V (26-29), L (29-70) | 18      | 1.5               | 0.5              | ≥32          | Yes        | No        | NT        | Microbiologic Failure |
| 14   | 62 | M Infected Pacemaker              | 13       | D (1-8), R (1-4), TS (4-8) | 7       | 3                  | 2                | 2            | No         | Yes       | Yes       | Died (Day 9) Paeomaker removed Day 7 |
| 15   | 38 | F LVAD                            | 12       | V (1-10), D (10-13), R (8-13), G (8-13)** | 258     | 1.5               | 0.5              | 0.5          | No         | No        | NT        | Microbiologic Failure Patient Died (Day 258) |
| 16   | 34 | F Skin/Wound                      | 12       | V (1-14)          | 1       | 1.5               | 0.5              | ≥32          | Yes        | No        | NT        | Success       |

* Definitions: DOB=duration of bacteremia; BMD=broth microdilution; MET=microdilution Etest; POP=population analysis; NT=not tested; V=vancomycin; T=tobramycin; G=gentamicin; C=chloramphenicol; L=linezolid; D=daptomycin; TS=trimethoprim/sulfamethoxazole; R=rifampin; LVAD=left ventricular assist device

** Of 14 patients treated with vancomycin, 8 did not have vancomycin levels (5 were hemodialysis, 2 on treatment < 48h, 1 not available). The initial (≤ 48h) vancomycin trough of the six remaining patients were 10-15 µg/mL (n=4) and >15 µg/mL (n=2). The definitive (>48h) vancomycin trough was 10-15 µg/mL (n=1), >15 µg/mL (n=4) and not available (n=1).

***Antibiotics from first admission reported in table. Patient treated consecutively for 258 days with the following agents: Vancomycin, Daptomycin, Rifampin, TMP/SMX, Linezolid, Quinupristin/Dalfopristin.