

# Reduced vancomycin susceptibility among clinical *Staphylococcus aureus* isolates ('the MIC Creep'): implications for therapy

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## Abstract

Methicillin-resistant *S. aureus* (MRSA) has emerged as the most common hospital-acquired pathogen and is associated with increased morbidity and mortality compared with other strains. Vancomycin has been the cornerstone of treatment of patients with serious MRSA infections for some decades and while more than 99% of clinical *S. aureus* isolates remain susceptible to vancomycin, we are beginning to see strains of MRSA with reduced susceptibility. This review discusses this phenomenon, the predictors of infection with such forms of MRSA, and current and future management options.

## Introduction

Methicillin-resistant *S. aureus* (MRSA) has emerged as the most common hospital acquired pathogen, accounting for >60% of the clinical *S. aureus* isolates recovered in US intensive care units [1]. Infection with MRSA is associated with increased morbidity, requirement of longer duration of antibiotic therapy, higher healthcare costs, prolonged hospitalization and an increased risk of death [2,3]. This risk is more pronounced in patients who have been treated sub-optimally, either with an ineffective antibiotic and/or inadequate surgical intervention.

Vancomycin has been the cornerstone of treatment of patients with serious MRSA infections. Consequently, vancomycin use has been increasing since the mid-1980's, resulting in the emergence of MRSA with reduced susceptibility to vancomycin. In patients with *S. aureus* bacteremia, higher vancomycin minimum inhibitory concentrations (MICs) have been associated with prolonged bacteremia or increased mortality [4]. While vancomycin non-susceptible strains (see Table 1), in the form of intermediate-resistant *S. aureus*, (VISA, vancomycin MIC 4-8 mg/L) remain rare and vanA-mediated vancomycin-resistant *S. aureus* (VRSA, vancomycin MIC > 16 mg/L) are limited to a handful of reported cases, the

rising MICs of vancomycin among vancomycin susceptible *S. aureus*, referred to as the 'vancomycin MIC creep', has caused a re-evaluation of vancomycin susceptibility criteria in cases of complicated infections like bacteremia and/or pneumonia [4,5].

This report will discuss the phenomenon of vancomycin MIC creep in *S. aureus*, its potential association with decreased vancomycin efficacy, the predictors of infections with MRSA with high vancomycin MIC, and briefly review the current and future management options.

## Vancomycin – the early years

Vancomycin is a complex tricyclic glycopeptide that was first isolated from *Amycolatopsis orientalis* found in a soil sample from Borneo in the mid 1950s that has activity against most clinically relevant Gram-positive pathogens [6]. In 1958, vancomycin was introduced for clinical use against penicillin-resistant *S. aureus*, but for the first 25 years after introduction, its use remained limited to patients with severe beta-lactam allergy and with MRSA infection, which were infrequent prior to 1980. However, the 1980s were marked by a surge in the use of cephalosporin and quinolone antibiotics in hospitals, which paralleled the emergence of methicillin resistance

**Table 1: Clinical Laboratory and Standards Institute (CLSI) susceptibility breakpoints for vancomycin**

Pathogen	Abbreviation	Susceptibility profile	MIC (µg/mL)
Vancomycin-susceptible <i>S. aureus</i>	VSSA	susceptible	<2
Heteroresistant <i>S. aureus</i> with intermediate susceptibility to vancomycin or glycopeptide	hVISA or hGISA	heteroresistant	1-2 <sup>a</sup>
<i>S. aureus</i> with intermediate susceptibility to vancomycin or glycopeptide	VISA	intermediate	4-8
Vancomycin-resistant <i>S. aureus</i>	VRSA	resistant	>16 <sup>b</sup>

<sup>a</sup>consists of subpopulations ( $\leq 1$  in 1,000,000) that may grow in media containing  $>2$  µg/mL of vancomycin.

<sup>b</sup>requires backup primary testing with 6µg/mL of vancomycin on an overnight plate.

initially in coagulase-negative staphylococci and subsequently *S. aureus*. During this early period, vancomycin dosing was centered around avoidance of nephrotoxicity and dosing convenience rather than efficacy or prevention of developing resistance, with target serum trough concentrations of 5-10 mg/L. In contrast, recent consensus guidelines have recommended troughs of 15-20 mg/L in the treatment of serious MRSA infections, not only for increasing pharmacodynamic exposure to improve *in vivo* efficacy but also to prevent selection of *S. aureus* strains with higher MICs that have been demonstrated *in vitro* in vancomycin concentrations  $< 10$  mg/L [7]. It is important to realize that a robust risk-benefit analysis for pushing serum concentrations of vancomycin to these heights is so far lacking.

### Vancomycin susceptibility testing

For antimicrobials, various laboratory methods of susceptibility testing against strains of bacteria are used to predict clinical response. The simplest of these assays is the "MIC", which refers to the lowest concentration of antibiotic that inhibits visible growth of a standard inoculum size of bacteria (generally  $10^4$  cfu for staphylococci) after overnight incubation in Mueller-Hinton broth. A result of "susceptible" by a clinical microbiology laboratory is considered to be predictive of a satisfactory clinical response whereas "nonsusceptible" or "resistant" predicts failure. While beyond the scope of this article, readers must appreciate that establishing these MIC so-called "breakpoints" for strains that are susceptible, intermediate, and resistant to antibiotics is controversial and wrought with laboratory oversimplifications of a very complex host-pathogen-antibiotic relationship. In short, microbiological breakpoints (maximum MIC thresholds for predicting successful treatment) are established by combining data of MIC distributions of groups of clinical strains and available pharmacodynamic and pharmacokinetic information, but not on any data that correlates clinical response to MIC. Furthermore, the MIC measures inhibition of growth, not killing potency of an antibiotic. Such methods are much more time consuming and are rarely used and/or available to

clinicians in a timely manner. Finally, *in vitro* assays of susceptibility like MIC measurements completely ignore any potential interaction of the antibiotic with innate host defense mechanisms that may occur *in vivo*.

Various tests and methodologies with variable sensitivity and specificity are available to measure vancomycin MICs. Broth microdilution is considered to be the gold standard for measuring vancomycin MIC. However, broth microdilution can be a cumbersome test and is not used routinely in clinical laboratories. Since it is reliant on a two-fold dilution, it offers limited quantitative information. Also, agar disc diffusion is not a suitable test for large molecules like vancomycin and moreover it does not measure the MIC directly and cannot detect VISA isolates. Currently, most clinical laboratories use Etest and automated susceptibility tests for measuring the vancomycin MIC. However, automated testing has been associated with underestimation of vancomycin MIC when compared to Etest and broth microdilution [8]. In 2006, CLSI revised the susceptibility breakpoints for vancomycin in response to evidence that vancomycin was poorly effective against MRSA isolates with MIC  $> 4$  mg/L [4].

### Vancomycin resistance and heteroresistance VRSA

VRSA (vancomycin resistant *S. aureus*) isolates remain rare and sporadic. Since the first report in 2002, there have been 13 confirmed cases of VRSA [9] from US, and one each from India and Iran). A few existing factors that seem to predispose patients to VRSA infection include prior MRSA and vancomycin-resistant enterococcal infections or colonization, underlying chronic medical conditions, and previous extensive treatment with vancomycin [10].

### VISA and hVISA

*S. aureus* isolates with broth microdilution MIC 4-8 µg/mL are termed as VISA. This definition has been excluded from the new EUCAST breakpoints which define an isolate with broth microdilution MIC  $\geq 4$  µg/mL as resistant rather than

having intermediate susceptibility. More common than VISA and VRSA is the occurrence of *S. aureus* isolates exhibiting heteroresistance to vancomycin. hVISA (heteroresistant VISA) is defined as a vancomycin-susceptible *S. aureus* (VSSA) strain that upon subculture produces sub-colonies with MIC in the VISA/VRSA range at the frequency of  $\geq 1 \times 10^6$  according to population analysis profile (PAP) (Figure 1). Quantitative definitions used for hVISA are an AUC (area under the curve) ratio of  $\geq 0.9$  of the AUC of type strain Mu3 (PAP/AUC) or modified high-inoculum Etest read at 48 h [11,9].

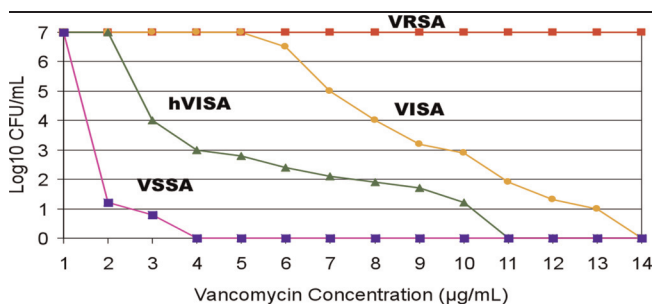
The hVISA phenotype is considered to be a precursor to VISA, and falls on the same continuum on the heteroresistance spectrum. The first case of hVISA was reported in Japan in 1996. Current reported prevalence of hVISA is variable (0-74%) because of difficulty of detection, cumbersome and non-standardized detection methods, variable selective vancomycin pressure, and instability of the phenotype once some isolates are frozen and stored [9]. The main risk factors for infections caused by VISA and hVISA strains are prior MRSA infection with high bacterial load (endocarditis, deep abscess, prosthetic joint infections) and prior vancomycin exposure (particularly a low serum concentration). Nosocomial spread and rare outbreaks caused by hVISA or VISA strains have also been reported.

The above discussion points out an obvious inherent flaw in establishing a microbiological breakpoint for vancomycin that correlates cleanly with clinical response. Vancomycin heteroresistance in hVISA is characterized by the existence of subpopulations of organisms among vancomycin-susceptible strains that are able to grow in

concentrations of vancomycin in the range of intermediate-resistant organisms. This property is present in all staphylococci but in varying degrees. If the proportion of organisms in the bacterial population is sufficiently high to be detected with standard low inocula of susceptibility testing, they may be detected and appropriately considered nonsusceptible. However, if the proportion of organisms in a bacterial population that can grow in vancomycin >2 mg/L is below the frequency of the testing inoculum density, then the strain will be characterized as susceptible by standard susceptibility testing. Infections with high bacterial load (e.g. pneumonia, endocarditis, abscess) will not only have a high risk for vancomycin treatment failure but also risk the rapid development of vancomycin intermediate resistance (VISA), especially if vancomycin is under-dosed and the organisms are exposed to vancomycin concentrations <10g/L [12]. This latter process is simply "enriching" the already present bacterial population for vancomycin-resistant subspecies to the point where subsequent susceptibility testing, after selection, will appropriately characterize these strains as VISA.

It is important to also consider that in situations where the infection is characterized by low bacterial inocula (e.g. UTI, simple cellulitis without abscess), vancomycin heteroresistance, and, therefore, the MIC creep, is likely to be less clinically relevant, if at all. The higher the vancomycin MIC, the more likely that a strain will exhibit subpopulations that can be grown in ranges of vancomycin concentrations comparable to vancomycin non-susceptible strains (Figure 2). Therefore, a high-inoculum infection like endocarditis caused by a vancomycin-susceptible *S. aureus* with an MIC of 2 mg/L is much more likely to fail vancomycin therapy than a low-inoculum infection such as a surgical wound infection without an abscess where the vancomycin MIC is lower. Both strains are vancomycin "susceptible", but clinical susceptibility to the drug is different in each of these cases.

**Figure 1. Population analysis among hVISA and VISA of heterogeneous resistance to vancomycin**

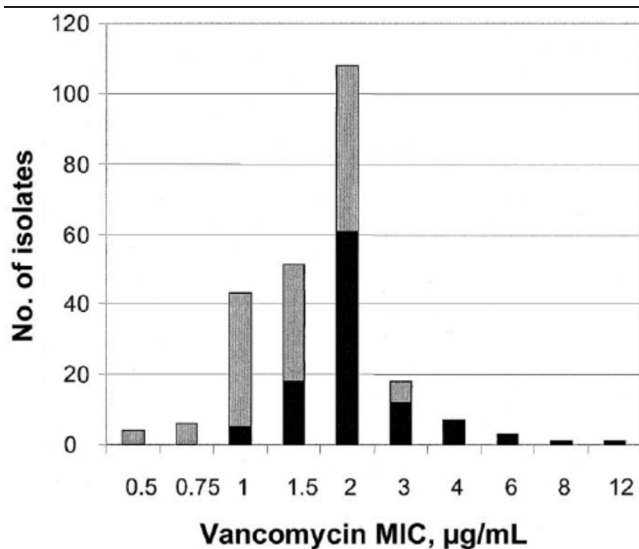


Population analysis demonstrating varying degrees of heterogeneous resistance to vancomycin among hVISA and VISA, relative to the more homogenous patterns seen in vancomycin-susceptible *S. aureus* (VSSA) and vanA-mediated vancomycin resistance in VRSA. (*Antimicrob Agents Chemother*, 2003, 47:3040-5, 10.1128/AAC.47.10.3040-3045.2003, reproduced/amended with permission from American Society for Microbiology [11].)

**Vancomycin Pharmacodynamics**

Based on neutropenic mouse models, *in vitro* studies and limited data from human studies, the AUC/MIC ratio has been used as a preferred parameter for measuring the effectiveness of vancomycin in treating *S. aureus* infections [13,14]. A specific AUC/MIC threshold of 400 has been advocated as a target to achieve clinical effectiveness with vancomycin, based on the initial clinical data from pneumonia [15] and more recent data from bacteremia [16]. These considerations superimposed on the vancomycin MIC creep have provided circumstantial evidence, without clinical trial validation, to support higher vancomycin trough concentrations in the

**Figure 2. Heteroresistance to vancomycin therapy among *S. aureus* isolates**



Fraction of hVISA (black) present among total *S. aureus* stratified by vancomycin MIC. (reproduced from Fred C. Tenover, Robert C. Moellering, Jr., The Rationale for Revising the Clinical and Laboratory Standards Institute Vancomycin Minimal Inhibitory Concentration Interpretive Criteria for *Staphylococcus aureus*, *Clin Infect Dis*, 2007, 44:1208-15 by permission of Oxford University Press [4].)

15-20 mg/L range. Using Monte Carlo simulations, it has been suggested that the probability of attaining this ratio is approximately 100% only in isolates with MIC  $\leq$  0.5 mg/L and the probability falls to 0% in isolates with MIC of 2 mg/L [17]. Based on simulated models, it has further been reported that a daily dose of 3-4 gm of vancomycin will be required to provide 90% probability of attaining the target AUC/MIC of 400 for an isolate with MIC 1 mg/L [18].

While vancomycin has traditionally been considered a bactericidal agent, its potency is reduced to bacteriostatic levels in the setting of high-inoculum infections, stationary growth phase, or in an anaerobic environment. Decreased bactericidal activity (vancomycin tolerance) has been associated with higher vancomycin MIC. However, not all MRSA strains with a vancomycin MIC of 2 mg/L are vancomycin tolerant. Vancomycin tolerance has been seen in 15% of wild-type MRSA strains compared with 74% of hVISA strains and 100% of VISA and VRSA strains [19]. This decreased bactericidal activity of vancomycin has been shown to be associated with poor clinical outcomes in bacteremia and can be independent of the MIC values [20]. Receipt of any vancomycin within 30 days predicts bacteriostatic activity among MRSA bacteremia isolates, whereas

vancomycin-naïve patients with MRSA bacteremia can be expected to have isolates against which vancomycin is bactericidal [21].

### Phenotypic changes associated with decreased vancomycin susceptibility

VISA has altered cellular physiology as a result of cumulative effects of mutations and/or modulation of regulatory systems. This altered physiology appears to change cell-wall metabolism in such a way as to result in increased numbers of D-Ala-D-Ala residues, which serve as dead-end binding sites for vancomycin. In addition, evaluation of *S. aureus* with reduced vancomycin susceptibility and isogenic vancomycin-susceptible progenitors showed cell walls with reduced peptidoglycan cross-linking, reduced cell-wall turnover, and reduced autolysis, causing considerable morphological cell-wall thickening. This altered cell wall results in a reduced diffusion coefficient of vancomycin, sequestration of vancomycin within the cell wall by these false targets, and prevention of vancomycin reaching its site of action [22-24].

### Vancomycin 'MIC creep' and clinical significance

Within the populations of *S. aureus* that are considered to be susceptible, a changing pattern of vancomycin MICs has been observed in some centers, demonstrating an overall population drift in the clinical isolates of *S. aureus* towards reduced vancomycin susceptibility. This phenomenon of "vancomycin MIC creep" varies considerably around the world and is summarized in Table 2. However, it is important to point out that this is not universal and some centers have noted no changes or even reductions in vancomycin MIC creep (GS, unpublished observations). Thus, when large numbers of *S. aureus* samples are pooled together and analyzed from multiple centers, as in the SENTRY database study, center to center heterogeneity leads to a net neutralization effect, with no overall changes in vancomycin susceptibility observed [19]. These variations are accounted for by differences in the epidemiological and clinical factors between study sites, including selective pressure generated by high-dose versus traditional use and dosing of vancomycin, severity of illness in the patient populations with different co-morbid conditions, difference in medical and surgical therapy of invasive *S. aureus* infections, and variation in susceptibility testing methods [25]. This variation is further confounded by the emergence of community-acquired MRSA and its prevalence in hospital settings. Community-acquired MRSA historically has a lower vancomycin MIC than MRSA from hospitals, presumably because of reduced vancomycin exposure, although virulence selection pressure may also play a role (discussed later). Furthermore, one needs to take reporting bias into account, whereby a

**Table 2: Summary of studies evaluating evidence of increasing Vancomycin MIC and MIC Creep in *S. aureus* (all MIC in µg/mL)**

Study	Study type	MIC method	Evidence of MIC creep	Comments
Wang 2006 [26]	2000-2004 Single center (US) 6003 SA, All clinical specimens	BMD	Yes	Shift of MIC from <0.5 to 1.0 over 5 year period. % isolates with MIC>1: 2000 (19.9%) vs. 2004 (70.4%). MIC creep noted in both MSSA and MRSA isolates.
Golan 2006 [8]	2002-2005 Single center (US)	BMD	Yes	Geometric mean vancomycin MIC increased from 0.9 to 1.4 over the study period. % isolates with MIC >0.5 increased for 67% to 96%, % isolates with MIC= 2 increased form 13% to 51% Isolates with MIC= 2 were frequently missed by automated systems.
Robert 2006 [43]	1983-2002 Single center (France) 1445 MRSA, All clinical specimens	Etest	Yes	Vancomycin MIC geometric mean increased from 1.56 in 1983 to 2.41 in 2002
Jones 2006 [19]	1998-2003 SENTRY database (>50 worldwide centers) 35,485 SA, 5902 CoNS All clinical specimen	BMD	No	No detected MIC creep in all gram positive organisms. Vancomycin tolerance was noted in wt MRSA (15.2%), VISA (73.9%), VISA (100%), VRSA (100%) with significant associated decrease in bactericidal activity.
Steinkraus 2007 [44]	2001-2005 Single center (US) 662 MRSA Blood Culture	Etest	Yes	1.5-fold increase in the geometric mean vancomycin MIC. % isolates with MIC=1 2001(16%) vs. 2005(69%). % isolates with MIC>1 2001 (0%) vs. 2005(7%).
Holmes 2008 [45]	1999-2006 Single center ( US) 240 MRSA Blood Culture	BMD	No	No change MIC and bactericidal activity of vancomycin in the pre-therapy isolates over the study period.
Alos 2008 [46]	2002-2006 Single center (Spain) 3141 SA, (566 MRSA) All clinical specimen	BMD	No	In an area of low vancomycin consumption, no statistically significant difference in 2 MIC groups ( $\leq 1$ vs. $\geq 2$ ).
Sader 2009 [47]	2002-2006 9 medical centers (US) 1800 MRSA Blood cultures	BMD	Possible/No	Geometric mean data showed a possible very low level MIC creep in 3/9 centers not evident on modal MICs. No overall increase in the resistance noted.
Musta 2009 [29]	1996-2006 period Single center (US) 489 blood cultures	Etest	No	% isolates with vancomycin MIC $\leq 1$ , 1.5, 2, 3 and % isolates with hVISA was unchanged over the 11 year period
Karas 2010 [48]	1998-2007 2 medical centers (UK) 259 MRSA Blood cultures	Agar Dilution	Possible	Increase in the % isolates with MIC>0.75 1998-2001 (4.4%) vs. 2002-04(4.9%) vs. 2005-07 (10.8%)
Ho 2010 [49]	1997-2008 5 hospitals (Hong Kong) 247 MRSA Blood cultures	Etest	Yes	Increase in the % isolates with MIC=1 1997-99(10.8%) vs. 2004(21.6%) vs. 2006-08 (38.3%)

Abbreviations: BMD, broth microdilution; MIC, minimum inhibitory concentration; MSSA, Methicillin-susceptible *S. aureus*; MRSA, Methicillin-resistant *S. aureus*; VISA, vancomycin-intermediate *S. aureus*; VRSA vancomycin-resistant *S. aureus*.

disproportionately higher numbers of centers with observed increases in vancomycin MIC may report their positive results in the literature, as opposed to the negative results from centers without this phenomenon. In short, the relevance of the vancomycin creep for individual clinicians, pharmacists, and microbiologists can only be assessed by the evaluation of their local susceptibility profiles, not published observations of others.

Infections caused by *S. aureus* isolates with higher vancomycin MIC, even those in the susceptible range, have shown to be associated with various poor clinical outcomes, including delayed early response, increased mortality, increased rate of relapse, prolonged hospital stay or overall increased cost of hospitalization (see Table 3).

Vancomycin MIC creep has been observed in both methicillin-susceptible and -resistant *S. aureus* isolates

[26,27]. Interestingly, elevated vancomycin *S. aureus* MIC has been associated with increased mortality in patients with methicillin-susceptible *S. aureus* infections when they are treated with either vancomycin or flucloxacillin [27].

### Predictors for vancomycin resistance

Recent guidelines have suggested considering alternative antibiotics in complicated MRSA infection when vancomycin MIC is  $\geq 2$  µg/mL [28]. Various studies have attempted to identify patients at risk of infections with *S. aureus* with higher vancomycin MIC. This is important, as many hospitals estimate vancomycin MIC using automated methods and do not routinely provide a measured MIC value for the medical record. Also up to 90% of MRSA isolates with a MIC of 2µg/mL can be missed by some automated systems [11]. Infections caused by MRSA with higher vancomycin MIC are seen in patients with recent exposure to vancomycin within one

**Table 3: Summary of studies evaluating clinical outcomes in infections with high Vancomycin MIC (all MICs expressed in µg/mL)**

Study	Study type	Clinical Isolates	MIC method	Clinical outcomes
Moise-B 2004 [50]	1998-2001 Multicenter (US)	122 MRSA infections (25 blood cultureD)	BMD	Increased failure rate was associated with higher MIC: 22 % (MIC 0.5) vs. 27% (MIC 1.0) vs. 51 % (MIC 2) and in patients with bacteremia of unknown origin, endocarditis, and respiratory infections.
Sakoulas 2004 [20]	1998-2001 Multicenter (US)	30 MRSA blood cultures	BMD	Treatment success with vancomycin was associated with lower MIC: 55.6 % (MIC≤0.5 vs. 9.5 % (MIC 1-2) and increased vancomycin killing (OR 10.73). No significant relation between vancomycin MIC and vancomycin bactericidal activity.
Rhee 2005 [51]	1994-1999 Single center (US) 2002-2004	171 SA blood culture/ osteomyelitis 52 clinical isolates	N/A	Decrease vancomycin susceptibility based on MBC data. % isolates with MBC≥1.6mcg/mL increased from 8% in 1994 to 30% in 1999. From 2002-2004, increased proportion of isolates with MBC≥2.
Maclayton 2006 [52]	2001-2003 Single center (US)	50 MRSA blood culture in HD patients	Vitek	Increased mortality in patients with higher vanco- mycin MIC: 35 % (MIC 1-2) vs. 24 % (≤ 0.5) vs 15% (controls). Increased cost of hospitalization was seen in group with MRSA infections with higher vancomycin MIC. Risk factors of infection with higher MIC were surgery within last 6 months and ICU admission.
Hidayat 2006 [53]	2004-2005 Single center (US)	95 MRSA infections	Etest	Overall response rate at end of therapy was lower in patients with infection with MRSA with higher MIC: 62% (MIC 2) vs. 85% (MIC≤1). Poor outcomes were associated with MIC 2 (OR 6.02) and severity of underlying illness (OR 3.14). Increased nephrotoxicity was seen with higher serum trough levels (15-20), duration of vancomycin treatment and use of concomitant nephrotoxic agents.
Neoh 2007 [54]	1998-2005 Single center (Japan)	22 MRSA blood cultures	Agar dilution	Significant difference in response was seen in patients with lower vancomycin susceptibility as expressed by area under curve (AUC) of population analysis.
Soriano 2008 [40]	1991-2005 Single center (Spain)	414 MRSA blood cultures	Etest	Higher mortality associated with increased vanco- mycin MIC: OR 2.86 (MIC 1.5) and OR 6.37 (MIC 2) vs. OR 3.62 (inappropriate therapy). Vancomycin MIC=2 was associated with decreased risk of shock (OR 0.33)
Lodise 2008 [30]	2005-2007 Single center (US)	92 MRSA blood cultures	Etest	2.4 fold increase in the risk of failure and longer hospital stay associated with vancomycin MIC≥1.5. Ability to achieve primary trough levels of 15mg/L was not associated with increased probability of success irrespective of MIC.
Musta 2008 [29]	1996-2006 Single center (US)	489 MRSA blood culture Mortality results on 285 isolates	Etest	Increased mortality with high vancomycin MIC: 19.4% (MIC≤1 vs. 27% (MIC 1.5 vs. 47.6% (MIC ≥ 2). hVISA seen in 14% of isolates and was not associated with increased mortality.
Yoon 2010 [55]	2006-2009 Single center (Korea)	96 MRSA blood cultures	Vitek 2	Persistent bacteremia associated with vancomycin MIC-2(OR 6.23), retention of medical devices (OR 10.35) and MRSA infection at more than two sites (OR 10.24). Increased MRSA bacteremia related mortality asso- ciated with higher vancomycin MIC: 50 % (MIC 2) vs. 19% (MIC≤1).
Wang 2010 [56]	2006 Single center, (Taiwan)	126 MRSA blood culture	BMD	Increased mortality at day 14 and 30 was associated with vancomycin MIC=2 (OR 3.76). Vancomycin MIC=2 was seen in patients with hospitalization >2 months ago (OR 4.56) and ICU admission prior to developing bacteremia (OR 4.83).
Haque 2010 [31]	2006-2007 4 US centers	158 MRSA pneumonia	Etest	Risk of death was increased 2.97 fold for increase of 1 µg/mL of vancomycin MIC. Heteroresistance was seen in 21.5% of isolates and was not associated with increased mortality.
Choi 2011 [57]	2008-2009 Single center (Korea)	70 MRSA pneumonia	Etest	Higher vancomycin MIC was associated with decreased early response 35.3 % (MIC≥1.5 vs. 63.9% (MIC≤1) and increased risk of relapse at day 28 29.6 % (MIC≥1.5 vs. 6.9% (MIC≤1).

Kullar 2011 [16]	2005-2010 Single center (US)	320 MRSA blood culture	Etest	Vancomycin failure was associated with Etest MIC > 1 (AOR 1.5), endocarditis (AOR 4.55), nosocomial infection (AOR 2.19) and initial vancomycin trough < 15 mg/L (AOR 2). Failure rates increased with vancomycin AUC/MIC: 61% (AUC/MIC < 421) vs. 48.6% (AUC/MIC > 421). Increased mortality associated with higher Etest MIC 12.2% (MIC ≤ 1.5) vs. 26.8% (MIC > 1.5). Increased mortality with high vancomycin regardless of methicillin susceptibility and treatment with either vancomycin (OR 2.25) or flucloxacillin (OR 2.25). High vancomycin MIC was associated with methicillin resistance, hospital onset of infection, vancomycin treatment, device associated infection and death and increased incidence of sepsis.
Holmes 2011 [27]	2007-2008 27 sites Australia/NZ	532 SA blood culture 266 patients treated with vancomycin	Etest /BMD	

month of the current infection, prior recent hospitalization, surgery within last 6 months and those with blood stream infections prior to admission in intensive care units [29-32]. One recent study attempted to produce a score for high vancomycin MIC in MRSA blood stream infections using factors like age, prior vancomycin exposure, history of MRSA bacteremia, chronic liver disease, and presence of non-tunneled central venous catheter as predictors. Using these factors and the local rate of high vancomycin MIC, different cut-off points can be established and effectively used to rule in or rule out an infection with an MRSA isolate with high vancomycin MIC and therefore help in selecting the early and empiric effective anti-MRSA therapy [33].

### Vancomycin “cross-resistance creeps”

Due to the complex nature of the mechanism of vancomycin heteroresistance, which represents a summation of a combination of genetic and gene expression events, it would be expected that the vancomycin MIC creep would influence not only susceptibility to vancomycin but other antibiotics as well. This hypothesis has been validated extensively in the literature with daptomycin, a cyclic lipopeptide of a different antibiotic class and mechanism of action. *S. aureus* strains with higher vancomycin MICs also tend to have higher daptomycin MICs [34]. As one “climbs the ladder” of vancomycin resistance, increasing daptomycin nonsusceptibility is observed, with about 50% of VISA being daptomycin nonsusceptible [35]. This selection of daptomycin nonsusceptibility by vancomycin occurs without exposure to daptomycin [36]. While the molecular mechanism(s) remain unknown, this cross-heteroresistance between vancomycin and daptomycin has important clinical implications. First, it further underscores the danger of vancomycin underdosing, as selection of hVISA and VISA in an infection like endocarditis, which contains a large number of organisms. Secondly, given the less likely scenario of daptomycin selecting for vancomycin heteroresistance, debates have surfaced

towards using daptomycin rather than vancomycin as first line therapy, particularly in healthcare settings where vancomycin MICs of 2 mg/L are expected. While microbiologically sound, daptomycin is much more costly, and consequently physicians may be reluctant to use it during the first 24-72 hour window where blood cultures are pending. Third, use of daptomycin in salvage of vancomycin failure in MRSA bacteremia has been suggested at higher than approved doses of 8 mg/kg/day rather than the approved 6 mg/kg/day dose [28]. While unproven in benefit, higher doses are recommended when prior vancomycin has already reduced the potency of daptomycin, both in terms of increasing MIC and tolerance. Nevertheless, case reports demonstrate that the prior selection of daptomycin heteroresistance by vancomycin is not universally overcome with high-dose daptomycin monotherapy, and combination therapy with aminoglycosides is recommended. We have reported great success using daptomycin in combination with antistaphylococcal beta-lactams in the most recalcitrant cases of MRSA bacteremia [37].

### Vancomycin heteroresistance and implications for virulence

Knowledge of the innate host response has revealed that it consists, in part, of antimicrobial peptides produced by various cell types (e.g. cathelicidins, defensins, platelet microbicidal proteins) that serve to kill invasive pathogens. These antimicrobial peptides can be viewed in simple terms as endogenous antibiotic molecules produced by mammals. Thus, it would be conceivable that cross-resistance may occur between prokaryotic antibiotics and the antimicrobial peptides of mammals. While this area remains largely unexplored, evidence is emerging which demonstrates cross-resistance between platelet microbicidal proteins (cationic peptides produced by platelets) and vancomycin and daptomycin. Platelet microbicidal protein is believed to be an important innate host response mechanism that clears bacteria from the bloodstream when transient bacteremia occurs

under physiologic circumstances. Thus, resistance to platelet microbicidal proteins has been shown to be an important virulence factor in the ability to establish an endovascular infection, like bacterial endocarditis.

In *S. aureus*, selection pressure by vancomycin *in vivo* and *in vitro* co-selects for platelet microbicidal protein resistance [38]. Thus, a vancomycin MIC creep seen in MRSA from patients previously treated with vancomycin carries with it consequences that extend beyond vancomycin susceptibility and pharmacodynamics. MRSA with higher vancomycin MIC may be less susceptible not only to daptomycin but also platelet microbicidal proteins and potentially other antimicrobial peptides of the innate host response. The downstream consequences of these phenotypes, including the risk of endocarditis in patients previously treated with vancomycin, requires further study.

It is an oversimplification, however, to say that *S. aureus* strains with higher vancomycin MICs are more virulent, as the virulence machinery of this pathogen is extremely elegant and complex. In fact, there is evidence toward the contrary when one considers traditional measures of virulence, such as severity of illness as well as laboratory *in vivo* models [39]. In one study, while higher vancomycin MICs were related to higher mortality in MRSA bacteremia, vancomycin MIC was inversely related with symptoms of septic shock [40]. Other studies have shown that while hVISA is associated with prolonged bacteremia, it is inversely associated with mortality [41,42].

## Conclusions

Using vancomycin as the cornerstone of MRSA therapy for the past five decades, particularly in the last 20 years, has exerted considerable selection pressure on *S. aureus* strains in the healthcare setting. While > 99% of clinical *S. aureus* isolates remain susceptible to vancomycin, the vancomycin MIC creep seen in many institutions demonstrates that this organism has not remained inert to this pressure. While appearing subtle by the relatively crude assays used in clinical microbiology laboratories, evidence is emerging suggesting that the vancomycin MIC creep is just the "tip of the iceberg" with regards to clinical consequences. Further complicating the matter is the variability in vancomycin MIC obtained between agar-based Etest and broth microdilutions, both automated and manual.

Physicians and pharmacists are meeting these challenges in a variety of ways, including: i) the adoption of rapid molecular tests to quickly differentiate MRSA from beta-lactam susceptible strains and, therefore, convert patients with the latter more rapidly to superior beta-lactam

therapy; ii) optimization of vancomycin doses; iii) switching early to alternative agents when vancomycin MIC is 2 mg/L; iv) using combination antibiotic therapy; and v) abandoning vancomycin altogether in serious MRSA infections.

While all of these have theoretical benefits and, in some single center experiences, have actually shown benefit, the case for global generalizability of these countermeasures remains unproven. Vancomycin has yet to be demonstrated to be inferior to any of the novel agents against MRSA bacteremia in prospective multi-center studies. This may reflect the fact that many clinical trials exclude the extremely difficult circumstances where enhanced activity in laboratory settings of novel agents may translate into a clinical benefit. Trials examining subgroups of patients representative of these most difficult cases would be extremely difficult to perform. Many questions remain unanswered and the bridge between laboratory science and clinical medicine is still in the early stages of development. What is clear is that *S. aureus* infections will remain a challenge for years to come for physicians, microbiologists, pharmacists, scientists, and, most importantly, patients.

## Abbreviations

MRSA, methicillin-resistant *S. aureus*; VRSA, vancomycin-resistant *S. aureus*; MIC, minimum inhibitory concentration; VISA, vancomycin-intermediate *S. aureus*; VSSA, vancomycin-susceptible *S. aureus*; hVISA, heteroresistant vancomycin-intermediate *S. aureus*; hGISA, heteroresistant glycopeptide-intermediate *S. aureus*; PAP, population analysis profile; AUC, area under the curve.

## Competing interests

**AD:** member of a speakers bureau and receiver of speaking honoraria from Cubist and Pfizer Pharmaceuticals.

**GS:** member of a speakers bureau and receiver of speaking honoraria from Cubist, Forest, Astellas, and Pfizer Pharmaceuticals; grant support from Cubist Pharmaceuticals; consulting fees from Cubist Pharmaceuticals.

## References

1. Wisplinghoff H, Bischoff T, Tallent SM, Seifert H, Wenzel RP, Edmond MB: **Nosocomial bloodstream infections in US hospitals: analysis of 24,179 cases from a prospective nationwide surveillance study.** *Clin Infect Dis* 2004, **39**:309-17.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

2. Soriano A, Martinez JA, Mensa J, Marco F, Almela M, Moreno-Martinez A, Sanchez F, Munoz I, Jimenez de Anta MT, Soriano E: **Pathogenic significance of methicillin resistance for patients**

with *Staphylococcus aureus* bacteremia. *Clin Infect Dis* 2000, **30**:368-73.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

3. Cosgrove SE, Sakoulas G, Perencevich EN, Schwaber MJ, Karchmer AW, Carmeli Y: **Comparison of mortality associated with methicillin-resistant and methicillin-susceptible *Staphylococcus aureus* bacteremia: a meta-analysis.** *Clin Infect Dis* 2003, **36**:53-9.

4. Tenover FC, Moellering RC, Jr: **The rationale for revising the Clinical and Laboratory Standards Institute vancomycin minimal inhibitory concentration interpretive criteria for *Staphylococcus aureus*.** *Clin Infect Dis* 2007, **44**:1208-15.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

5. Sakoulas G, Moellering RC, Jr: **Increasing antibiotic resistance among methicillin-resistant *Staphylococcus aureus* strains.** *Clin Infect Dis* 2008, **46**:S360-7.

6. Levine DP: **Vancomycin: a history.** *Clin Infect Dis* 2006, **42**:S5-12.
7. Rybak MJ, Lomaestro BM, Rotschafer JC, Moellering RC, Jr, Craig WA, Billeter M, Dalovisio JR, Levine DP: **Therapeutic monitoring of vancomycin in adults summary of consensus recommendations from the American Society of Health-System Pharmacists, the Infectious Diseases Society of America, and the Society of Infectious Diseases Pharmacists.** *Pharmacotherapy* 2009, **29**:1275-9.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

8. Golan Y, Baez-Giangreco C, O'Sullivan C, Snyderman D: **Trends in vancomycin susceptibility among consecutive MRSA isolates.** In: Abstracts of the Forty-fourth Annual Meeting of the Infectious Diseases Society of America, Toronto, Ontario, Canada. Alexandria, VA, USA: Infectious Diseases Society of America; 2006, p. 238 Abstract LB-11.
9. Howden BP, Davies JK, Johnson PD, Stinear TP, Grayson ML: **Reduced vancomycin susceptibility in *Staphylococcus aureus*, including vancomycin-intermediate and heterogeneous vancomycin-intermediate strains: resistance mechanisms, laboratory detection, and clinical implications.** *Clin Microbiol Rev* 2010, **23**:99-139.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

10. CDC Health care-associated infections: [[http://www.cdc.gov/HAI/settings/lab/vrsa\\_lab\\_search\\_containment.html](http://www.cdc.gov/HAI/settings/lab/vrsa_lab_search_containment.html)]
11. Liu C, Chambers HF: ***Staphylococcus aureus* with heterogeneous resistance to vancomycin: epidemiology, clinical significance, and critical assessment of diagnostic methods.** *Antimicrob Agents Chemother* 2003, **47**:3040-5.
12. Sakoulas G, Eliopoulos GM, Moellering RC, Jr, Novick RP, Venkataraman L, Wennersten C, DeGirolami PC, Schwaber MJ, Gold HS: ***Staphylococcus aureus* accessory gene regulator (agr) group II: is there a relationship to the development of intermediate-level glycopeptide resistance?** *J Infect Dis* 2003, **187**:929-38.
13. Craig WA: **Basic pharmacodynamics of antibacterials with clinical applications to the use of beta-lactams, glycopeptides, and linezolid.** *Infect Dis Clin North Am* 2003, **17**:479-501.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

14. Rybak MJ: **The pharmacokinetic and pharmacodynamic properties of vancomycin.** *Clin Infect Dis* 2006, **42**:S35-9.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

15. Moise-Broder PA, Forrest A, Birmingham MC, Schentag JJ: **Pharmacodynamics of vancomycin and other antimicrobials in**

patients with *Staphylococcus aureus* lower respiratory tract infections. *Clin Pharmacokinet* 2004, **43**:925-42.

16. Kullar R, Davis SL, Levine DP, Rybak MJ: **Impact of vancomycin exposure on outcomes in patients with methicillin-resistant *Staphylococcus aureus* bacteremia: support for consensus guidelines suggested targets.** *Clin Infect Dis* 2011, **52**:975-81.
17. Mohr JF, Murray BE: **Point: Vancomycin is not obsolete for the treatment of infection caused by methicillin-resistant *Staphylococcus aureus*.** *Clin Infect Dis* 2007, **44**:1536-42.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

18. del Mar Fernandez de Gatta Garcia M, Revilla N, Calvo MV, Dominguez-Gil A, Sanchez Navarro A: **Pharmacokinetic/pharmacodynamic analysis of vancomycin in ICU patients.** *Intensive Care Med* 2007, **33**:279-85.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

19. Jones RN: **Microbiological features of vancomycin in the 21st century: minimum inhibitory concentration creep, bactericidal/static activity, and applied breakpoints to predict clinical outcomes or detect resistant strains.** *Clin Infect Dis* 2006, **42**(Suppl 1):S13-24.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

20. Sakoulas G, Moise-Broder PA, Schentag J, Forrest A, Moellering RC, Jr, Eliopoulos GM: **Relationship of MIC and bactericidal activity to efficacy of vancomycin for treatment of methicillin-resistant *Staphylococcus aureus* bacteremia.** *J Clin Microbiol* 2004, **42**:2398-402.
21. Moise PA, Smyth DS, El-Fawal N, Robinson DA, Holden PN, Forrest A, Sakoulas G: **Microbiological effects of prior vancomycin use in patients with methicillin-resistant *Staphylococcus aureus* bacteraemia.** *J Antimicrob Chemother* 2008, **61**:85-90.
22. Sieradzki K, Tomasz A: **Alterations of cell wall structure and metabolism accompany reduced susceptibility to vancomycin in an isogenic series of clinical isolates of *Staphylococcus aureus*.** *J Bacteriol* 2003, **185**:7103-10.
23. Sieradzki K, Tomasz A: **Inhibition of the autolytic system by vancomycin causes mimicry of vancomycin-intermediate *Staphylococcus aureus*-type resistance, cell concentration dependence of the MIC, and antibiotic tolerance in vancomycin-susceptible *S. aureus*.** *Antimicrob Agents Chemother* 2006, **50**:527-33.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

24. Pereira PM, Filipe SR, Tomasz A, Pinho MG: **Fluorescence ratio imaging microscopy shows decreased access of vancomycin to cell wall synthetic sites in vancomycin-resistant *Staphylococcus aureus*.** *Antimicrob Agents Chemother* 2007, **51**:3627-33.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

25. van Hal SJ, Wehrhahn MC, Barbagiannakos T, Mercer J, Chen D, Paterson DL, Gosbell IB: **Performance of various testing methodologies for detection of heteroresistant vancomycin-intermediate *Staphylococcus aureus* in bloodstream isolates.** *J Clin Microbiol* 2011, **49**:1489-94.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

26. Wang G, Hindler JF, Ward KW, Bruckner DA: **Increased vancomycin MICs for *Staphylococcus aureus* clinical isolates from a university hospital during a 5-year period.** *J Clin Microbiol* 2006, **44**:3883-6.
27. Holmes NE, Turnidge JD, Munckhof WJ, Robinson JO, Korman TM, O'Sullivan MV, Anderson TL, Roberts SA, Gao W, Christiansen KJ, Coombs GW, Johnson PD, Howden BP:

**Antibiotic Choice May Not Explain Poorer Outcomes in Patients With Staphylococcus aureus Bacteremia and High Vancomycin Minimum Inhibitory Concentrations.** *J Infect Dis* 2011, **204**:340-347.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

28. Liu C, Bayer A, Cosgrove SE, Daum RS, Fridkin SK, Gorwitz RJ, Kaplan SL, Karchmer AVW, Levine DP, Murray BE, J. Rybak M, Talan DA, Chambers HF: **Clinical Practice Guidelines by the Infectious Diseases Society of America for the Treatment of Methicillin-Resistant Staphylococcus Aureus Infections in Adults and Children.** *Clinical Infectious Diseases* 2011, **52**: e18-55.
29. Musta AC, Riederer K, Shemes S, Chase P, Jose J, Johnson LB, Khatib R: **Vancomycin MIC plus heteroresistance and outcome of methicillin-resistant Staphylococcus aureus bacteremia: trends over 11 years.** *J Clin Microbiol* 2009, **47**:1640-4.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

30. Lodise TP, Graves J, Evans A, Graffunder E, Helmecke M, Lomaestro BM, Stellrecht K: **Relationship between vancomycin MIC and failure among patients with methicillin-resistant Staphylococcus aureus bacteremia treated with vancomycin.** *Antimicrob Agents Chemother* 2008, **52**:3315-20.

F1000 Factor 7

Evaluated by George Sakoulas 18 Jan 2012, Paul Johnson 13 Nov 2008

31. Haque NZ, Zuniga LC, Peyrani P, Reyes K, Lamerato L, Moore CL, Patel S, Allen M, Peterson E, Wiemken T, Cano E, Mangino JE, Kett DH, Ramirez JA, Zervos MJ: **Improving Medicine through Pathway Assessment of Critical Therapy of Hospital-Acquired Pneumonia (IMPACT-HAP) Investigators: Relationship of vancomycin minimum inhibitory concentration to mortality in patients with methicillin-resistant Staphylococcus aureus hospital-acquired, ventilator-associated, or health-care-associated pneumonia.** *Chest* 2010, **138**:1356-62.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

32. Lodise TP, Miller CD, Graves J, Evans A, Graffunder E, Helmecke M, Stellrecht K: **Predictors of high vancomycin MIC values among patients with methicillin-resistant Staphylococcus aureus bacteraemia.** *J Antimicrob Chemother* 2008, **62**:1138-41.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

33. Lubin AS, Snyderman DR, Ruthazer R, Bide P, Golan Y: **Predicting high vancomycin minimum inhibitory concentration in methicillin-resistant Staphylococcus aureus bloodstream infections.** *Clin Infect Dis* 2011, **52**:997-1002.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

34. Sakoulas G, Alder J, Thauvin-Eliopoulos C, Moellering RC, Jr, Eliopoulos GM: **Induction of daptomycin heterogeneous susceptibility in Staphylococcus aureus by exposure to vancomycin.** *Antimicrob Agents Chemother* 2006, **50**:1581-5.

35. Kelley PG, Gao W, Ward PB, Howden BP: **Daptomycin non-susceptibility in vancomycin-intermediate Staphylococcus aureus (VISA) and heterogeneous-VISA (hVISA): implications for therapy after vancomycin treatment failure.** *J Antimicrob Chemother* 2011, **66**(5):1057-1060.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

36. van Hal SJ, Paterson DL, Gosbell IB: **Emergence of daptomycin resistance following vancomycin-unresponsive Staphylococcus**

**aureus bacteraemia in a daptomycin-naive patient—a review of the literature.** *Eur J Clin Microbiol Infect Dis* 2011, **30**:603-10.

37. Dhand A, Bayer AS, Pogliano J, Yang SJ, Bolaris M, Nizet V, Wang G, Sakoulas G: **Use of antistaphylococcal beta-lactams to increase daptomycin activity in eradicating persistent bacteremia due to methicillin-resistant Staphylococcus aureus: role of enhanced daptomycin binding.** *Clin Infect Dis* 2011, **53**:158-63.
38. Sakoulas G, Eliopoulos GM, Fowler VG, Jr, Moellering RC, Jr, Novick RP, Lucindo N, Yeaman MR, Bayer AS: **Reduced susceptibility of Staphylococcus aureus to vancomycin and platelet microbicidal protein correlates with defective autolysis and loss of accessory gene regulator (agr) function.** *Antimicrob Agents Chemother* 2005, **49**:2687-92.
39. Peleg AY, Monaga D, Pillai S, Mylonakis E, Moellering RC, Jr, Eliopoulos GM: **Reduced susceptibility to vancomycin influences pathogenicity in Staphylococcus aureus infection.** *J Infect Dis* 199:532-6.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

40. van Hal SJ, Jones M, Gosbell IB, Paterson DL: **Vancomycin heteroresistance is associated with reduced mortality in ST239 methicillin-resistant Staphylococcus aureus blood stream infections.** *PLoS One* 2011, **6**:e21217.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

41. Soriano A, Marco F, Martinez JA, Pisos E, Almela M, Dimova VP, Alamo D, Ortega M, Lopez J, Mensa J: **Influence of vancomycin minimum inhibitory concentration on the treatment of methicillin-resistant Staphylococcus aureus bacteremia.** *Clin Infect Dis* 2008, **46**:193-200.

F1000 Factor 10

Evaluated by George Sakoulas 18 Jan 2012, Richard Wunderink 30 Apr 2008, Ralph Corey 08 Apr 2008

42. Holmes NE, Turnidge JD, Munckhof WJ, Robinson JO, Korman TM, O'Sullivan MVN, Anderson TL, Roberts SA, Gao W, Christiansen KJ, Coombs GW, Johnson PDR, Howden BP: **Antibiotic choice may not explain poorer outcomes in patients with Staphylococcus aureus bacteremia and high vancomycin minimum inhibitory concentrations.** *J Infect Dis* 2011, **204**:340-7.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

43. Robert J, Bismuth R, Jarlier V: **Decreased susceptibility to glycopeptides in methicillin-resistant Staphylococcus aureus: a 20 year study in a large French teaching hospital, 1983-2002.** *J Antimicrob Chemother* 2006, **57**:506-10.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

44. Steinkraus G, White R, Friedrich L: **Vancomycin MIC creep in non-vancomycin-intermediate Staphylococcus aureus (VISA), vancomycin-susceptible clinical methicillin-resistant S. aureus (MRSA) blood isolates from 2001-05.** *J Antimicrob Chemother* 2007, **60**:788-94.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

45. Holmes RL, Jorgensen JH: **Inhibitory activities of 11 antimicrobial agents and bactericidal activities of vancomycin and daptomycin against invasive methicillin-resistant Staphylococcus aureus isolates obtained from 1999 through 2006.** *Antimicrob Agents Chemother* 2008, **52**:757-60.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

46. Alos JI, Garcia-Canas A, Garcia-Hierro P, Rodriguez-Salvanes F: **Vancomycin MICs did not creep in Staphylococcus aureus**

isolates from 2002 to 2006 in a setting with low vancomycin usage. *J Antimicrob Chemother* 2008, **62**:773-5.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

47. Sader HS, Fey PD, Limaye AP, Madinger N, Pankey G, Rahal J, Rybak MJ, Snyderman DR, Steed LL, Waites K, Jones RN: **Evaluation of vancomycin and daptomycin potency trends (MIC creep) against methicillin-resistant Staphylococcus aureus isolates collected in nine U.S. medical centers from 2002 to 2006.** *Antimicrob Agents Chemother* 2009, **53**:4127-32.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

48. Karas JA, Enoch DA, Fish D, Foweraker JE: **Vancomycin susceptibility of methicillin-resistant Staphylococcus aureus (MRSA) bacteraemia isolates from two UK hospitals over a decade.** *Int J Antimicrob Agents* 2010, **36**:189-90.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

49. Ho PL, Lo PY, Chow KH, Lau EH, Lai EL, Cheng VC, Kao RY: **Vancomycin MIC creep in MRSA isolates from 1997 to 2008 in a healthcare region in Hong Kong.** *J Infect* 2010, **60**:140-5.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

50. Moise-Broder PA, Sakoulas G, Eliopoulos GM, Schentag JJ, Forrest A, Moellering RC, Jr: **Accessory gene regulator group II polymorphism in methicillin-resistant Staphylococcus aureus is predictive of failure of vancomycin therapy.** *Clin Infect Dis* 2004, **38**:1700-5.
51. Rhee KY, Gardiner DF, Charles M: **Decreasing in vitro susceptibility of clinical Staphylococcus aureus isolates to vancomycin at the New York Hospital: quantitative testing redux.** *Clin Infect Dis* 2005, **40**:1705-6.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

52. Maclayton DO, Suda KJ, Coval KA, York CB, Garey KW: **Case-control study of the relationship between MRSA bacteremia**

**with a vancomycin MIC of 2 microg/mL and risk factors, costs, and outcomes in inpatients undergoing hemodialysis.** *Clin Ther* 2006, **28**:1208-16.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

53. Hidayat LK, Hsu DI, Quist R, Shriner KA, Wong-Beringer A: **High-dose vancomycin therapy for methicillin-resistant Staphylococcus aureus infections: efficacy and toxicity.** *Arch Intern Med* 2006, **166**:2138-44.
54. Neoh HM, Hori S, Komatsu M, Oguri T, Takeuchi F, Cui L, Hiramatsu K: **Impact of reduced vancomycin susceptibility on the therapeutic outcome of MRSA bloodstream infections.** *Ann Clin Microbiol Antimicrob* 2007, **6**:13.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

55. Yoon YK, Kim JY, Park DW, Sohn JW, Kim MJ: **Predictors of persistent methicillin-resistant Staphylococcus aureus bacteraemia in patients treated with vancomycin.** *J Antimicrob Chemother* 2010, **65**:1015-8.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

56. Wang JL, Wang JT, Sheng WH, Chen YC, Chang SC: **Nosocomial methicillin-resistant Staphylococcus aureus (MRSA) bacteremia in Taiwan: mortality analyses and the impact of vancomycin, MIC = 2 mg/L, by the broth microdilution method.** *BMC Infect Dis* 2010, **10**:159.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012

57. Choi EY, Huh JW, Lim CM, Koh Y, Kim SH, Choi SH, Kim YS, Kim MN, Hong SB: **Relationship between the MIC of vancomycin and clinical outcome in patients with MRSA nosocomial pneumonia.** *Intensive Care Med* 2011, **37**:639-47.

F1000 Factor 6

Evaluated by George Sakoulas 18 Jan 2012