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Efficacy of Telavancin against Penicillin-Resistant Pneumococci and *Staphylococcus aureus* in a Rabbit Meningitis Model and Determination of Kinetic Parameters

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The penetration of telavancin was 2% into inflamed meninges and ca. 1‰ into noninflamed meninges after two intravenous injections (30 mg/kg of body weight). In experimental meningitis, telavancin was significantly superior to vancomycin combined with ceftriaxone against a penicillin-resistant pneumococcal strain. Against a methicillin-sensitive staphylococcal strain, telavancin was slightly but not significantly superior to vancomycin.

The continuous spread of penicillin-resistant pneumococci worldwide represents one of the major challenges for clinicians and infectious disease specialists. The epidemiological situation in Europe varies considerably. Increasing rates of penicillin resistance are observed everywhere. Although some countries in the northern part of Europe have a relatively low percentage of penicillin-resistant pneumococci, the highest rates have been reported from Hungary, with over 50% of the strains being penicillin resistant, followed by Spain and Portugal (1). Increasing rates of penicillin resistance have been registered in the United States as well in recent years, with resistance rates over 34% in some regions (3). In addition, resistance to cephalosporins has further jeopardized the therapeutic options for penicillin-resistant strains (11). Generally, β-lactam antibiotics remain the first-line drugs for the treatment of pneumococcal diseases, except when their penetration into target organs might be insufficient, as is the case in meningitis. Based on actual guidelines, the empirical treatment of pneumococcal meningitis consists of a combination of an expanded-spectrum cephalosporin with vancomycin, especially when resistant strains are suspected (7, 8). Another challenging issue remains the treatment of staphylococcal meningitis. *Staphylococcus aureus* is the cause of about 1 to 9% of all cases of bacterial meningitis, with mortality rates ranging from 14 to 77% (9, 14). In the majority of cases, meningitis due to *Staphylococcus aureus* is a nosocomially acquired disease and occurs in patients after neurosurgical procedures or head trauma. *S. aureus* is the second cause of cerebrospinal fluid (CSF) shunt infections (12 to 19% of the cases) (9, 14). In patients with community-acquired *S. aureus* meningitis, underlying conditions include sinusitis, endocarditis, abscess, cellulitis, osteomyelitis, and pneumonia. The rate of mortality in patients with *S. aureus* meningitis has been reported to be higher when the path of infection is hematogenous rather than postoperative (56% versus 18%) (6). In the case of β-lactam allergy or when methicillin-resistant *S. aureus* strains are suspected, vancomycin remains the treatment of choice.

Telavancin is a recently developed lipoglycopeptide with excellent and rapid bactericidal activity against the most relevant gram-positive microorganisms (10). The aim of this study was to evaluate the activities of telavancin against a penicillin-resistant pneumococcal strain and a methicillin-sensitive staphylococcal strain in experimental meningitis and to determine the kinetic parameters of its penetration into the CSF.

Pneumococcal strain. Pneumococcal strain WB4 (a penicillin-resistant serotype 6 strain; MIC, 4 mg/liter) was isolated from the blood of a patient at the Inselspital in Bern, Switzerland, and was provided by the Institute for Infectious Diseases at the University of Bern. This strain was grown in Muller-Hinton broth (MHB) to a density of approximately 10^8 CFU/ml and was then diluted to circa 10^6 CFU/ml for in vivo experiments. The MICs were determined in liquid cultures; and growth was controlled after 6, 12, and 24 h because of the spontaneous autolysis of the pneumococci. The MICs were as follows: ceftriaxone, 0.5 mg/liter, vancomycin, 0.12 mg/liter; and telavancin, 0.06 mg/liter.

Staphylococcal strain. Methicillin-susceptible *S. aureus* strain 1112 was kindly provided by José Entenza, Department of Infectious Diseases, University Hospital Lausanne. This strain has routinely been used in experimental models of endocarditis (4). The strain was grown in MHB to a density of approximately 10^8 CFU/ml and was then diluted for in vivo experiments. The MICs were determined in liquid cultures. The MICs were as follows: vancomycin, 1 mg/liter; and telavancin, 2 mg/liter.

Experimental meningitis model. The experimental rabbit meningitis model described by Dacey and Sande (2) was used in this project. The experimental protocols were approved by the federal veterinary office of the County of Bern.

Pathogen-free New Zealand rabbits (weight, 2.5 to 3 kg) were provided by the Zentraltierställe der Medizinischen Fakultät der Universität Bern, where all the experiments were performed.

One day before an experiment, the rabbits were anesthetized by intramuscular injection of a combination of ketamine and xylazine to fit a prosthesis on the calvaria to facilitate subsequent placement within a stereotactic frame. On the day

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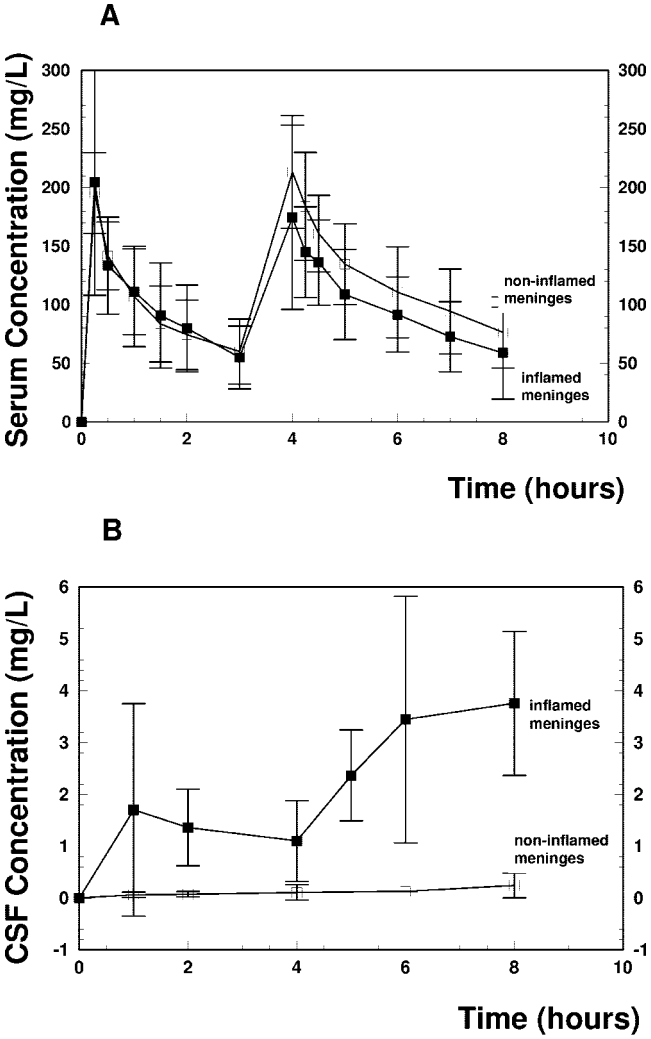

FIG. 1. Mean (±standard deviation) serum (A) and CSF (B) concentrations of telavancin in rabbits following intravenous administration of 30 mg/kg at 0 and 4 h.

of the experiment, the rabbits received 1.75 g/kg of body weight ethylcarbamate (urethane) subcutaneously and then 10 mg/kg pentobarbital intravenously to induce deep anesthesia. The animals were fixed in stereotactic frames, and a 3.5-in. (25-gauge) spinal needle was introduced into the cisterna magna. Following the withdrawal of 0.2 ml of CSF, either pneumococci or staphylococci (1×10^5 CFU in 0.2 ml of saline solution) were injected into the subarachnoid space. After inoculation, the animals were placed back in their cages for the night. The next day, the rabbits were again fitted in the frames by using the techniques and anesthesia described above. A catheter was fixed in the femoral artery for serum sampling. A spinal needle was again fixed in the subarachnoid space. Antibiotics were injected intravenously at the doses described in the literature (12, 13): ceftriaxone at 100 mg/kg and vancomycin at 20 mg/kg for the group infected with pneumococci and vancomycin at 20 mg/kg for the group infected with staphylococci. The ceftriaxone and vancomycin doses were the standard doses used for rabbits.

Telavancin was administered at a dose (twice at 30 mg/kg) that mimics the levels measured in the serum of humans. Vancomycin and telavancin were given at hours 0 and 4 and ceftriaxone was given at hour 0, according to their pharmacokinetic properties. CSF (0.2 ml) was sampled at 0, 1, 2, 4, 5, 6, and 8 h after the initiation of therapy. Blood samples were collected at 0.25, 0.5, 1, 2, 3, 4, 4.25, 4.5, 5, 6, 7, and 8 h after the initiation of therapy. Each group included untreated controls, which received a comparable volume of saline.

Determination of antibiotic levels and CFU titers. The free telavancin concentrations in serum and CSF were determined by high-performance liquid chromatography (kindly performed by Theravance Company). The numbers of CFU were measured by serial dilution of CSF, which was plated on agar plates (*S. aureus*) or on agar plates with 5% sheep blood (*Streptococcus pneumoniae*) and incubated overnight at 37°C in 5% CO_2.

Statistical analysis. The Student *t* test and one-way analysis of variance (Newman-Keuls multiple-comparisons test) were used for parametric data. Positive and negative cultures were compared by the two-tailed Fisher exact test. A *P* value of <0.05 was considered significant.

The kinetics of the free (non-protein-bound) telavancin (administered twice at 30 mg/kg) are presented in Fig. 1A and B. After one injection, serum telavancin levels peaked at about 200 mg/liter and slowly decreased to 50 mg/liter 4 h later. The second injection led to peak levels from 160 to 200 mg/liter and decreased to 70 to 50 mg/liter at the end of the treatment period (Fig. 1A). The dose used in this study (two doses of 30 mg/kg) corresponded approximately to the dose of 15 mg/kg recently tested in humans (15). The peak levels in serum were similar in both species, i.e., 210 to 213 mg/kg in rabbits and 203 mg/kg in humans. Because of the shorter half-life of telavancin in rabbits (4 h in rabbits versus 8 h in humans), a second dose was injected into the rabbits after 4 h in order to mimic the kinetics of telavancin in the serum of humans. In the CSF, the first injection of telavancin produced a peak level of 1.8 mg/liter in inflamed meninges and remained stable without a significant decrease during the next 4 h. In contrast, the second injection led to a progressive increase to 3.8 mg/liter at the end of the experimental period (Fig. 1B). As known from a previ-

TABLE 1. Noncompartmental pharmacokinetic parameters for telavancin following intravenous administration to rabbits

Compartment and parameter[a]	Healthy rabbits	Rabbits with inflamed meninges
Plasma		
No. of rabbits	3	5
C_{max} (mg/liter)	213	210
AUC_{0-8} (mg · h/liter)	931	830
$t_{1/2}$ (h)	4.0	2.3
CSF		
No. of rabbits	2	7
C_{max} (mg/liter)	0.130	5.14
AUC_{0-8} (mg · h/liter)	0.542	16.3
C_{max} ratio[b]	0.00076	0.0271
AUC ratio[c] (CSF/plasma)	0.00078	0.0191

[a] C_{max}, maximum concentration of drug; AUC_{0-8}, area under the concentration-time curve from time zero to 8 h; $t_{1/2}$, half-life.
[b] C_{max} for CSF/C_{max} for plasma.
[c] AUC for CSF/AUC for plasma.

TABLE 2. Activity of telavancin monotherapy compared to that of ceftriaxone combined with vancomycin against penicillin-resistant *S. pneumoniae* strain WB4 and compared to that of vancomycin against a methicillin-sensitive staphylococcal strain in experimental meningitis

Group (strain)[a]	Inoculum ($\log_{10}$ CFU/ml)	Killing rate/h ($\Delta\log_{10}$ CFU/ml · h)	Killing rate/8 h ($\Delta\log_{10}$ CFU/ml · 8 h)
Controls (Pen^r)	5.80 ± 0.15	+0.09 ± 0.04	+0.70 ± 0.20
TLV (Pen^r)	6.69 ± 0.53	−0.84 ± 0.23	−6.12 ± 1.14[b]
CRO + V (Pen^r)	5.87 ± 0.66	−0.61 ± 0.13	−4.75 ± 0.13[b]
Controls (MSSA)	5.31 ± 0.37	+0.09 ± 0.04	+0.80 ± 0.26
TLV (MSSA)	5.02 ± 0.19	−0.51 ± 0.19	−4.32 ± 0.93
V (MSSA)	5.20 ± 0.36	−0.44 ± 0.16	−3.58 ± 1.19
TLV (MSSA)	5.02 ± 0.19	−0.51 ± 0.19	−4.32 ± 0.93
V (MSSA)	5.20 ± 0.36	−0.44 ± 0.16	−3.58 ± 1.19

[a] Each group contained 10 animals. Strain abbreviations: Pen^r, penicillin resistant; MSSA, methicillin-susceptible *S. aureus.* Drug abbreviations: TLV, telavancin; CRO, ceftriaxone; V, vancomycin.
[b] $P < 0.001$ for telavancin versus ceftriaxone plus vancomycin.

ous study (5), vancomycin, when it is used at standard doses (twice at 20 mg/kg) in rabbits, produces CSF levels (1.5 to 4.0 mg/liter). Similar to those produced by telavancin. During the entire treatment period, the telavancin CSF levels remained above the MIC for both strains, leading to CSF/MIC ratios that ranged from 30 to 63 for the pneumococcal strain and from 0.9 to 1.9 for the staphylococcal strain. The penetration of unbound telavancin into inflamed meninges was about 2%, whereas that into noninflamed meninges was negligible and was lower than 1‰. However, due to the high level of protein binding (ca. 93%) (15), the real penetration of telavancin into inflamed meninges might be much higher. The pharmacokinetic parameters are summarized in Table 1.

The antibacterial activity of telavancin against the penicillin-resistant pneumococcal strain is presented in Table 2. In untreated controls, bacterial titers increased slowly (less than 1 $\log_{10}$ CFU/ml over 8 h). Before the initiation of treatment, the bacterial titer was significantly higher in the telavancin group (6.69 ± 0.53 versus 5.87 ± 0.66 for the comparator regimen; $P < 0.006$). Telavancin was highly bactericidal (−0.84 ± 0.24 change in $\log_{10}$ CFU/ml · h [$\Delta\log_{10}$ CFU/ml · h]) and produced a decrease in the viable cell count of 6.12 $\log_{10}$ at the end of the treatment period, managing to sterilize the CSF from 6 of 10 rabbits. The standard regimen based on a combination of vancomycin with ceftriaxone produced a less rapid decrease in the bacterial titer (−0.61 ± 0.13 versus −0.84 ± 0.23 $\Delta\log_{10}$ CFU/ml · h for telavancin; $P < 0.01$) and was significantly less efficacious after 8 h. The CSF from 4 of 10 of rabbits was sterile at the end of the experimental period when the standard regimen was used. The high degree of efficacy of telavancin against pneumococci is based on its high intrinsic bactericidal activity due to the unique dual mechanism of action of telavancin and on the high CSF/MIC ratios (from 30 to 63), which seems to be the major pharmacodynamic parameter of telavancin (16). Addition of a cephalosporin, e.g., ceftriaxone, as used in the standard regimen, might increase the efficacy of telavancin, in analogy to the synergy between β-lactam and glycopeptide antibiotics.

The activities of telavancin and vancomycin against a methicillin-sensitive staphylococcal strain are also summarized in Table 2. Before the start of treatment, the bacterial titer was comparable in all groups, ranging from 5.32 to 5.02 $\log_{10}$ CFU/ml. In untreated controls, the bacterial growth progressed slowly over 8 h (+0.8 $\log_{10}$ CFU/ml). Similar antibacterial activities were detected in both treatment groups. Although telavancin was slightly superior to vancomycin, the difference was statistically not significant after 8 h (−4.32 ± 0.93 versus −3.58 ± 1.19 $\log_{10}$ CFU/ml for telavancin and vancomycin, respectively). However, telavancin managed to sterilize the CSF of 6 of 10 rabbits, whereas the CSF samples of only 3 of 10 rabbits in the vancomycin treatment group were sterile.

Two factors are probably responsible for the relatively reduced activity of telavancin against staphylococci. First, the low CSF/MIC ratio of from 0.9 to 1.9 and the lower starting bacterial titer before the initiation of therapy may explain the lower decrease in the viable cell count after 8 h, although the number of CSF samples that were sterile was similar in both groups (staphylococci and pneumococci). Two of the four CSF samples from rabbits infected with *Staphylococcus aureus* that were not sterilized contained the lowest levels of telavancin. A higher dosage of telavancin will probably be more efficacious for the treatment of staphylococcal meningitis, mainly by increasing the CSF/MIC ratio. We are aware that vancomycin is not the standard regimen for the treatment of methicillin-sensitive staphylococcal meningitis, but the aim of this study was to compare telavancin to a standard glycopeptide antibiotic.

In summary, despite its relatively low level of penetration into inflamed meninges, telavancin was a very efficacious monotherapy against a penicillin-resistant pneumococcal strain and was even superior to the standard regimen (vancomycin plus ceftriaxone). Higher dosages of telavancin for the treatment of staphylococcal meningitis should be tested.

REFERENCES

1. **Adam, D.** 2002. Global antibiotic resistance in Streptococcus pneumoniae. J. Antimicrob. Chemother. **50**(Suppl.)**:**1–5.
2. **Dacey, R. G., and M. A. Sande.** 1974. Effect of probenecid on cerebrospinal fluid concentrations of penicillin and cephalosporin derivatives. Antimicrob. Agents Chemother. **6:**437–441.
3. **Doern, G. V., S. S. Richter, A. Miller, N. Miller, C. Rice, K. Heilmann, and S. Beekmann.** 2005. Antimicrobial resistance among Streptococcus pneumoniae in the United States: have we begun to turn the corner on resistance to certain antimicrobial classes? Clin. Infect. Dis. **41:**139–148.
4. **Entenza, J. M., J. Vouillamoz, M. P. Glauser, and P. Moreillon.** 1997. Levofloxacin versus ciprofloxacin, flucloxacillin, or vancomycin for treatment of experimental endocarditis due to methicillin-susceptible or -resistant *Staphylococcus aureus*. Antimicrob. Agents Chemother. **41:**1662–1667.
5. **Gerber, C. M., M. Cottagnoud, K. A. Neftel, M. G. Tauber, and P. Cottagnoud.** 1999. Meropenem alone and in combination with vancomycin in experimental meningitis caused by a penicillin-resistant pneumococcal strain. Eur. J. Clin. Microbiol. Infect. Dis. **18:**866–870.
6. **Jensen, A. G., F. Espersen, P. Skinhoj, V. T. Rosdahl, and N. Frimodt-Moller.** 1993. Staphylococcus aureus meningitis. A review of 104 nationwide, consecutive cases. Arch. Intern. Med. **153:**1902–1908.
7. **Kaplan, S. L.** 2002. Management of pneumococcal meningitis. Pediatr. Infect. Dis. J. **21:**589–591.
8. **Kaplan, S. L., and E. O. Mason, Jr.** 2002. Mechanisms of pneumococcal antibiotic resistance and treatment of pneumococcal infections in 2002. Pediatr. Ann. **31:**250–260.
9. **Kim, J. H., C. van der Horst, C. D. Mulrow, and G. R. Corey.** 1989. Staphylococcus aureus meningitis: review of 28 cases. Rev. Infect. Dis. **11:**698–706.
10. **King, A., I. Phillips, and K. Kaniga.** 2004. Comparative in vitro activity of telavancin (TD-6424), a rapidly bactericidal, concentration-dependent antiinfective with multiple mechanisms of action against gram-positive bacteria. J. Antimicrob. Chemother. **53:**797–803.

11. **Klugman, K. P.** 1996. Epidemiology, control and treatment of multiresistant pneumococci. Drugs **52**(Suppl. 2)**:**42–46.
12. **Paris, M. M., S. M. Hickey, M. I. Uscher, S. Shelton, K. D. Olsen, and G. H. McCracken, Jr.** 1994. Effect of dexamethasone on therapy of experimental penicillin- and cephalosporin-resistant pneumococcal meningitis. Antimicrob. Agents Chemother. **38:**1320–1324.
13. **Paris, M. M., O. Ramilo, and G. H. McCracken, Jr.** 1995. Management of meningitis caused by penicillin-resistant *Streptococcus pneumoniae*. Antimicrob. Agents Chemother. **39:**2171–2175.
14. **Schlesinger, L. S., S. C. Ross, and D. R. Schaberg.** 1987. Staphylococcus aureus meningitis: a broad-based epidemiologic study. Medicine (Baltimore) **66:**148–156.
15. **Shaw, J. P., J. Seroogy, K. Kaniga, D. L. Higgins, M. Kitt, and S. Barriere.** 2005. Pharmacokinetics, serum inhibitory and bactericidal activity, and safety of telavancin in healthy subjects. Antimicrob. Agents Chemother. **49:**195–201.
16. **Van Bambeke, F.** 2004. Glycopeptides in clinical development: pharmacological profile and clinical perspectives. Curr. Opin. Pharmacol. **4:**471–478.