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Vancomycin 24-Hour Area under the Curve/Minimum Bactericidal Concentration Ratio as a Novel Predictor of Mortality in Methicillin-Resistant *Staphylococcus aureus* Bacteremia

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While previous studies have examined the association between vancomycin (VAN) exposure and MIC with regard to outcomes in methicillin-resistant *Staphylococcus aureus* bacteremia (MRSA-B), none have explored if a relationship exists with the VAN minimum bactericidal concentration (MBC). The objective of this study was to evaluate the VAN 24-h area under the curve (AUC_{24})/MBC ratio as a pharmacodynamic predictor of mortality. This retrospective cohort study included patients treated with VAN for MRSA-B with the primary outcome of 30-day all-cause mortality. Data collected included patient demographics, comorbidities, antimicrobial treatment data, therapeutic drug levels, and laboratory and microbiological data. Vancomycin MICs and MBCs were determined by Etest (MIC only) and broth microdilution (BMD). The vancomycin AUC_{24} was determined by pharmacokinetic maximum *a posteriori* probability Bayesian (MAP-Bayesian) analysis. The most significant breakpoint for 30-day mortality was determined by classification and regression tree (CART) analysis. The association between pharmacodynamic parameters (VAN AUC_{24}/MIC_{BMD}, VAN AUC_{24}/MIC_{Etest}, and AUC_{24}/MBC_{BMD}) and mortality were determined by χ^2 and multivariable Poisson regression. Overall mortality in this cohort ($n = 53$) was 20.8% ($n = 11/53$), and all corresponding MRSA blood isolates were VAN susceptible (MIC range, 0.5 to 2 μg/ml; MIC_{50}, 1 μg/ml; MIC_{90}, 1 μg/ml). The CART-derived breakpoints for mortality were 176 (VAN AUC_{24}/MBC) and 334 (VAN AUC_{24}/MIC_{BMD}). In multivariable analysis, the association between a VAN AUC_{24}/MBC of ≥176 and survival persisted, but VAN AUC_{24}/MIC_{BMD} values (≥334 or ≥400) were not associated with improved mortality. In conclusion, VAN AUC_{24}/MBC was a more important predictor of 30-day mortality than VAN AUC_{24}/MIC for MRSA-B.

The incidence of methicillin-resistant *Staphylococcus aureus* (MRSA) infections in health care and community settings has increased in recent years (1). Vancomycin (VAN) has been a mainstay in the armamentarium against MRSA for nearly half a century, but changes in susceptibility patterns have presented new treatment challenges (2). The gradual reduction in susceptibility of MRSA isolates to VAN occurring over the last 2 decades has been associated with increased treatment failure and mortality (3, 4). While the relationship between VAN MIC and clinical outcomes in MRSA bacteremia (MRSA-B) is well established, the importance of VAN minimum bactericidal concentration (MBC) is less clear (5).

Multiple studies have highlighted the importance of bactericidal therapy in treating *S. aureus* bacteremia (SAB). Previous research has demonstrated that *in vitro* 24-h bactericidal activity by VAN is associated with a decrease in the duration of MRSA-B clinically compared to *in vitro* bacteriostatic activity (6). Additionally, decreased *in vitro* VAN killing activity over 72 h also has been associated with an increase in 30-day mortality in patients with MRSA-B treated with VAN (7). Sakoulas et al. reported an association between elevated VAN MIC and treatment failure but also noted that reduced bactericidal activity had a similar effect (8). Lastly, multiple studies have demonstrated improved clinical outcomes with β-lactam therapy compared to those with VAN for methicillin-susceptible SAB, which may be partially attributed to the superior killing activity of β-lactams compared to that of VAN (9, 10). Thus, there is evidence to suggest that VAN bactericidal activity plays an influential role in clinical outcomes of SAB, including MRSA-B.

Consensus recommendations support a VAN area under the curve (AUC)/MIC ratio of ≥400 as a pharmacodynamic predictor of therapeutic effectiveness for MRSA infections (11). An AUC/MIC ratio of ≥421 also has been associated with clinical success in MRSA-B (12). Due to the trend toward improved outcomes with increased bactericidal activity of VAN, we hypothesized that VAN exposure as a function of MBC rather than MIC may be a more important predictor of VAN effectiveness in the treatment of MRSA-B. Therefore, the objectives of this study were to (i) evaluate the association between pharmacodynamic parameters (VAN AUC_{24}/MIC ratio determined by broth microdilution [MIC_{BMD}], VAN AUC_{24}/MIC ratio determined by Etest [MIC_{Etest}], and VAN AUC_{24}/MBC ratio) and 30-day mortality and (ii) quantify the VAN AUC_{24}/MBC threshold associated with an increased probability of mortality in a cohort of hospitalized patients with MRSA-B.

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MATERIALS AND METHODS

Study population and data sources. This was a retrospective study of patients treated for MRSA-B at the University of Kansas Hospital, a tertiary-care academic medical center, from September 2012 through June 2014. Inclusion criteria were (i) age of ≥18 years, (ii) positive blood culture for MRSA, and (iii) intravenous treatment with VAN. Patients were excluded if they were on hemodialysis, had polymicrobial bacteremia at onset, or were treated with VAN for <48 h. For patients with multiple clinical MRSA blood isolates during the study period, only the first isolate and corresponding clinical case of MRSA-B were evaluated. Clinical data were collected by retrospective chart review. Variables abstracted included baseline patient demographics, setting of bacteremia onset (community versus hospital-acquired [admission within ≥72 h]), comorbidities, Charlson comorbidity index, Pitt bacteremia score (PBS), acute physiology and chronic health evaluation II (APACHE II) score, laboratory data (including VAN therapeutic drug monitoring), microbiology data, antimicrobial treatment data (including previous VAN exposure within 30 days), and vital signs. The focus of MRSA-B was determined as documented by a treating physician and categorized according to mortality risk (13). Immunosuppression was defined as chronic steroid (equivalent to ≥20 mg prednisone) or antineoplastic use, neutropenia, or leukopenia. The primary clinical outcome was 30-day all-cause mortality. This study was approved by the University of Kansas Medical Center institutional review board.

AUC determination. Vancomycin AUC values were estimated with ADAPT 5 software (University of Southern California, Los Angeles, CA, USA). Using data from a previously published 2-compartment VAN model, the mean parameter vector and full covariance matrix were embedded into the PRIOR subroutine of ADAPT 5 (14, 15). For each individual patient, the pharmacokinetic parameter values were estimated using the maximum *a posteriori* probability (MAP) procedure. After the MAP-Bayesian step, values of the AUC for 0 to 72 h (AUC_{0-72}) and AUC_{0-96} were estimated for each patient. For the purposes of these analyses, the AUC at 72 to 96 h (AUC_{24}) of therapy was used, because this is the 24-h exposure variable that most closely resembles a steady-state AUC (16). The vancomycin AUC_{24} was calculated as the difference in integrated AUC_{0-72} and AUC_{0-96} values.

Microbiological analysis. (i) Bacterial strains. Clinical *S. aureus* blood isolates from MRSA-B cases encountered during the study period were stored at −70°C and passed for 3 consecutive days on tryptic soy agar (TSA) to ensure uniform metabolic activity prior to testing.

(ii) Antimicrobials. Vancomycin was commercially purchased from Sigma-Aldrich Co. (St. Louis, MO, USA). Stock solutions of VAN were prepared daily.

(iv) Media. Mueller-Hinton II broth (Becton-Dickinson, Sparks, MD, USA) was used as media for susceptibility testing. Brain heart infusion agar (Becton-Dickinson, Sparks, MD, USA) was used as the medium for the Etest. Tryptic soy agar (Becton-Dickinson, Sparks, MD, USA) was used for colony counts.

(v) Susceptibility testing. Vancomycin MICs were determined by broth microdilution (BMD) at concentrations of 0.125 to 64 μg/ml at a standard inoculum of approximately 1×10^6 CFU/ml according to Clinical and Laboratory Standards Institute (CLSI) guidelines (17). The vancomycin MIC also was determined for all isolates by Etest according to manufacturer recommendations (bioMérieux, Marcy l'Etoile, France). *Staphylococcus aureus* ATCC 25923 was used as the control strain, and each test was performed in duplicate to ensure reproducibility.

(vi) MBC determination. Vancomycin MBCs were determined by the microdilution method according to CLSI guidelines (18). To account for antibiotic carryover, 100-μl aliquots of all wells with no visible growth after 24 h of incubation at 35°C (i.e., above the MIC_{BMD} endpoint) were streaked onto TSA, allowed to visibly dry at room temperature, and cross-streaked using a sterile cotton-tipped swab (18, 19). The vancomycin MBC was defined as the lowest concentration with ≥99.9% killing after 24 h of incubation at 35°C. Vancomycin tolerance was defined as a MBC/MIC ratio of ≥32.

Statistical analysis. Categorical variables were compared by χ^2 or two-tailed Fisher's exact test, and continuous variables were compared by *t* test or Mann-Whitney *U* test. The AUC_{24} variable was divided by MIC_{BMD}, MIC_{Etest}, and MBC, respectively, to compute the VAN pharmacodynamic variables of interest. Classification and regression tree (CART) analyses were used to identify the breakpoints in these pharmacodynamic parameters associated with an increased probability of the primary outcome (30-day mortality). A secondary analysis examining the relationship between VAN exposure within the preceding 30 days and VAN MIC_{BMD} and MBC was performed using the Cochran-Armitage trend test.

The association between VAN pharmacodynamic parameters and clinical outcomes was evaluated using backward stepwise Poisson regression with CART-derived breakpoints forced into respective models. Variables which were associated with the pharmacodynamic parameter of interest or 30-day mortality ($P < 0.2$) were entered into the model, and only those that confounded the relationship between the pharmacodynamic breakpoint and 30-day mortality (≥10% change in the associated risk ratio) were retained in the final parsimonious model. The association between the guideline concordant VAN AUC_{24}/MIC_{BMD} target of ≥400 and 30-day mortality also was evaluated in this cohort (11). Time-to-event analyses were conducted for 30-day mortality using the Kaplan-Meier method and log-rank test. Survivors were right-censored at the end of treatment with VAN, and backward stepwise Cox regression was performed. Statistical analyses were performed using SAS (version 9.2; SAS Institute, Inc., Cary, NC, USA), and a two-tailed *P* value of <0.05 was considered statistically significant for all tests.

RESULTS

During the study period, a total of 100 patients with MRSA-B who were treated with VAN were identified. Patients were excluded due to treatment with VAN for <48 h ($n = 13$) and due to hemodialysis ($n = 34$). The final analysis included a total of 53 patients. All corresponding clinical MRSA blood isolates were VAN susceptible (MIC range, 0.5 to 2 μg/ml; MIC_{50}, 1 μg/ml; MIC_{90}, 1 μg/ml). Among these isolates, 13/53 (24.5%) were VAN tolerant (MBC range, 0.5 to 64 μg/ml; MBC_{50}, 1 μg/ml; MBC_{90}, 64 μg/ml). Additionally, 25/53 (47.2%) of clinical MRSA isolates had dissociated MIC_{BMD} and MBC values.

Overall, 30-day mortality was 20.8% ($n = 11/53$). Factors associated with 30-day mortality among MRSA-B patients treated with VAN are included in Table 1. In univariable analysis, mean age, an age of ≥85 years, previous VAN exposure within 30 days, intensive care unit (ICU) admission, high-risk infection focus, malignancy, median Charlson comorbidity index, PBS, and APACHE II score were significantly associated with increased 30-day mortality. Conversely, a medium-risk focus of infection was associated with decreased 30-day mortality.

In this cohort, the mean duration of VAN therapy was 8.73 days (standard deviations [SD], 5.23 days). The mean VAN AUC_{24} at steady state [$AUC_{24(ss)}$] was 681 mg · h/liter (SD, 331 mg · h/liter). The median VAN $AUC_{24(ss)}$ was 661.7 mg · h/liter (interquartile range, 440.6 to 834.1 mg · h/liter). The CART-derived 30-day mortality breakpoints were 334 for VAN AUC_{24}/MIC_{BMD} ratio and 176 for VAN AUC_{24}/MBC ratio. Classification and regression tree analysis yielded no optimal breakpoint for the association between AUC_{24}/MIC_{Etest} and 30-day mortality, so no further analyses were conducted for this parameter. In univariable analysis, mortality was significantly lower when the VAN AUC_{24}/

TABLE 1 Factors associated with 30-day mortality among MRSA-B patients treated with VAN

Factor[a]	Value for patients with: No mortality ($n = 42$)	Mortality ($n = 11$)	*P* value
Age (yr), means ± SD	58.5 ± 15.9	70.6 ± 16.6	0.031
≥65 yr, *n* (%)	17 (40.5)	7 (63.6)	0.170
≥85 yr, *n* (%)	1 (2.4)	3 (27.3)	0.025
Female gender, *n* (%)	15 (35.7)	3 (27.3)	0.730
Previous VAN exposure within 30 days, *n* (%)	4 (9.5)	5 (45.5)	0.031
Hospital-acquired MRSA-B, *n* (%)	12 (28.6)	6 (54.5)	0.105
ICU, *n* (%)	10 (23.8)	6 (54.5)	0.048
Sepsis, *n* (%)	27 (64.3)	9 (81.8)	0.267
Septic shock, *n* (%)	6 (14.3)	2 (18.2)	0.665
Immunosuppression, *n* (%)	10 (23.8)	4 (36.4)	0.453
MRSA-B focus			
High risk, *n* (%)	10 (23.8)	8 (72.7)	0.002
Medium risk, *n* (%)	26 (61.9)	2 (18.2)	0.016
Low risk, *n* (%)	6 (14.3)	1 (9.1)	1.000
MRSA bacteriuria, *n* (%)	4 (9.5)	1 (9.1)	1.000
Diabetes mellitus, *n* (%)	15 (35.7)	4 (36.4)	0.968
Alcoholism, *n* (%)	4 (9.5)	1 (9.1)	1.000
Cirrhosis, *n* (%)	3 (7.1)	1 (9.1)	1.000
Congestive heart failure, *n* (%)	3 (7.1)	2 (18.2)	0.275
Malignancy, *n* (%)	8 (19.0)	7 (63.6)	0.003
HIV positive, *n* (%)	1 (2.4)	0 (0.0)	1.000
Chronic obstructive pulmonary disease, *n* (%)	5 (11.9)	2 (18.2)	0.626
Charlson comorbidity index, median (IQR)	6 (3–7)	7 (6–9)	0.017
PBS, median (IQR)	1 (0–3)	4 (1–6)	0.028
PBS ≥ 4, *n* (%)	7 (16.7)	6 (54.5)	0.009
APACHE II score, median (IQR)	11 (7–14)	17 (14–25)	0.002

[a] IQR, interquartile range.

TABLE 2 Patient characteristics by CART-derived AUC_{24}/MBC breakpoint for VAN in MRSA-B

Characteristic[a]	Value by AUC_{24}/MBC category: <176 ($n = 21$)	≥176 ($n = 32$)	*P* value
Age (years), means ± SD	67.6 ± 15.5	55.2 ± 15.9	0.007
≥65 yr, *n* (%)	13 (61.9)	11 (34.4)	0.049
≥85 yr, *n* (%)	4 (19.0)	0 (0.0)	0.020
Female gender, *n* (%)	6 (28.6)	12 (37.5)	0.502
Previous VAN exposure within 30 days, *n* (%)	7 (33.3)	2 (6.2)	0.021
Hospital-acquired MRSA-B, *n* (%)	11 (52.4)	7 (21.9)	0.022
ICU, *n* (%)	8 (38.1)	8 (25.0)	0.310
Sepsis, *n* (%)	16 (76.2)	20 (62.5)	0.296
Septic shock, *n* (%)	6 (28.6)	2 (6.2)	0.047
Immunosuppression, *n* (%)	4 (19.0)	10 (31.2)	0.324
MRSA-B focus, *n* (%)			
High risk	10 (47.6)	8 (25.0)	0.089
Medium risk	7 (33.3)	21 (65.6)	0.021
Low risk	4 (19.0)	3 (9.4)	0.415
MRSA bacteriuria, *n* (%)	2 (9.5)	2 (9.4)	1.000
Diabetes mellitus, *n* (%)	7 (33.3)	12 (37.5)	0.757
Alcoholism, *n* (%)	1 (4.8)	4 (12.5)	0.637
Cirrhosis, *n* (%)	0 (0.0)	4 (12.5)	0.143
Congestive heart failure, *n* (%)	5 (23.8)	0 (0.0)	0.007
Malignancy, *n* (%)	7 (33.3)	8 (25.0)	0.546
HIV positive, *n* (%)	0 (0.0)	1 (3.1)	1.000
Chronic obstructive pulmonary disease, *n* (%)	4 (19.0)	3 (9.4)	0.415
Charlson comorbidity index, median (IQR)	7 (5–9)	6 (3–7)	0.058
PBS, median (IQR)	2 (1–4)	2 (0–4)	0.528
PBS ≥ 4, *n* (%)	5 (23.8)	8 (25.0)	0.922
APACHE II, median (IQR)	13 (9–18)	10 (7–15)	0.084

[a] IQR, interquartile range.

MIC_{BMD} was ≥334 (7.7% [$n = 2/26$] versus 33.3% [$n = 9/27$]; $P = 0.021$) and when the VAN AUC_{24}/MIC_{BMD} was ≥400 (14.3% [$n = 6/42$] versus 45.5% [$n = 5/11$]; $P = 0.037$). Mortality also was significantly lower when the VAN AUC_{24}/MBC was ≥176 (9.4% [$n = 3/32$] versus 38.1% [$n = 8/21$]; $P = 0.017$). Comparisons of baseline patient characteristics by CART-derived breakpoints are displayed in Tables 2 and 3. As shown, there were many significant differences between groups with regard to these characteristics. In the secondary trend analysis, prior VAN exposure within 30 days was associated with a linear increase in VAN MIC_{BMD} ($P = 0.033$) and VAN MBC ($P = 0.001$).

The association between pharmacodynamic parameters and 30-day mortality were evaluated in multivariable Poisson regression analyses (Table 4). As shown, the association between a VAN AUC_{24}/MBC ratio of <176 and 30-day mortality persisted after adjusting for age, malignancy, PBS, and cirrhosis (Table 4, model 1). After adjusting for confounding factors, VAN AUC_{24}/MIC_{BMD} ratios of <334 and <400 were not significantly associated with mortality in this cohort (Table 4, models 2 and 3).

In Kaplan-Meier analysis, the CART-derived VAN AUC_{24}/MBC breakpoint of ≥176 was significantly associated with improved survival ($P = 0.011$ by log-rank test). When evaluated by multivariable Cox regression, the association between an AUC_{24}/MBC ratio of ≥176 and improved survival persisted (adjusted hazard ratio [aHR], 7.32; 95% confidence interval [CI], 1.66 to 32.29; $P = 0.009$) after adjusting for APACHE II score (aHR, 1.09; 95% CI, 1.01 to 1.19; $P = 0.042$) and PBS of ≥4 (aHR, 6.80, 95% CI, 1.68 to 27.60; $P = 0.007$).

DISCUSSION

The present investigation aimed to evaluate the relationship between various VAN pharmacodynamic parameters and 30-day mortality in MRSA-B. Primarily, we sought to derive an associated VAN AUC_{24}/MBC breakpoint and evaluate this breakpoint alongside other pharmacodynamic parameters in a series of multivariable analyses. To our knowledge, this is the first study to establish a VAN AUC_{24}/MBC breakpoint for 30-day mortality. Multiple studies have derived VAN AUC/MIC breakpoints, and current consensus recommendations target a VAN AUC/MIC ratio of ≥400 to maximize therapeutic effectiveness, although this goal is only clinically feasible with some MIC values in the suscep-

TABLE 3 Patient characteristics by CART-derived AUC_{24}/MIC_{BMD} breakpoint for VAN in MRSA-B[a]

Characteristic	Value by AUC_{24}/MIC_{BMD} category		*P* value
	<334 (*n* = 27)	≥334 (*n* = 26)	
Age (yr), means ± SD	63.5 ± 19.1	56.5 ± 13.5	0.136
≥65 yr, *n* (%)	14 (51.9)	10 (38.5)	0.328
≥85 yr, *n* (%)	4 (14.8)	0 (0.0)	0.111
Female gender, *n* (%)	8 (29.6)	10 (38.5)	0.497
Previous VAN exposure within 30 days, *n* (%)	8 (29.6)	1 (3.8)	0.024
Hospital-acquired MRSA-B, *n* (%)	12 (44.4)	6 (23.1)	0.101
ICU, *n* (%)	10 (37.0)	6 (23.1)	0.268
Sepsis, *n* (%)	20 (74.1)	16 (61.5)	0.328
Septic shock, *n* (%)	7 (25.9)	1 (3.8)	0.050
Immunosuppression, *n* (%)	6 (22.2)	8 (30.8)	0.480
MRSA-B focus, *n* (%)			
High risk	12 (44.4)	6 (23.1)	0.101
Medium risk	11 (40.7)	17 (65.4)	0.072
Low risk	4 (14.8)	3 (11.5)	0.725
MRSA bacteriuria, *n* (%)	2 (7.4)	3 (11.5)	0.669
Diabetes mellitus, *n* (%)	9 (33.3)	10 (38.5)	0.697
Alcoholism, *n* (%)	1 (3.7)	4 (15.4)	0.192
Cirrhosis, *n* (%)	0 (0.0)	4 (15.4)	0.051
Congestive heart failure, *n* (%)	5 (18.5)	0 (0.0)	0.051
Malignancy, *n* (%)	9 (33.3)	6 (23.1)	0.407
HIV positive, *n* (%)	1 (3.7)	0 (0.0)	1.000
Chronic obstructive pulmonary disease, *n* (%)	4 (14.8)	3 (11.5)	0.725
Charlson comorbidity index, median (IQR)	7 (5–9)	5 (3–7)	0.099
PBS, median (IQR)	2 (1–4)	2 (0–3)	0.238
PBS ≥ 4, *n* (%)	7 (25.9)	6 (23.1)	0.810
APACHE II, median (IQR)	13 (6–17)	11 (8–14)	0.232

[a] IQR, interquartile range.

TABLE 4 Poisson regression models for 30-day mortality by pharmacodynamic parameter among MRSA-B patients

Model and characteristic	Adjusted risk ratio (95% CI)	*P* value
Model 1		
VAN $AUC_{24}/MBC < 176$	3.68 (1.17–11.56)	0.025
Age ≥85 yr	4.24 (1.75–10.23)	0.001
Malignancy	5.62 (2.31–13.65)	<0.001
PBS ≥ 4	3.63 (1.48–8.67)	0.005
Cirrhosis	6.33 (0.60–67.38)	0.120
Model 2		
VAN $AUC_{24}/MIC_{BMD} < 334$	5.12 (0.85–30.67)	0.074
Malignancy	3.70 (1.25–10.97)	0.019
MRSA pneumonia	3.56 (1.13–11.26)	0.031
Cirrhosis	9.88 (0.72–34.79)	0.086
Model 3		
VAN $AUC/MIC_{BMD} < 400$	1.95 (0.92–4.15)	0.084
Malignancy	3.89 (1.31–11.53)	0.014
MRSA pneumonia	3.59 (1.16–11.11)	0.026
Cirrhosis	3.73 (0.39–36.07)	0.255

tible range (11, 12, 16). In the present investigation, a VAN AUC_{24}/MIC_{BMD} ratio of ≥334 and VAN AUC_{24}/MIC_{BMD} ratio of ≥400 both were significantly associated with improved mortality in univariable analysis. However, the CART-derived VAN AUC_{24}/MBC breakpoint of 176 was the only pharmacodynamic parameter that was significantly associated with improved mortality after adjusting for confounding factors. A vancomycin AUC_{24}/MBC ratio of ≥176 also was associated with improved survival in Kaplan-Meier analysis and multivariable Cox regression.

The association between VAN MIC and clinical outcomes has been evaluated exhaustively in the literature, while little attention has been paid to the potential importance of bactericidal activity (3, 5). Studies that have evaluated VAN bactericidal activity have noted an apparent association between reduced bactericidal activity and worse clinical outcomes (7, 8). Although VAN MIC and MBC values are correlated, it is not uncommon for these values to be either slightly or greatly dissociated (20–22). In the present study, nearly half of corresponding MRSA isolates had some level of dissociation between VAN MIC_{BMD} and MBC. Because bactericidal activity is preferred in the treatment of MRSA-B, a VAN AUC_{24}/MIC target theoretically would be less likely to predict therapeutic effectiveness when MIC and MBC values are dissociated (6). Vancomycin AUC_{24}/MBC estimations would allow for the differentiation of these cases and potentially provide a more sensitive predictor of VAN effectiveness. The results of the present study appear to support this logic.

The finding that prior VAN exposure can influence VAN activity as well as clinical outcomes in MRSA-B is consistent with previous research (23, 24). This observation potentially has significant clinical implications in the treatment of MRSA-B, as previous VAN exposure was associated with increased VAN MIC_{BMD} and VAN MBC and a lower probability of achieving optimal AUC_{24}/MIC_{BMD} and AUC_{24}/MBC ratios in univariable analysis. Therefore, alternatives to VAN may be warranted for treatment of MRSA-B in patients with recent VAN exposure, even if the corresponding isolate tests susceptible.

The utility of the VAN AUC_{24}/MBC ratio as a pharmacodynamic predictor of VAN effectiveness likely is dependent on the prevalence of dissociated VAN MIC and MBC values, including VAN tolerance. The prevalence of VAN tolerance is approximately 20% among MRSA blood isolates, although tolerance is as high as 43% in some institutions (21). Therefore, the proportion of VAN tolerant isolates observed in the present study is consistent with previous research and likely similar to tolerance rates typically found in U.S. hospitals. In institutions where VAN tolerance is more common, the difference between AUC_{24}/MIC and AUC_{24}/MBC targets likely would be even more profound. It should be noted that $AUC_{72–96}$ was used in this study as a measure of 24-h VAN exposure. The decision to select VAN $AUC_{72–96}$ was informed by the literature (16). Furthermore, the vast majority of patients will have achieved steady-state concentrations of VAN and most clinicians will have commenced VAN therapeutic drug monitoring by this time point. Unfortunately, the literature is not clear regarding the optimal VAN AUC time interval for predicting clinical outcomes, and this question should be addressed in future studies (16, 25–27).

The present investigation has a number of noteworthy limitations. Most notably, this was a retrospective analysis and suffers from the limitations of such a design. Although the VAN AUC_{24}/MIC_{BMD} ratio was not predictive of 30-day mortality in this cohort after adjusting for other factors, there is a concern for type II error due to the relatively small number of patients included. It is important to consider that the utility of the VAN AUC_{24}/MBC ratio as a pharmacodynamic predictor of VAN effectiveness likely will depend on institution-specific VAN tolerance rates, which may not be known. Previous research has demonstrated that other phenotypic characteristics of MRSA, such as biofilm formation and susceptibility to host defense cationic peptide, play an influential role in clinical outcomes of MRSA-B (28). Although the purpose of this study was to investigate the relationship between VAN AUC_{24}/MBC ratios and clinical outcomes, it is important to consider the possible influence of unmeasured phenotypic and genotypic microbiological characteristics (28, 29). Host defense cationic peptide phenotypes have been associated with reduced susceptibility to vancomycin and defective autolytic mechanisms (30). These phenotypic changes, in addition to enhanced biofilm production, have been associated with poor clinical outcomes in MRSA-B, including persistent infection (20, 28, 30). Additionally, the cause of death was not known and we were unable to ascertain attributable mortality. Important strengths of this study were the utilization of a method for MBC testing which appropriately accounted for antibiotic carryover, use of a MAP-Bayesian approach to determine VAN AUC_{24}, and application of CART analysis to define a novel VAN AUC_{24}/MBC breakpoint for 30-day mortality.

In summary, the VAN AUC_{24}/MBC was a more important predictor of 30-day mortality than the VAN AUC_{24}/MIC_{BMD} in a cohort of patients with MRSA-B. The VAN AUC_{24}/MBC breakpoint most predictive of survival was $\geq$176. Overall, the results of this study add to the growing body of literature underscoring the importance of VAN bactericidal activity in the treatment of MRSA-B. Alternatives to VAN may be warranted in cases of MRSA-B where the achievement of optimal AUC_{24}/MBC is unlikely, such as in patients with VAN-tolerant clinical isolates or recent VAN exposure. This research should be replicated in a larger analysis and at institutions with various rates of VAN tolerance.

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