Efficacy of telavancin in a murine model of bacteraemia induced by methicillin-resistant Staphylococcus aureus

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Objectives: The efficacy of telavancin, a bactericidal lipoglycopeptide, was compared with vancomycin against methicillin-resistant Staphylococcus aureus (MRSA) in an immunocompromised murine model of bacteraemia.

Methods: Immunocompromised mice were inoculated intraperitoneally with S. aureus ATCC 33591 and treated with two subcutaneous doses (once every 12 h) of vehicle or test compound. Mouse pharmacokinetic data were generated and used to choose doses of telavancin (40 mg/kg) and vancomycin (110 mg/kg) in order to equate clinical exposures. Reduction in bacterial titre (in blood and spleen) and mortality were the two pharmacodynamic endpoints of the study.

Results: Mortality was 100% in animals treated with vehicle or vancomycin but was significantly lower (7%) in telavancin-treated animals. Telavancin produced significantly greater reductions in blood and spleen bacterial titres compared with vancomycin.

Conclusions: The data described here demonstrate that telavancin’s in vivo bactericidal activity is superior to that of vancomycin against a single strain of MRSA and results in successful infection resolution and, consequently, improved survival in the murine bacteraemia model.

Keywords: lipoglycopeptides, MRSA, neutropenic, mice

Introduction

Methicillin-resistant Staphylococcus aureus (MRSA) has become a frequent cause of serious nosocomial infections including bacteraemia. Telavancin is a novel lipoglycopeptide that operates through multiple mechanisms to produce potent and rapid bactericidal activity against clinically relevant Gram-positive bacteria including MRSA. Previous studies have shown that telavancin exhibits potent antibacterial activity against MRSA in animal models of soft-tissue infection, endocarditis and pneumonia. In the studies described here, we assessed the efficacy of telavancin in an immunocompromised murine model of bacteraemia caused by MRSA.

Materials and methods

Telavancin for injection (250 mg/vial) and vancomycin (Sigma-Aldrich, St Louis, MO, USA) were reconstituted in 5% dextrose in water. MRSA strain ATCC 33591, used for all studies, was obtained from the American Type Culture Collection (Manassas, VA, USA). MICs were determined by the broth microdilution method according to protocol M7-A5 of the Clinical Laboratory Standards Institute (formerly NCCLS). All in vivo studies were approved by the Institutional Animal Care and Use Committee at Theravance, Inc. The experimental model of infection that was employed in the present study was similar to that described previously with some modifications. Lethal bacteraemia was produced by intraperitoneal inoculation of the organism in immunocompromised mice. Immunocompromised mice were used to produce a robust infection that had a predictable time course. Besides measuring blood titres, the bacterial burden in the spleen was also evaluated to assess the level of infection in a systemic organ. Female NSA mice (Harlan, Indianapolis, IN, USA), weighing between 18 and 28 g, were rendered neutropenic by treatment with 250 mg/kg of cyclophosphamide, intraperitoneally, at 4 and 2 days prior to infection. Neutropenic animals were inoculated intraperitoneally with ~0.8 mL of inoculum containing ~10⁵ cfu/mL of bacteria. The inoculum size of 10⁵ was chosen to produce a robust and consistent infection.

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Telavancin's efficacy in MRSA bacteraemia

Table 1. Pharmacokinetic parameters of telavancin and vancomycin in humans and infected mice

<table>
<thead>
<tr>
<th></th>
<th>Telavancin</th>
<th>Vancomycin</th>
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</thead>
<tbody>
<tr>
<td>Dose</td>
<td>mouse</td>
<td>humana</td>
</tr>
<tr>
<td></td>
<td>40 mg/kg sc two dosesb</td>
<td>10 mg/kg iv once daily</td>
</tr>
<tr>
<td>Cmax (µg/L)</td>
<td>98.9 (5.6)</td>
<td>87.5 (4.9)</td>
</tr>
<tr>
<td>AUC0-24 (µg·h/L)</td>
<td>1107 (63)</td>
<td>858 (50)</td>
</tr>
<tr>
<td>t1/2 (h)</td>
<td>2.7</td>
<td>7.5</td>
</tr>
</tbody>
</table>

aData reported as total drug with free drug in parentheses. Free drug concentrations were computed based on 94% and 42% protein binding for telavancin and vancomycin, respectively.

bData projected from single dose data.

cData from ref. (9).

dData from ref. (10).

In pharmacokinetic studies, immunosuppressed infected mice were dosed 4 h after inoculation with a single subcutaneous (sc) injection of either telavancin (40 mg/kg, sc) or vancomycin (110 mg/kg, sc) (n = 3 per group). Blood samples were collected using cardiac puncture at pre-dose, 2, 5, 15 and 30 min, and 1, 2, 4, 8 and 24 h post-dose. Blood samples were processed to plasma by centrifugation (12,000 rpm, 4 min, 4°C) and stored at −80°C until analysis. The plasma samples were subjected to solid phase extraction and analysed by liquid chromatography with tandem mass spectrometry (LC/MS/MS). Deuterated telavancin was used as the internal standard. The limit of quantification was 0.25 mg/L and the coefficient of variance for replicate samples at each level was <20%. The pharmacokinetic parameters of single doses of telavancin and vancomycin were analysed by non-compartmental methods using WinNonlin (Version 4.0.1, Pharsight, Mountain View, CA, USA).

In studies intended to investigate effects of treatment on survival, infected immunosuppressed animals were administered two doses of telavancin (40 mg/kg, sc) or vancomycin (110 mg/kg, sc) at 4 and 16 h post-infection (n = 15 per group). A separate group of control animals (n = 15) were left untreated. Animals had access to food and water ad libitum. Animals were observed twice daily and deaths recorded over a 14 day period.

For bacterial titre studies, infected immunosuppressed animals were administered two doses of telavancin (40 mg/kg, sc) or vancomycin (110 mg/kg, sc) at 4 and 16 h post-infection (n = 15 per group). A separate group of control animals (n = 10) were left untreated. At 1, 4, 10, 16, 22, 28, 34, 40 and 120 h post-inoculation, designated groups of control and drug-treated surviving animals were euthanized via CO2 asphyxiation. Subsequently, blood and spleen were collected via cardiac puncture, serially diluted and plated onto tryptic-soy agar (TSA) plates. Each spleen was excised, weighed, placed in saline, homogenized, serially diluted and plated onto TSA plates. All plates were incubated overnight at 37°C and the bacterial colonies were counted the following day.

Results and discussion

The MICs of telavancin and vancomycin for S. aureus ATCC 33591 were 0.5 and 1 mg/L, respectively.

Following sc administration into neutropenic infected mice, telavancin (40 mg/kg) and vancomycin (110 mg/kg) had a Cmax of 102.3 and 108.1 mg/L, mean AUC0-24 of 574.5 and 152.7 mg·h/L, clearance of 0.070 and 0.721 L/h/kg and half-life of 2.7 and 0.97 h, respectively. The projected exposures for two doses (every 12 h) of telavancin (40 mg/kg, sc) and vancomycin (110 mg/kg, sc) in comparison with human exposures of the two antibiotics are shown in Table 1. The free-fraction of drug was computed based on protein binding estimates of 94% and 42% for telavancin and vancomycin, respectively. The total and free Cmax and AUC of telavancin and vancomycin were not significantly different between the mouse and human doses. The free mouse and human AUC/MICs were 126 and 100 for telavancin and 130 and 132 for vancomycin, respectively. The t > MIC was >24 h in mice and humans for telavancin and 11 and >24 h in mice and humans, respectively, for vancomycin.

The pre-treatment bacterial titres were 4.3 log cfu/mL and 8.7 log cfu/g in the blood and spleen, respectively. In untreated animals, the titre in the blood and spleen increased to 7.5 log cfu/mL and 9.7 log cfu/g, respectively. Analysis of the kill curves for both blood and spleen revealed that telavancin exhibited significantly greater killing activity than vancomycin (P < 0.05, two-way ANOVA) (Figure 1). At 6 h after the first dose, the titres in the blood were reduced to a greater extent by telavancin (~2.5 log cfu/mL) compared with vancomycin (~1.2 log cfu/mL). At 6 h after the second dose, the splenic titres were reduced to a greater extent by telavancin (~3.8 log cfu/g) when compared with vancomycin (~1.4 log cfu/g). Animals treated with vancomycin had a rebound increase in both their splenic and blood titres between 28 and 40 h post-infection. In contrast, telavancin produced sustained suppression of titres and eventually resolved the infection in the blood and spleen by 28 and 120 h, respectively.

The proportion of survivors after 14 days in the control, vancomycin and telavancin groups were 0/15 (0%), 0/15 (0%) and 14/15 (93%), respectively (Figure 2). Comparison of the survival curves of the telavancin-treated and vancomycin-treated groups indicated that telavancin produced significantly greater improvement in survival (P < 0.05, log-rank test).

We propose that the superior efficacy demonstrated by telavancin in this model of bacteraemia can be attributed, at least in part, to its enhanced bactericidal potency and longer post-antibiotic effect (PAE), compared with vancomycin. We have reported previously on the differential bactericidal properties of telavancin and vancomycin against S. aureus ATCC 33591; at equal multiples (8×) of their MIC telavancin produced >3 log10 decrease in bacterial count as opposed to ~1 log10 decrease with vancomycin.
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Figure 1. Effects of telavancin (TLV) and vancomycin (VAN) on spleen (a) and blood (b) bacterial titres in the murine bacteraemia model. $n=10$ for controls at all time points except at $t=10h$ ($n=9$) and $t=16h$ ($n=4$). $n=5$ for the VAN group at all time points except at $t=40h$ ($n=4$), $n=5$ for the TLV group at all time points except at $t=28h$ ($n=4$). Data represent mean $\pm$ 1 SEM. Arrow denotes time of dosing. LOQ, limit of quantification.

Figure 2. Effects of telavancin (TLV) and vancomycin (VAN) on survival in the murine MRSA bacteraemia model. $n=15$ at start of the study. Arrow denotes time of dosing. $^*P<0.05$ versus control and vancomycin groups.

for the greater efficacy of telavancin could be its longer plasma half-life in mice compared with vancomycin, resulting in a longer $t >$ MIC. However, AUC/MIC, as opposed to $t >$ MIC, has been implicated as the pharmacodynamically linked variable for both telavancin and vancomycin. A caveat of the present study is that a single strain of MRSA was examined and it remains to be determined whether the findings of this investigation extend to other strains.

Conclusions

In summary, the data from the present study demonstrate the efficacy of telavancin in reducing splenic and blood bacterial titres and improving survival in a model of bacteraemia caused by MRSA in neutropenic mice. These findings suggest that telavancin may have utility for treatment of MRSA bacteraemic infections.

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Transparency declarations

All authors are employees of Theravance and own stock in the company.

References

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