Comparative Pharmacodynamics of Telavancin and Vancomycin in the Neutropenic Murine Thigh and Lung Infection Models against *Staphylococcus aureus*

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ABSTRACT The pharmacodynamics of telavancin and vancomycin were compared using neutropenic murine thigh and lung infection models. Four *Staphylococcus aureus* strains were included. The telavancin MIC ranged from 0.06 to 0.25 mg/liter, and the vancomycin MIC ranged from 1 to 4 mg/liter. The plasma pharmacokinetics of escalating doses (1.25, 5, 20, and 80 mg/kg of body weight) of telavancin and vancomycin were linear over the dose range. Epithelial lining fluid (ELF) pharmacokinetics for each drug revealed that penetration into the ELF mirrored the percentage of the free fraction (the fraction not protein bound) in plasma for each drug. Telavancin (0.3125 to 80 mg/kg/6 h) and vancomycin (0.3125 to 1,280 mg/kg/6 h) were administered by the subcutaneous route in treatment studies. Dose-dependent bactericidal activity against all four strains was observed in both models. A sigmoid maximum-effect model was used to determine the area under the concentration-time curve (AUC)/MIC exposure associated with net stasis and 1-log₁₀ kill relative to the burden at the start of therapy. The 24-h plasma free drug AUC (fAUC)/MIC values associated with stasis and 1-log kill were remarkably congruent. Net stasis for telavancin was noted at fAUC/MIC values of 83 and 40.4 in the thigh and lung, respectively, and 1-log kill was noted at fAUC/MIC values of 215 and 76.4, respectively. For vancomycin, the fAUC/MIC values for stasis were 77.9 and 45.3, respectively, and those for 1-log kill were 282 and 113, respectively. The 24-h ELF total drug AUC/MIC targets in the lung model were very similar to the 24-h plasma free drug AUC/MIC targets for each drug. Integration of human pharmacokinetic data for telavancin, the results of the MIC distribution studies, and the pharmacodynamic targets identified in this study suggests that the current dosing regimen of telavancin is optimized to obtain drug exposures sufficient to treat *S. aureus* infections.

KEYWORDS epithelial lining fluid, pharmacodynamics, pneumonia, telavancin, vancomycin

Telavancin (TD-6424) is a semisynthetic lipoglycopeptide with enhanced activity against Gram-positive bacteria, including multidrug-resistant Gram-positive organisms (1). Telavancin has a dual mechanism of action targeting both peptidoglycan synthesis and cell membrane function, which is purported to be responsible for its improved bactericidal activity compared to that of vancomycin (2, 3). Efficacy has been demonstrated in clinical trials in patients with complicated skin and skin structure infection (cSSSI) and hospital-acquired pneumonia (hospital-acquired bacterial pneumonia/ventilator-acquired bacterial pneumonia [HABP/VABP]) due to Gram-positive pathogens (4, 5). Subsequently, telavancin was also demonstrated to have efficacy for the treatment of

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TABLE 1 *In vitro* activities of telavancin and comparator agents against select *S. aureus* isolates determined using CLSI methods

Organism	MIC (mg/liter)					Phenotype
	Telavancin	Vancomycin	Daptomycin	Linezolid	Dalbavancin	
29213	0.0625	1	0.5	2	0.0625	MSSA
33591	0.125	1	0.5	1	0.125	MRSA
LSI653	0.125	2	1	0.5	0.25	MRSA
LSI1856	0.25	4	1	2	0.25	VISA

patients with cSSSI and for patients with HABP/VABP who had concurrent *Staphylococcus aureus* bacteremia (6).

Preclinical pharmacodynamic (PD) models are critical components in optimizing dosing regimen design and setting susceptibility breakpoints (7). Numerous *in vivo* studies with telavancin demonstrating its promising activity have been performed (8); however, pharmacodynamic analyses are sparse (9). Therefore, the following studies were designed to characterize the *in vivo* pharmacokinetic (PK)/PD characteristics of telavancin and vancomycin in neutropenic murine thigh and lung infection models. Studies included those designed to identify the magnitude of the PK/PD index required for efficacy among multiple *S. aureus* isolates, including beta-lactam-resistant and vancomycin-intermediate strains. The goals were to (i) examine the PK/PD characteristics of telavancin and vancomycin using plasma and epithelial lining fluid (ELF) drug pharmacokinetics, (ii) define the target area under the concentration-time curve (AUC)/MIC exposures for both drugs associated with efficacy endpoints of net stasis and 1-log₁₀ kill for each infection model, (iii) compare the PK/PD efficacy targets between telavancin and vancomycin, and (iv) bridge the *in vivo* telavancin PK/PD efficacy analysis in this study to clinical practice by examining the PK/PD targets identified in the context of the human telavancin PK and MIC distribution.

RESULTS

***In vitro* susceptibility testing.** The MICs of telavancin and vancomycin for the four *S. aureus* strains used in the studies are shown in Table 1. The MIC range for both antibiotics was 4-fold, and elevated MIC of one drug was mirrored by a similar increase in the MIC of the comparator.

Plasma pharmacokinetics. The single-dose plasma pharmacokinetics of telavancin and select PK parameters are shown in Fig. 1A. At the doses studied, exposure to telavancin increased in a dose-proportional manner across the dose range studied (1.25 to 80 mg/kg of body weight). Maximum concentrations (C_{max}) ranged from 4.6 to 115 μ g/ml. AUC from time zero to infinity ($AUC_{0-\infty}$) values ranged from 10.5 to 595 μ g · h/ml. The AUC was linear across the dose range ($R^2 = 0.99$). The elimination half-life

FIG 1 Telavancin concentration-time curves in plasma (A) and ELF (B) in groups of three mice after single subcutaneous doses. Pharmacokinetic parameters, including the maximum concentration (C_{max}), area under the drug concentration-time curve from time zero to infinity ($AUC_{0-\infty}$), and elimination half-life ($t_{1/2}$), are shown for each dose at each site.

TABLE 2 ELF penetration of telavancin and vancomycin

Drug	Dose (mg/kg)	24-h total drug AUC (mg · h/liter)		Penetration into ELF (%)
		Plasma	ELF	
Telavancin	1.25	10.5	0.42	4.0
	5	42.3	1.3	3.1
	20	139	5.1	3.7
	80	595	19.2	3.2
Vancomycin	1.25	1.4	1.1	79
	5	6.2	4.5	73
	20	19.0	15.7	83
	80	68.1	40.1	59

($t_{1/2}$) ranged from 1.1 to 2.2 h. The ELF pharmacokinetic results for telavancin are similarly shown in Fig. 1B. $C_{\max}$ ranged from 0.2 to 7.8 $\mu\text{g}/\text{ml}$. $\text{AUC}_{0-\infty}$ values ranged from 0.4 to 19.2 $\mu\text{g} \cdot \text{h}/\text{ml}$. Despite increased variability in the concentration of drug in the first hour between different doses, the ELF AUC was linear across the dose range ($R^2 = 0.99$). The elimination half-life in ELF was similar to that in plasma and ranged from 2.0 to 2.9 h. The level of penetration into the ELF on the basis of the plasma AUC values is shown in Table 2 and was approximately 3 to 4%. This value is very similar to the amount of free drug that would be expected in plasma on the basis of 96% protein binding in mice, and thus, nearly 100% of free plasma drug penetrated into the ELF.

The single-dose plasma pharmacokinetics of vancomycin and select PK parameters are shown in Fig. 2A. Similar to the findings for telavancin, the drug concentration exposures increased in a dose-proportional manner across the dose range, and AUC was linear for both plasma and ELF concentration measurements ($R^2 = 0.99$). In plasma, the $C_{\max}$ ranged from 0.9 to 45.1 $\mu\text{g}/\text{ml}$, $\text{AUC}_{0-\infty}$ values ranged from 1.43 to 68.1 $\mu\text{g} \cdot \text{h}/\text{ml}$, and the elimination half-life ranged from 0.54 to 0.57 h. Vancomycin ELF drug concentrations for each dose are shown in Fig. 2B. $C_{\max}$ ranged from 0.6 to 21.5 $\mu\text{g}/\text{ml}$. $\text{AUC}_{0-\infty}$ values ranged from 1.1 to 40.1 $\mu\text{g} \cdot \text{h}/\text{ml}$. The elimination half-life ranged from 0.9 to 1.0 h. The penetration into the ELF on the basis of the plasma AUC is shown in Table 2 and was between 70 and 80% for all but the highest dose, where the value was almost 60%. On the basis of a plasma protein binding level of 25%, all but the highest vancomycin dose achieved nearly 100% of the free plasma levels in the ELF. Thus, for both drugs, after accounting for differences in protein binding, approximately 100% of free plasma drug penetrates into the ELF.

Pharmacodynamic target determination. The dose-response curves for telavancin against each strain in the thigh and lung infection models are shown in Fig. 3A and B, respectively. For each infection site, the dose-response curves were quite similar,

FIG 2 Vancomycin concentration-time curves in plasma (A) and ELF (B) in groups of three mice after single subcutaneous doses. Pharmacokinetic parameters, including the maximum concentration ($C_{\max}$), area under the drug concentration-time curve from time zero to infinity ($\text{AUC}_{0-\infty}$), and elimination half-life ($t_{1/2}$), are shown for each dose at each site.

FIG 3 Telavancin dose-response curves in the thigh (A) and lung (B) infection models. Groups of mice were infected with each *S. aureus* strain, and treatment with one of five different dosing regimens of telavancin by the subcutaneous route commenced starting 2 h after infection. After 24 h, mice were euthanized and the thigh or lung was aseptically harvested, homogenized, and serially plated to determine the bacterial burden as the number of CFU. Each point in the thigh model represents the mean and standard deviation from four thigh replicates, and each point in the lung model represents the mean and standard deviations from three lung replicates. The dashed horizontal line represents the burden at the start of therapy. Points above the line represent net growth, whereas those below the line represent killing (cidal) activity.

consistent with the relatively narrow MIC range. At the start of therapy, mice had $6.4 \pm 0.1 \log_{10}$ CFU/thigh and $6.3 \pm 0.1 \log_{10}$ CFU/lung. The burden increased in untreated controls to $9.6 \pm 0.2 \log_{10}$ CFU/thigh and $8.4 \pm 0.6 \log_{10}$ CFU/lung after 24 h. The growth of each strain in untreated animals is also shown in Table 3. In the thigh model, maximum efficacy was noted at between 1- and 2-log kill for 3 of 4 isolates, whereas in the lung we observed >2 -log kill for all 4 isolates.

The dose-response curves for vancomycin against each strain in the thigh and lung infection models are shown in Fig. 4A and B, respectively. As with telavancin, the dose-response curves were quite similar, given the narrow MIC range. At the start of therapy, mice had $6.3 \pm 0.1 \log_{10}$ CFU/thigh and $6.2 \pm 0.3 \log_{10}$ CFU/lung. The burden increased in untreated controls to $9.4 \pm 0.4 \log_{10}$ CFU/thigh and $8.3 \pm 0.6 \log_{10}$ CFU/lung after 24 h. The growth in untreated animals for each strain is also shown in Table 4. In the thigh model, maximum efficacy was noted at between 1- and 2-log kill for all 4 isolates, whereas in the lung we observed ≥ 2 -log kill for all 4 isolates.

One can see for both drugs that there was a shift to the left in the dose-response curves when comparing the thigh and lung models, with increased efficacy being

TABLE 3 *In vitro* and *in vivo* efficacies of telavancin against select *S. aureus* isolates using plasma AUC/MIC as the predictive pharmacodynamic index^a

Model	Organism and parameter	Control growth ($\log_{10}$ no. of CFU)	MIC (mg/liter)	24-h static dose (mg/kg)	24-h static dose tAUC/MIC	24-h static dose fAUC/MIC	24-h 1-log-kill dose (mg/kg)	24-h 1-log-kill dose tAUC/MIC	24-h 1-log-kill dose fAUC/MIC
Thigh	29213	3.1	0.0625	25.8	3,260	130	68.4	7,519	301
	33591	3.3	0.125	10.7	717	28.7	25.6	1,619	64.8
	LSI653	3.3	0.125	34.3	2,053	82.1	127	6,970	279
	LSI1856	2.8	0.25	83.5	2,268	90.7	NA	NA	NA
	Mean			38.6	2,075	83.0	73.6	5,369	215
	Median			30.1	2,160	86.4	68.4	6,970	279
	SD			31.5	1,047	41.9	50.8	3,259	130
Lung	29213	2.6	0.0625	13.6	1,817	72.7	26.9	3,372	135
	33591	2.3	0.125	20.9	1,386	55.4	39.4	2,311	92.4
	LSI653	2.1	0.125	9.9	664	26.6	22.8	1,478	59.1
	LSI1856	1.4	0.25	5.2	173	6.9	14.3	479	19.1
	Mean			12.4	1,010	40.4	25.9	1,910	76.4
	Median			11.8	1,025	41	24.9	1,895	75.8
	SD			6.6	733	29.3	10.5	1,229	49.2

^atAUC, total drug AUC; fAUC, free drug AUC; NA, not achieved.

FIG 4 Vancomycin dose-response curves in the thigh (A) and lung (B) infection models. Groups of mice were infected with each *S. aureus* strain, and treatment with one of seven different dosing regimens of telavancin by the subcutaneous route commenced starting 2 h after infection. After 24 h, mice were euthanized and the thigh or lung was aseptically harvested, homogenized, and serially plated to determine the bacterial burden as the number of CFU. Each point in the thigh model represents the mean and standard deviation from four thigh replicates, and each point in the lung model represents the mean and standard deviations from three lung replicates. The dashed horizontal line represents the burden at the start of therapy. Points above the line represent net growth, whereas those below the line represent killing (cidal) activity.

noted at each dose level. This in part may be explained by the difference in growth (i.e., fitness) between the two models that we observed in untreated animals. The growth in untreated animals in the lung model was significantly lower ($P < 0.001$) in experiments with both drugs.

The telavancin and vancomycin doses necessary to achieve net stasis or 1-log₁₀ kill were calculated for each strain using the maximum-effect (E_{max}) Hill equation and are shown in Tables 3 and 4, respectively. The mean 24-h static dose for telavancin was 38.6 mg/kg in the thigh model and 12.4 mg/kg in the lung model. The mean 24-h 1-log-kill doses for telavancin were approximately 2-fold higher. Once again, when comparing the two sites, the 1-log-kill doses were numerically higher in the thigh model at 73.6 mg/kg than the lung model at 25.9 mg/kg. The mean 24-h static dose for vancomycin against the four isolates was 159 mg/kg in the thigh model and 78.2 mg/kg in the lung model. The mean 1-log-kill doses for vancomycin were approximately 3-fold higher than the static doses and were 582 mg/kg in the thigh model and 204 mg/kg in the lung model.

TABLE 4 *In vitro* and *in vivo* efficacies of vancomycin against select *S. aureus* isolates using plasma AUC/MIC as the predictive pharmacodynamic index^a

Model	Organism and parameter	Control growth (log ₁₀ no. of CFU)	MIC (mg/liter)	24-h static dose (mg/kg)	24-h static dose tAUC/MIC	24-h static dose fAUC/MIC	24-h 1-log-kill dose (mg/kg)	24-h 1-log-kill dose tAUC/MIC	24-h 1-log-kill dose fAUC/MIC
Thigh	29213	3.4	1	186	163	122	606	515	386
	33591	3.2	1	180	158	118	792	673	505
	LSI653	2.8	2	156	69.2	51.9	566	240	180
	LSI1856	3.0	4	115	26.1	19.6	365	77.5	58.2
	Mean			159	104	77.9	582	376	282
	Median			168	113	85.0	586	378	283
	SD			32.2	67.3	50.5	175	268	201
Lung	29213	2.6	1	107	97.9	73.4	225	194	146
	33591	2.5	1	120	108	81.3	385	327	245
	LSI653	2.1	2	53.0	26.5	19.8	162	71.3	53.5
	LSI1856	2.3	4	33.1	9.02	6.76	46.6	11.9	8.90
	Mean			78.2	60.4	45.3	204	151	113
	Median			80.0	62.2	46.6	193	133	100
	SD			41.7	50.0	37.5	141	140	105

^atAUC, total drug AUC; fAUC, free drug AUC.

FIG 5 Relationship between telavancin plasma 24-h total drug AUC/MIC and treatment outcome in the thigh (A) and lung (B) infection models. Each point in the thigh model represents the mean from four thigh replicates, and each point in the lung model represents the mean from three lung replicates. The dashed horizontal line represents the burden at the start of therapy. Points above the line represent net growth, whereas those below the line represent killing (cidal) activity. The curved line represents the best-fit line based on the sigmoid $E_{\max}$ Hill equation. Also shown is the maximum effect ($E_{\max}$), the AUC/MIC associated with the 50% maximal effect (ED_{50}), the slope of the best-fit line (N), and the result of nonlinear least-squares multivariate regression analysis (R^2).

The dose-response data were then modeled utilizing both plasma and ELF PK data. The relationship between the telavancin plasma total drug 24-h AUC/MIC and the therapeutic effect in the thigh and lung models is shown in Fig. 5A and B. The data fit after applying the Hill $E_{\max}$ model to the plasma 24-h AUC/MIC and treatment effect data were fairly robust for each infection site ($R^2 = 0.85$ for the thigh and $R^2 = 0.91$ for the lung). Similar to the dose-response curves, the AUC/MIC exposures associated with stasis or cidal activity were shifted to the left (i.e., smaller) for the lung model in comparison to the thigh model. Additionally, the slope of the curve was steeper in the lung model, indicating that more cidal activity was observed with incremental increases in AUC/MIC exposure. ELF pharmacokinetic data for telavancin were also modeled, and the relationship between the ELF total drug 24-h AUC/MIC and therapeutic effect in the lung model is shown in Fig. 6A. As expected, the best-fit line based on the Hill equation from ELF analyses was almost identical to that for the plasma AUC/MIC. However, as only ~4% of plasma drug penetrated into the ELF, the ELF AUC/MIC was proportionally

FIG 6 Relationship between ELF 24-h AUC/MIC and treatment outcome in the lung model for telavancin (A) and vancomycin (B). Each point represents the mean from three lung replicates. The dashed horizontal line represents the burden at the start of therapy. Points above the line represent net growth, whereas those below the line represent killing (cidal) activity. The curved line represents the best-fit line based on the sigmoid $E_{\max}$ Hill equation. Also shown is the maximum effect ($E_{\max}$), the AUC/MIC associated with the 50% maximal effect (ED_{50}), the slope of the best-fit line (N), and the result of nonlinear least-squares multivariate regression analysis (R^2).

FIG 7 Relationship between vancomycin plasma 24-h total drug AUC/MIC and treatment outcome in the thigh (A) and lung (B) infection models. Each point in the thigh model represents the mean from four thigh replicates, and each point in the lung model represents the mean from three lung replicates. The dashed horizontal line represents the burden at the start of therapy. Points above the line represent net growth, whereas those below the line represent killing (cidal) activity. The curved line represents the best-fit line based on the sigmoid $E_{\max}$ Hill equation. Also shown is the maximum effect ($E_{\max}$), the AUC/MIC associated with the 50% maximal effect (ED_{50}), the slope of the best-fit line (N), and the result of nonlinear least-squares multivariate regression analysis (R^2).

lower than the plasma AUC/MIC targets. The same relationship between the vancomycin plasma 24-h AUC/MIC and treatment outcome was examined for both models and is shown in Fig. 7A and B. The relationship between AUC/MIC and therapeutic effect was equally strong, with R^2 values of 0.93 and 0.86 for the thigh and lung models, respectively. ELF pharmacokinetic data for vancomycin modeled to the 24-h ELF AUC/MIC and therapeutic effect in the lung are shown in Fig. 6B.

The plasma AUC/MIC values associated with the stasis and 1-log-kill endpoints were calculated for each strain in each infection site and are shown in Table 3 for telavancin and Table 4 for vancomycin. Both plasma total and free drug concentrations were considered. The telavancin mean 24-h plasma free drug AUC/MIC values for stasis were 83.0 and 40.4 for the thigh and lung models, respectively ($P = 0.22$). The telavancin mean 24-h plasma free drug AUC/MIC values associated with 1-log kill were 215 and 76.4 for the thigh and lung models, respectively ($P = 0.15$). In the case of vancomycin, the mean 24-h plasma free drug AUC/MIC values for stasis were 77.9 and 45.3 for the thigh and lung models, respectively ($P = 0.34$). The vancomycin mean 24-h plasma free drug AUC/MIC values associated with 1-log kill were 282 and 113 for the thigh and lung models, respectively ($P = 0.19$). The free plasma AUC/MIC targets were similar when telavancin and vancomycin were compared for each endpoint and infection site (P value range, 0.55 to 0.88).

The ELF 24-h AUC/MIC targets in the lung model are shown in Table 5 for each isolate for telavancin and vancomycin. The mean ELF 24-h AUC/MIC values associated with net stasis and 1-log kill for telavancin were 32.4 and 60.8, respectively. The mean ELF 24-h AUC/MIC values associated with net stasis and 1-log kill for vancomycin were 50.8 and 92.0, respectively. While the values were numerically lower for telavancin than vancomycin, the target AUC/MIC exposures were not statistically significantly different between the two drugs ($P = 0.89$ and $P = 0.51$, respectively).

DISCUSSION

Antimicrobial agents for the treatment of methicillin-resistant *S. aureus* (MRSA) pneumonia remain limited, and patient treatment outcome is suboptimal (10, 11). Currently, the two most common agents utilized for the treatment of MRSA pneumonia are vancomycin and linezolid. However, these agents have significant shortcomings, such as pharmacokinetic variability, toxicity-related adverse effects, and limited cidal activity. Novel potent therapies are necessary to address the paucity of therapeutic options for MRSA pneumonia. Telavancin is a potent and cidal anti-MRSA agent well equipped to

TABLE 5 *In vivo* efficacy in the murine lung model of telavancin and vancomycin against select *S. aureus* isolates using ELF AUC/MIC as the predictive pharmacodynamic index

Drug	Organism and parameter	MIC (mg/liter)	24-h static dose (mg/kg)	24-h static dose tAUC/MIC ^a	24-h 1-log-kill dose (mg/kg)	24-h 1-log-kill dose tAUC/MIC
Telavancin	29213	0.0625	13.6	58.0	26.9	106
	33591	0.125	20.9	42.4	39.4	75.9
	LSI653	0.125	9.9	22.4	22.8	45.7
	LSI1856	0.25	5.2	6.91	14.3	15.2
	Mean		12.4	32.4	25.9	60.8
	Median		11.8	32.4	24.9	60.8
	SD		6.6	22.4	10.5	39.3
Vancomycin	29213	1	107	76.6	225	120
	33591	1	120	98.7	385	191
	LSI653	2	53.0	20.9	162	47.3
	LSI1856	4	33.1	6.87	46.6	9.3
	Mean		78.2	50.8	204	92.0
	Median		80.0	48.7	193	83.7
	SD		41.7	43.9	141	80.6

^atAUC, total drug AUC.

fill this void. Previous studies in patients with hospital-acquired pneumonia due to Gram-positive pathogens (ATTAIN trials) demonstrated efficacy comparable to that of vancomycin (5). Preclinical studies have demonstrated the robust efficacy of telavancin in a range of animal models of soft tissue, cardiac, systemic, lung, bone, brain, and device-associated infections involving clinically relevant Gram-positive pathogens (8). However, pharmacodynamic evaluations to optimize dosing strategies for telavancin remain limited to a study with a single isolate performed by Hegde and colleagues (9).

We report the results of our studies examining the telavancin pharmacodynamic AUC/MIC target using well-characterized murine neutropenic thigh and lung infection models. Previous studies have demonstrated that lipoglycopeptides exhibit concentration-dependent activity with moderate to prolonged postantibiotic effects (12–15). The PD indices associated with this pattern of activity are $C_{\max}$ /MIC and AUC/MIC. In the case of telavancin, a previous dose fractionation study demonstrated AUC/MIC to be the PD index most closely associated with efficacy (9). Indeed, we demonstrated a robust relationship between the telavancin AUC/MIC and treatment efficacy in both models based on regression analysis utilizing the Hill $E_{\max}$ model ($R^2 = 0.85$ for the thigh and $R^2 = 0.91$ for the lung). The mean free drug AUC/MIC targets for stasis against a diverse group of *S. aureus* strains, including those with decreased susceptibility to beta-lactams and vancomycin, were 83 and 40.4 in the thigh and lung infection models, respectively. These values are similar to the targets identified for another lipoglycopeptide, dalbavancin, using the same infection model and methods of PD analyses (12, 13). The telavancin exposure-response relationships were relatively steep, such that 1-log₁₀-kill endpoints were, in general, achieved with an approximately 2-fold increase in AUC/MIC exposure. These targets are particularly robust, given that we utilized multiple strains with a range of *in vitro* susceptibilities to telavancin to generate the estimates. Additionally, we were able to compare the pharmacodynamic activity of telavancin to that of vancomycin, which in many settings remains the “gold standard” therapy for serious MRSA infection, including pneumonia. The pharmacodynamic activity and target exposures associated with stasis and cidal endpoints for telavancin in each infection site were very similar to those for vancomycin.

An important consideration in PK/PD studies is accounting for the protein binding of a drug, as it is generally accepted that only free drug is available for antimicrobial activity at the site of infection (16, 17). In the current studies, when plasma total drug AUC/MIC targets are compared between telavancin and vancomycin, it would appear that significant differences occur, as the plasma total drug AUC/MIC targets for telavancin are 10- to 20-fold higher than those for vancomycin. However, the plasma free drug AUC/MIC targets are congruent. Protein binding also appears to impact the

ELF pharmacokinetics of each drug. Again, it would appear that there are significant differences between the drugs, as almost 20-fold less telavancin than vancomycin penetrates into ELF (4% versus 70 to 80%, respectively). However, when one examines the penetration into ELF in comparison to the free fraction of drug available, both drugs perform similarly. Approximately 4% of the telavancin plasma drug concentration penetrates into the ELF, which is equivalent to the proportion of free drug in plasma based on protein binding of 96%. The same held true for vancomycin, where we observed 70 to 80% penetration into ELF, which approximates the proportion of free drug that would be expected in the plasma on the basis of protein binding of 25%. Thus, the penetration into ELF for both drugs is approximately 100% of the free plasma fraction. On the basis of this, it is not surprising that we found PK/PD targets using the total drug ELF AUC/MIC to closely approximate the plasma free drug AUC/MIC targets for each drug in the lung infection model.

Whether stasis endpoints or cidal endpoints are more important in translating preclinical PK/PD results to clinical medicine remains an ongoing debate. In general, for pneumonia many experts place more emphasis on preclinical cidal endpoints, such as 1-log kill. Human pharmacodynamic studies with vancomycin have been performed in a number of settings evaluating the significance of total drug AUC/MIC targets in patient outcome. The first study examining this was published in 2004 by Moise-Broder and colleagues (18). They examined 108 patients receiving vancomycin for *S. aureus* pneumonia and found that 24-h total drug AUC/MIC ratios of >350 were associated with a statistically significantly higher odds of clinical success and that ratios of >400 were associated with faster bacterial eradication. Another study published in 2012 demonstrated that patients with vancomycin 24-h total drug AUC/MIC exposures in excess of 211 had significantly lower attributable mortality from complicated bacteremia and endocarditis due to MRSA (19). Finally, in 2013 Holmes and colleagues demonstrated that a vancomycin 24-h total drug AUC/MIC threshold of >373 was associated with reduced mortality in patients with *S. aureus* bacteremia (20). Overall, the limited pharmacodynamic data from clinical studies have led many experts to recommend vancomycin total drug 24-h AUC/MIC targets of >400 as a goal to optimize outcomes (21). Our *in vivo* animal model data demonstrated that these targets are very congruent with the 1-log-kill targets identified in the murine thigh model. The vancomycin 24-h total drug AUC/MIC value associated with this outcome was 376. Additionally, we observed plateauing of maximal kill effects at AUC/MIC values just in excess of 400. Thus, the murine thigh model 1-log-kill targets are likely to be clinically relevant for translating preclinical PK/PD targets to clinical medicine.

An interesting observation in our study was a shift to the left for the exposure-response curves in the lung infection model compared to the thigh infection model, indicating that less drug was needed to achieve similar outcomes. This resulted in static doses, 1-log-kill doses, and AUC/MIC targets associated with these endpoints which are approximately 2-fold larger for the thigh model than the lung model; however, these differences were not statistically significant. There are important considerations associated with the differences in the exposure-response relationships observed between different infection models. Two very relevant factors are the initial burden of organism in each model and the ability of the organism to grow and produce infection over time in untreated animals (i.e., fitness). While the infection burdens at the start of therapy were remarkably congruent, we did note a consistently lower growth in the untreated control mice in the murine lung model than the thigh model which was statistically significant ($P < 0.001$). Thus, we feel that the differences in the ability of the organism to produce infection at each of these sites were likely a factor in our observation that PD targets were numerically lower in the lung.

Human PK studies have been performed with telavancin (6, 22). The 24-h AUC after a single 10-mg/kg intravenous dose of telavancin in humans is approximately 666 mg · h/liter and increases slightly to 780 mg · h/liter with multiple daily doses. Protein binding in humans is approximately 90% (6, 23). Thus, free drug 24-h AUCs would be approximately 67 and 78 mg · h/liter, respectively. Using the most conservative

estimates in the PD targets identified in this study (i.e., the thigh targets), a stasis target would be expected to be achievable for isolates with telavancin MICs of ≤ 0.5 mg/liter and 1-log kill would be expected to be achievable for isolates with telavancin MICs of ≤ 0.25 mg/liter. These MIC thresholds are consistent with the current MIC breakpoints proposed by EUCAST, CLSI, and FDA, which have set the susceptible breakpoint at ≤ 0.12 mg/liter (6, 24–26). Additionally, $>99\%$ of all *S. aureus* isolates, including MRSA isolates, exhibit an MIC distribution of ≤ 0.12 mg/liter against telavancin (27–29). Thus, the PD targets associated with efficacy identified in this study and human PK data suggest that telavancin would be predicted to be effective against nearly all *S. aureus* isolates.

In conclusion, these studies demonstrate that telavancin exhibits dose-dependent *in vivo* activity against various strains of *S. aureus* in the neutropenic murine thigh and lung infection models. The PK/PD index AUC/MIC was a very strong predictor of treatment efficacy in both models. The PK/PD characteristics of telavancin and the efficacy AUC/MIC targets compare very favorably with those of vancomycin. Both static and killing endpoints were achieved in both models at relatively modest AUC/MIC targets. The preclinical vancomycin 1-log-kill targets align very well with clinical targets (AUC/MIC > 400), suggesting that this endpoint may be the most clinically relevant. Importantly, for telavancin the 1-log-kill targets identified in these studies, epidemiological susceptibility surveys, and human PK data suggest that the current dosing regimen in humans would be predicted to provide telavancin exposures that are sufficient to achieve optimal efficacy. Additionally, the proposed breakpoint of ≤ 0.12 mg/liter appears to be appropriate on the basis of the results of these studies. In total, these findings support telavancin as an efficacious option with PK/PD exposures optimized for serious *S. aureus* infections, including MRSA pneumonia.

MATERIALS AND METHODS

Organisms, media, and antibiotic. Four *Staphylococcus aureus* strains were used for these studies, including 1 methicillin-susceptible *S. aureus* (MSSA) and 3 methicillin-resistant *S. aureus* (MRSA) strains (Table 1). One MRSA strain also demonstrated a vancomycin-intermediate-susceptibility phenotype (vancomycin-intermediate *S. aureus* [VISA]). Organisms were grown, subcultured, and quantified using Mueller-Hinton broth (MHB) and agar (Difco Laboratories, Detroit, MI). Telavancin was supplied by Theravance Biopharma Antibiotics, Inc. (George Town, Cayman Islands), and vancomycin was obtained from the University of Wisconsin (Madison, WI) pharmacy.

***In vitro* susceptibility testing.** *In vitro* MIC testing was performed according to CLSI standard techniques (24). Specifically, for telavancin the MICs were determined in cation-adjusted MHB supplemented with 0.002% polysorbate 80 (30). All MICs were performed in duplicate on at least three occasions. The median MIC is reported and was used for analysis (Table 1).

Murine thigh and lung infection models. Animals were maintained in accordance with the Association for Assessment and Accreditation of Laboratory Animal Care (AAALAC) criteria (31). All animal studies were approved by the Animal Research Committees of the William S. Middleton Memorial VA Hospital and the University of Wisconsin. Six-week-old, specific-pathogen-free, female ICR/Swiss mice weighing 24 to 27 g were used for all studies (Harlan Sprague-Dawley, Indianapolis, IN). Mice were rendered neutropenic (neutrophil count, $<100/\text{mm}^3$) by injecting cyclophosphamide (Mead Johnson Pharmaceuticals, Evansville, IN) intraperitoneally at 4 days (150 mg/kg) and 1 day (100 mg/kg) before infection. Broth cultures of freshly plated bacteria were grown overnight to logarithmic phase to an absorbance at 580 nm of 0.3 (Spectronic 88; Bausch and Lomb, Rochester, NY). After 1:10 dilution into fresh MHB, the bacterial counts of the inoculum were $7.0 \pm 0.1 \log_{10}$ CFU/ml and $6.7 \pm 0.1 \log_{10}$ CFU/ml for telavancin and vancomycin, respectively, in the thigh infection model. Infection with each of the isolates was produced by injection of 0.1 ml of inoculum into the thigh of isoflurane-anesthetized mice (32). Four thigh infections were included in each treatment and control group. In the lung infection model, the bacterial counts of the inoculum were $7.7 \pm 0.1 \log_{10}$ CFU/ml and $7.8 \pm 0.1 \log_{10}$ CFU/ml for telavancin and vancomycin, respectively. Lung infection with each isolate was produced by pipetting 50 μl of inoculum into the anterior nares of isoflurane-anesthetized mice held upright to allow for inhalation into the lungs (32). Three mice were included in each treatment and control group. Drug treatment was commenced 2 h after infection was induced. The treatment duration was 24 h, at which time mice were euthanized by CO_2 asphyxiation and the thighs or lungs were aseptically harvested for determination of the number of CFU by serial plating on MHB.

Drug pharmacokinetics. The single-dose plasma pharmacokinetics of telavancin and vancomycin were studied in mice. Animals were administered a single subcutaneous dose (0.2 ml/dose) at dose levels of 1.25, 5, 20, and 80 mg/kg. Groups of three mice were sampled at each time point and dose level. There were 7 sampling times that ranged from 0.5 to 12 h. Samples were collected from each mouse by cardiac puncture. Plasma was obtained by centrifugation for 5 min at 4,000 rpm and immediately frozen at -70°C until PK assay. Plasma concentrations were determined by Q² Solutions (Ithaca, NY) using a

validated liquid chromatography-tandem mass spectrometry (LC-MS/MS) method (9, 33). Pharmacokinetic parameters ($\pm$ standard deviation), including elimination half-life ($t_{1/2}$), $AUC_{0-\infty}$ and C_{max} , were calculated by noncompartmental analysis. The half-life was determined by linear least-squares regression. The $AUC_{0-\infty}$ was calculated from the mean concentrations using the trapezoidal rule. Due to the linearity of the results over the dose range, pharmacokinetic estimates for dose levels that were not measured were calculated using linear interpolation for dose levels between those with measured kinetics (e.g., between 5 and 20 mg/kg) and linear extrapolation for dose levels above or below the highest and lowest dose levels with kinetic measurements (i.e., 1.25 and 80 mg/kg). Protein binding of 96% was used to determine the free drug fraction for telavancin, and protein binding of 25% was used to determine the free drug fraction for vancomycin (9, 34).

Single-dose ELF pharmacokinetics were also determined for the same doses and time points listed above for each drug. Bronchial alveolar lavage (BAL) fluid was obtained by instillation of 1 ml of sterile saline into the lungs of each animal followed by immediate removal. The BAL fluid was centrifuged to remove blood and cellular debris. The supernatant was then collected, immediately frozen, and stored at -70°C until drug assay by LC-MC/MS. ELF concentrations were calculated from BAL fluid concentrations by the urea correction methodology using the formula $[\text{drug}]_{\text{ELF}} = [\text{drug}]_{\text{BAL fluid}} \times ([\text{urea}]_{\text{plasma}} / [\text{urea}]_{\text{BAL fluid}})$, where $[\text{drug}]_{\text{ELF}}$ is the drug concentration in ELF, $[\text{drug}]_{\text{BAL fluid}}$ is the drug concentration in BAL fluid, $[\text{urea}]_{\text{plasma}}$ is the urea concentration in plasma, and $[\text{urea}]_{\text{BAL fluid}}$ is the urea concentration in BAL fluid. Total drug concentrations from ELF were utilized for all analyses. The penetration of each drug into the ELF space was calculated by comparing ELF/plasma AUC ratios.

Pharmacodynamic target determination. Neutropenic mice were infected with one of four *S. aureus* strains as described above. Both the thigh and lung infection models were utilized for all strains. Treatment with telavancin or vancomycin was initiated at 2 h after infection. Five total doses of telavancin, which ranged from 1.25 to 320 mg/kg/24 h, were fractionated into 6-h dosing regimens. Similarly, seven total doses of vancomycin, which ranged from 1.25 to 5,120 mg/kg/24 h, were fractionated into 6-h dosing regimens. The drug doses were administered subcutaneously. Mice were euthanized after 24 h, and the infected thighs or lungs were aseptically removed and processed for determination of the number of CFU. Dose-response curves for telavancin and vancomycin were generated from the treatment data for both infection models.

Pharmacodynamic analyses utilized data from the PK study, *in vitro* susceptibility results, and the treatment outcome data. The PK/PD 24-h AUC/MIC ratio was used for all analyses, as this has been the pharmacodynamically linked index in previous glycopeptide, lipopeptide, and lipoglycopeptide studies (8, 9, 12, 15, 35–37). The correlation between treatment efficacy and AUC/MIC was determined by nonlinear least-squares multivariate regression (SigmaPlot software, version 12.3; Systat Software, San Jose, CA). The model is derived from the Hill equation: $E = (E_{\text{max}} \times D^N) / (ED_{50}^N + D^N)$, where E is the effect or, in this case, the log change in the number of CFU per thigh between treated mice and untreated controls after the 24-h period of study, E_{max} is the maximum effect, D is the 24-h total dose, ED_{50} is the dose required to achieve 50% of E_{max} , and N is the slope of the dose-effect curve. The indices E_{max} , ED_{50} , and N were calculated using nonlinear least-squares regression. A sigmoid dose-response E_{max} model derived from the four-parameter Hill equation was used to calculate the dose of telavancin or vancomycin that produced net bacteriostatic and 1-log₁₀-kill endpoints over 24 h (static and 1-log-kill doses). The pharmacodynamic target AUC/MIC was determined for each endpoint and for each isolate using total plasma, free plasma, and total ELF drug concentrations. The PD targets identified in the thigh and lung models were compared by *t* test or the Mann-Whitney rank-sum test to determine if significant differences existed between infection sites or between each of the drugs at the same site.

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REFERENCES

- Karlowsky JA, Nichol K, Zhanel GG. 2015. Telavancin: mechanisms of action, *in vitro* activity, and mechanisms of resistance. *Clin Infect Dis* 61(Suppl 2):S58–S68. <https://doi.org/10.1093/cid/civ534>.
- Higgins DL, Chang R, Debatov DV, Leung J, Wu T, Krause KM, Sandvik E, Hubbard JM, Kaniga K, Schmidt DE, Jr, Gao Q, Cass RT, Karr DE, Benton BM, Humphrey PP. 2005. Telavancin, a multifunctional lipoglycopeptide, disrupts both cell wall synthesis and cell membrane integrity in methicillin-resistant *Staphylococcus aureus*. *Antimicrob Agents Chemother* 49:1127–1134. <https://doi.org/10.1128/AAC.49.3.1127-1134.2005>.
- Lunde CS, Hartouni SR, Janc JW, Mammen M, Humphrey PP, Benton BM. 2009. Telavancin disrupts the functional integrity of the bacterial membrane through targeted interaction with the cell wall precursor lipid II. *Antimicrob Agents Chemother* 53:3375–3383. <https://doi.org/10.1128/AAC.01710-08>.
- Stryjowski ME, Graham DR, Wilson SE, O'Riordan W, Young D, Lentnek A, Ross DP, Fowler VG, Hopkins A, Friedland HD, Barriere SL, Kitt MM, Corey GR. Assessment of Telavancin in Complicated Skin and Skin-Structure Infections Study. 2008. Telavancin versus vancomycin for the treatment of complicated skin and skin-structure infections caused by gram-positive organisms. *Clin Infect Dis* 46:1683–1693. <https://doi.org/10.1086/587896>.
- Rubinstein E, Lalani T, Corey GR, Kanafani ZA, Nannini EC, Rocha MG, Rahav G, Niederman MS, Kollef MH, Shorr AF, Lee PC, Lentnek AL, Luna CM, Fagon JY, Torres A, Kitt MM, Genter FC, Barriere SL, Friedland HD, Stryjowski ME, ATTAIN Study Group. 2011. Telavancin versus vancomycin for hospital-acquired pneumonia due to gram-positive pathogens. *Clin Infect Dis* 52:31–40. <https://doi.org/10.1093/cid/ciq031>.
- Food and Drug Administration. 2017. Vibativ (telavancin) prescribing information. Food and Drug Administration, Silver Spring, MD. http://www.accessdata.fda.gov/drugsatfda_docs/label/2014/022110s011bl.pdf. Accessed 29 February 2017.
- Craig WA. 1998. Pharmacokinetic/pharmacodynamic parameters: rationale for antibacterial dosing of mice and men. *Clin Infect Dis* 26:1–10. <https://doi.org/10.1086/516284>.
- Hegde SS, Janc JW. 2014. Efficacy of telavancin, a lipoglycopeptide antibiotic, in experimental models of Gram-positive infection. *Expert Rev*

- Anti Infect Ther 12:1463–1475. <https://doi.org/10.1586/14787210.2014.979789>.
9. Hegde SS, Reyes N, Wiens T, Vanasse N, Skinner R, McCullough J, Kaniga K, Pace J, Thomas R, Shaw JP, Obedencio G, Judice JK. 2004. Pharmacodynamics of telavancin (TD-6424), a novel bactericidal agent, against Gram-positive bacteria. *Antimicrob Agents Chemother* 48:3043–3050. <https://doi.org/10.1128/AAC.48.8.3043-3050.2004>.
 10. Moellering RC, Jr. 2012. MRSA: the first half century. *J Antimicrob Chemother* 67:4–11. <https://doi.org/10.1093/jac/dkr437>.
 11. Liu C, Bayer A, Cosgrove SE, Daum RS, Fridkin SK, Gorwitz RJ, Kaplan SL, Karchmer AW, Levine DP, Murray BE, Rybak MJ, Talan DA, Chambers HF, Infectious Diseases Society of America. 2011. Clinical practice guidelines by the Infectious Diseases Society of America for the treatment of methicillin-resistant *Staphylococcus aureus* infections in adults and children. *Clin Infect Dis* 52:e18–e55. <https://doi.org/10.1093/cid/ciq146>.
 12. Andes D, Craig WA. 2007. In vivo pharmacodynamic activity of the glycopeptide dalbavancin. *Antimicrob Agents Chemother* 51:1633–1642. <https://doi.org/10.1128/AAC.01264-06>.
 13. Lepak A, Marchillo K, VanHecker J, Andes D. 2015. Impact of glycopeptide resistance in *Staphylococcus aureus* on the dalbavancin in vivo pharmacodynamic target. *Antimicrob Agents Chemother* 59:7833–7836. <https://doi.org/10.1128/AAC.01717-15>.
 14. Boylan CJ, Campanale K, Iversen PW, Phillips DL, Zeckel ML, Parr TR, Jr. 2003. Pharmacodynamics of oritavancin (LY333328) in a neutropenic-mouse thigh model of *Staphylococcus aureus* infection. *Antimicrob Agents Chemother* 47:1700–1706. <https://doi.org/10.1128/AAC.47.5.1700-1706.2003>.
 15. Ambrose PG, Drusano GL, Craig WA. 2012. In vivo activity of oritavancin in animal infection models and rationale for a new dosing regimen in humans. *Clin Infect Dis* 54(Suppl 3):S220–S228. <https://doi.org/10.1093/cid/cis001>.
 16. Craig WA, Ebert SC. 1989. Protein binding and its significance in antibacterial therapy. *Infect Dis Clin North Am* 3:407–414.
 17. Zeitlinger MA, Derendorf H, Mouton JW, Cars O, Craig WA, Andes D, Theuretzbacher U. 2011. Protein binding: do we ever learn? *Antimicrob Agents Chemother* 55:3067–3074. <https://doi.org/10.1128/AAC.01433-10>.
 18. Moise-Broder PA, Forrest A, Birmingham MC, Schentag JJ. 2004. Pharmacodynamics of vancomycin and other antimicrobials in patients with *Staphylococcus aureus* lower respiratory tract infections. *Clin Pharmacokinet* 43:925–942. <https://doi.org/10.2165/00003088-200443130-00005>.
 19. Brown J, Brown K, Forrest A. 2012. Vancomycin AUC₂₄/MIC ratio in patients with complicated bacteremia and infective endocarditis due to methicillin-resistant *Staphylococcus aureus* and its association with attributable mortality during hospitalization. *Antimicrob Agents Chemother* 56:634–638. <https://doi.org/10.1128/AAC.05609-11>.
 20. Holmes NE, Turnidge JD, Munckhof WJ, Robinson JO, Korman TM, O'Sullivan MV, Anderson TL, Roberts SA, Warren SJ, Gao W, Howden BP, Johnson PD. 2013. Vancomycin AUC/MIC ratio and 30-day mortality in patients with *Staphylococcus aureus* bacteremia. *Antimicrob Agents Chemother* 57:1654–1663. <https://doi.org/10.1128/AAC.01485-12>.
 21. Rybak MJ, Lomaestro BM, Rotschafer JC, Moellering RC, Jr, Craig WA, Billeter M, Dalovisio JR, Levine DP. 2009. 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* 29:1275–1279. <https://doi.org/10.1592/phco.29.11.1275>.
 22. Shaw JP, Seroogy J, Kaniga K, Higgins DL, Kitt M, Barriere S. 2005. Pharmacokinetics, serum inhibitory and bactericidal activity, and safety of telavancin in healthy subjects. *Antimicrob Agents Chemother* 49:195–201. <https://doi.org/10.1128/AAC.49.1.195-201.2005>.
 23. Lodise TP, Jr, Gotfried M, Barriere S, Drusano GL. 2008. Telavancin penetration into human epithelial lining fluid determined by population pharmacokinetic modeling and Monte Carlo simulation. *Antimicrob Agents Chemother* 52:2300–2304. <https://doi.org/10.1128/AAC.01110-07>.
 24. Clinical and Laboratory Standards Institute. 2016. Performance standards for antimicrobial susceptibility testing, 26th ed. CLSI supplement M100S. Clinical and Laboratory Standards Institute, Wayne, PA.
 25. Farrell DJ, Mendes RE, Rhomberg PR, Jones RN. 2014. Revised reference broth microdilution method for testing telavancin: effect on MIC results and correlation with other testing methodologies. *Antimicrob Agents Chemother* 58:5547–5551. <https://doi.org/10.1128/AAC.03172-14>.
 26. EUCAST. Breakpoint tables for interpretation of MICs and zone diameters, version 6.0. http://www.eucast.org/fileadmin/src/media/PDFs/EUCAST_files/Breakpoint_tables/v_6_0_Breakpoint_table.pdf. Accessed 29 February 2017.
 27. Mendes RE, Farrell DJ, Sader HS, Streit JM, Jones RN. 2015. Update of the telavancin activity in vitro tested against a worldwide collection of Gram-positive clinical isolates (2013), when applying the revised susceptibility testing method. *Diagn Microbiol Infect Dis* 81:275–279. <https://doi.org/10.1016/j.diagmicrobio.2014.12.011>.
 28. Mendes RE, Flamm RK, Farrell DJ, Sader HS, Jones RN. 2016. Telavancin activity tested against Gram-positive clinical isolates from European, Russian and Israeli hospitals (2011 to 2013) using a revised broth microdilution testing method: redefining the baseline activity of telavancin. *J Chemother* 28:83–88. <https://doi.org/10.1179/1973947815Y.0000000050>.
 29. Mendes RE, Sader HS, Flamm RK, Farrell DJ, Jones RN. 2015. Telavancin in vitro activity against a collection of methicillin-resistant *Staphylococcus aureus* isolates, including resistant subsets, from the United States. *Antimicrob Agents Chemother* 59:1811–1814. <https://doi.org/10.1128/AAC.04616-14>.
 30. Clinical and Laboratory Standards Institute. 2014. Performance standards for antimicrobial susceptibility testing, 24th informational supplement. M100-S24. Clinical and Laboratory Standards Institute, Wayne, PA.
 31. National Research Council Committee on the Care and Use of Laboratory Animals, Institute of Laboratory Animal Resources, and Commission on Life Sciences. 1996. Guide for the care and use of laboratory animals. National Academy Press, Washington, DC.
 32. Andes DR, Lepak AJ, Fridmott-Moller N. 2015. Evaluation of antimicrobials in experimental animal infections, p 603–677. In Amsterdam D (ed), *Antibiotics in laboratory medicine*, 6th ed. Wolters Kluwer, Philadelphia, PA.
 33. Reyes N, Skinner R, Benton BM, Krause KM, Shelton J, Obedencio GP, Hegde SS. 2006. Efficacy of telavancin in a murine model of bacteraemia induced by methicillin-resistant *Staphylococcus aureus*. *J Antimicrob Chemother* 58:462–465. <https://doi.org/10.1093/jac/dkl222>.
 34. Knudsen JD, Fuursted E, Espersen F, Fridmott-Moller N. 1997. Activities of vancomycin and teicoplanin against penicillin-resistant pneumococci in vitro and in vivo and correlation to pharmacokinetic parameters in the mouse peritonitis model. *Antimicrob Agents Chemother* 41:1910–1915.
 35. Rybak MJ. 2006. The pharmacokinetic and pharmacodynamic properties of vancomycin. *Clin Infect Dis* 42(Suppl 1):S35–S39. <https://doi.org/10.1086/491712>.
 36. Safdar N, Andes D, Craig WA. 2004. In vivo pharmacodynamic activity of daptomycin. *Antimicrob Agents Chemother* 48:63–68. <https://doi.org/10.1128/AAC.48.1.63-68.2004>.
 37. Craig WA. 2003. Basic pharmacodynamics of antibacterials with clinical applications to the use of beta-lactams, glycopeptides, and linezolid. *Infect Dis Clin North Am* 17:479–501. [https://doi.org/10.1016/S0891-5520\(03\)00065-5](https://doi.org/10.1016/S0891-5520(03)00065-5).