MAJOR ARTICLE

Pharmacokinetics of Intravenous Polymyxin B in Critically Ill Patients

Alexandre P. Zavascki,[1,4] Luciano Z. Goldani,[1] Guoying Cao,[6,7] Silvana V. Superti,[5] Larissa Lutz,[2] Afonso L. Barth,[2] Fabiano Ramos,[4] Márcio M. Boniatti,[3] Roger L. Nation,[6] and Jian Li[6]

[1]Division of Infectious Diseases, [2]Microbiology Unit, Clinical Pathology Service, and [3]Intensive Care Unit, Hospital de Clínicas de Porto Alegre, and [4]Infectious Diseases Service and [5]Microbiology Service, Hospital São Lucas da Pontifícia Universidade Católica do Rio Grande do Sul, Porto Alegre, Brazil; [6]Facility for Anti-infective Drug Development and Innovation, Monash Institute of Pharmaceutical Sciences, Monash University, Parkville, Victoria, Australia; and [7]Department of Pharmacy, Beijing Hospital, Beijing, People's Republic of China

Background. Although not much pharmacokinetic knowledge is available, polymyxin B is increasingly used for treatment of infections caused by gram-negative bacteria that are resistant to all other antibiotics.

Methods. This study involved 8 patients who received intensive care after intravenous administration of a 60-min infusion of polymyxin B at currently recommended doses. Blood and urine samples were collected, and plasma protein binding of polymyxin B was determined. Concentrations of polymyxin B in plasma and urine samples were measured by a specific high-performance liquid chromatographic method.

Results. Polymyxin B was well tolerated. The peak plasma concentrations at the end of the infusion varied from 2.38 to 13.9 mg/L. For 4 patients from whom it was possible to collect urine samples over a dosing interval, only 0.04%–0.86% of the dose was recovered in the urine in unchanged form. Plasma protein binding of polymyxin B was higher in samples from patients (range, 78.5%–92.4%) than in plasma samples from healthy human subjects (mean ± standard deviation, 55.9% ± 4.7%). Unbound plasma concentrations of polymyxin B were in the vicinity of or lower than the minimum inhibitory concentration of the pathogen.

Conclusion. To our knowledge, this is the first study to report plasma concentrations over time and urinary recovery of polymyxin B in critically ill patients after intravenous administration. Polymyxin B is eliminated mainly by nonrenal pathways, and the total body clearance appears to be relatively insensitive to renal function. Additional investigations are required to assess the appropriateness of currently recommended doses of this drug for the treatment of severe infections in critically ill persons.

Since the 1990s, the increasing prevalence of infections caused by multidrug-resistant *Pseudomonas aeruginosa*, *Acinetobacter baumannii*, and *Klebsiella pneumoniae* has presented a major clinical problem worldwide [1–3]. In many cases, these pathogens are resistant to all currently available antibiotics except polymyxins [4, 5]. Unfortunately, there is a mismatch between the above-mentioned medical need and antimicrobial development; no novel class of antibiotics is being developed in the drug discovery and development pipeline for these multidrug-resistant, gram-negative pathogens [3]. Therefore, clinicians have been forced to use polymyxins as the last line of defense [4–8].

Polymyxins were commercially released in the late 1950s [9]. Subsequently, the frequency of use of these agents decreased because of concerns about their potential toxicity (e.g., nephrotoxicity and neurotoxicity) and the availability of less toxic antibiotics [4–8]. Interestingly, recent clinical reports have demonstrated the tolerability, safety, and effectiveness of intravenously administered polymyxins [10–13]. There are 2 polymyxins used clinically, polymyxin B and colistin (polymyxin E), which have similar antibacterial spectra and rapid bactericidal activity [14–16]. Unlike colistin, which is administered parenterally and by inhalation in the form of its inactive prodrug (colistin methanesulfonate sodium) [4], polymyxin B is administered as its sulfate salt (i.e., the microbiologically active entity) [5, 6]. Similar to colistin, polymyxin B is a multicompo-

Received 2 May 2008; accepted 23 July 2008; electronically published 7 October 2008.

Reprints or correspondence: Dr. Jian Li, Facility for Anti-infective Drug Development and Innovation, Monash Institute of Pharmaceutical Sciences, Monash University, 381 Royal Parade, Parkville, Victoria 3052, Australia (Jian.Li@pharm.monash.edu.au).

Clinical Infectious Diseases 2008; 47:1298–1304

1058-4838/2008/4710-0008$15.00
DOI: 10.1086/592577

nent antibiotic; polymyxin B1 and B2 are the forms that predominate [6].

Because polymyxins became available clinically before the advent of contemporary drug-development procedures, there are substantial gaps in knowledge of their pharmacology [4]. Recent progress in elucidating the pharmacology of the polymyxins has focused mainly on colistin [13, 17–21]. One exception was a prospective observational pharmacokinetic study of the polymyxin B1 component after administration of polymyxin B to a small sample of 9 adult patients who apparently were not critically ill [22]. In the product information for polymyxin B [23], no data are available on plasma concentration over time after intravenous administration [5]. Although the level of resistance to polymyxin B is currently low [24], resistance has been reported recently [25], possibly because of use of suboptimal dosing regimens arising from insufficient understanding of the pharmacokinetics and pharmacodynamics of the drug [26]. Therefore, the aim of the present study was to address the urgent need to investigate the clinical pharmacokinetics of polymyxin B among critically ill patients.

PATIENTS, MATERIALS, AND METHODS

Patients and ethics. The study protocol was approved by the Ethics Review Board of Pontificia Universidade Católica do Rio Grande do Sul and Hospital de Clínicas de Porto Alegre, Brazil, and the Monash University Standing Committee on Ethics in Research Involving Humans, Australia. Informed consent was obtained from the legal representatives of patients before entry in the study. Eight critically ill patients (4 male and 4 female patients) who were receiving intensive care and had ventilator-associated pneumonia caused by *P. aeruginosa* or *A. baumannii* entered this study. Clinical and demographic data for the 8 patients are presented in table 1. Criteria for exclusion were age <18 years; death within 72 h after the initiation of treatment; known hypersensitivity and/or allergy to polymyxin B, colistin, or other polymyxins; and no indication for treatment with polymyxin B by the attending physician.

Each dose of polymyxin B sulfate (Polymyxin B for Injection, Eurofarma; 500,000 U per vial; 1 mg contained ~10,000 U) was given as a 50-mL infusion over a 60-min period; the size of each dose and the dosing interval for each patient are shown in table 1. The decision to treat with polymyxin B and the dosing regimen for each patient were at the discretion of the attending physician. Renal function was monitored by daily determination of serum creatinine level; estimated creatinine clearance was calculated with use of the Cockcroft-Gault equation. Neurotoxicity was not systematically assessed, unless there was any clinical indicator of such toxicity. After at least 2 days of therapy had elapsed, blood samples (~4 mL) for pharmacokinetic analysis were collected in 4-mL heparinized Vacuette containers (Becton Dickinson) before the start of a 60-min polymyxin B infusion and at 60, 90, 120, 180, 360, and 720 min after the start of the infusion. The samples were immediately centrifuged for 10 min (4000 *g*) at 4°C, and plasma samples were stored at −80°C until analysis. Quantitative urine analyses were completed over the dosing intervals for patients 3 (day 3), 5 (day 5), 6 (day 3), and 8 (day 4).

Determination of polymyxin B concentrations in plasma and urine samples. The concentrations of polymyxin B in plasma and urine samples were measured by a sensitive high-performance liquid chromatographic (HPLC) method that was developed in our laboratory. In brief, proteins were precipitated by mixing plasma or urine samples (100 μL) with an equal volume of acetonitrile (100 μL), and after centrifugation (at 10,000 g for 10 min), the supernatant was transferred to a solid-phase extraction C_{18} cartridge (Sep-Pak; Waters), in which fluorescent derivatives were formed using 9-fluorenylmethyl chloroformate (Sigma). Elution of the derivatives was followed by reversed-phase HPLC with detection by fluorescence at 315 nm after excitation at 260 nm. The Shimadzu HPLC system consisted of an LC-10AS pump, an SIL-10ADvp auto injector, and an RF-10AXL fluorescence detector connected to a data processing system (Class-VP, version 6.14SP1). A 50×4.6 mm (internal diameter) Onyx Monolithic C_{18} column coupled with a 4×3.0 mm C_{18} guard column was used (Phenomenex). The mobile phase was acetonitrile-tetrahydrofuran-water (50:30:20), and the run time was 7 min. The concentrations of polymyxin B were calculated on the basis of the sum of the chromatographic peak areas of polymyxin B1 and B2 in the HPLC assay. The mean recovery percentages ($\pm$SD) achieved for polymyxin B in plasma samples were 103% $\pm$ 12.4% at 0.50 mg/L and 104% $\pm$ 6.0% at 4.00 mg/L; the corresponding values for urine samples were 89.7% $\pm$ 15.9% and 93.8% $\pm$ 9.0%. The limit of quantification was 0.10 mg/L. Analysis of independently prepared quality-control plasma (0.30, 3.00, and 15.0 mg/L) and urine (0.20 and 2.0 mg/L) samples indicated good accuracy (quality-control plasma and urine samples were within 9.3% and 10.0% of the actual concentrations, respectively) and reproducibility (coefficients of variation of 6.8% and 9.0%, respectively).

Pharmacokinetic analysis. Because blood samples for pharmacokinetic analysis were collected at least 2 days after the initiation of therapy with polymyxin B, it was assumed that the concentrations of polymyxin B in plasma were at a steady state. The areas under the plasma concentration–versus-time curves during a dosage interval for those patients for whom the dosing interval was 12 h were calculated with use of the linear trapezoidal rule (PK Functions for Microsoft Excel; Department of Pharmacokinetics and Drug Metabolism, Allergan). Total body clearance was determined as the dose divided by the area under the curve. Urinary recovery of polymyxin B was calculated as the amount of unchanged drug recovered in

Table 1. Clinical and demographic data for patients.

Patient	Age, years	Sex	APACHE II score	Body weight, kg	Polymyxin B dose, mg/kg	Dosing interval, h	Duration of polymyxin B treatment, days	Estimated creatinine clearance, mL/min			Time from initiation of treatment to sample collection, days	Pathogen (polymyxin B MIC, mg/L)	Coadministered antibiotic(s)
								At initiation of treatment	On the day of sample collection	At the end of the treatment			
1	80	F	22	60	0.83	48	8	<10	<10[a]	<10	3	Pa (NA)	Meropenem
2	52	M	26	80	1.25	12	14	83	108	139	3	Pa (NA)	Levofloxacin and vancomycin (after day 9)
3	42	M	26	50	1.0	12	9	98	61	57	3	Pa (2)	Ceftazidime (days 1 and 2) and trimethoprim-sulfamethoxazole (days 1–9) and ciprofloxacin (days 3–9)
4	72	F	21	75	1.0	12	14	60	60	55	5	Pa (2)	Aztreonam (days 8–15)
5	86	F	27	70	1.0	12	14	34	34	21	4	Ab (1)	None
6	72	M	24	67	1.5[b]	48	14	26	18	29	3	Pa (1)	None
7	65	M	23	60	0.5[c]	48	14	<10	<10[a]	24	3	Pa (0.5)	Vancomycin (days 7–15)
8	48	F	20	68	1.0	12	14	246	246	238	4	Pa (1)	Trimethoprim-sulfamethoxazole (days 1–14)

NOTE. The mean age (±SD) was 65 ± 16 years, the mean Acute Physiology and Chronic Health Evaluation (APACHE II) score (±SD) was 23.6 ± 2.6, the mean body weight (±SD) was 66 ± 9.5 kg, and the mean polymyxin B dose (±SD) was 1.0 ± 0.3 mg/kg. Ab, *Acinetobacter baumannii;* NA, not available; Pa, *Pseudomonas aeruginosa*.

[a] Receiving intermittent hemodialysis; pharmacokinetic analysis was conducted on a nondialysis day.

[b] First dose was 3.0 mg/kg.

[c] First dose was 2.0 mg/kg.

urine samples during the dosing interval divided by the dose; this information, together with total body clearance, allowed determination of renal clearance.

Binding of polymyxin B in human plasma. The binding of polymyxin B in plasma in vitro was measured by equilibrium dialysis. A Perspex dialysis cell unit contained 2 reservoirs (volume in each, 1 mL) separated by a semipermeable membrane (Spectra/Por-2; Spectrum Laboratories). Pooled plasma samples (volume, 1 mL) from each patient (except patient 6 because of insufficient sample volume) were dialyzed against the same volume of isotonic phosphate buffer (0.067 mol/L; pH, 7.4) at 37°C. The binding of polymyxin B was also determined in pooled human plasma samples from the Australian Red Cross; polymyxin B (sulfate) was added to these plasma samples to achieve a concentration of 8 mg/L. Samples of plasma and buffer were removed from each equilibrium dialysis reservoir after 12 h (shown in preliminary studies to be the time required for equilibration) and were stored at −80°C until they were analyzed as described above. The extent of protein binding was calculated as 1 minus the ratio of polymyxin B concentration in buffer to that in plasma and was expressed as a percentage.

RESULTS

No adverse events were reported during the study. Data on the renal function of the patients are presented in table 1. The plasma concentrations of polymyxin B over time for all patients are shown in figure 1. The peak plasma concentrations at the end of the 60-min infusion ranged from 2.38 to 13.9 mg/L. The concentrations of polymyxin B1 were ~4-fold higher than the concentrations of polymyxin B2 (data not shown). The pharmacokinetic parameters of polymyxin B are summarized in table 2. There was relatively little interindividual variability in total body clearance (range, 0.27–0.81 mL/min/kg) and volume of distribution (range, 71–194 mL/kg). Urinary recovery was extremely low, with each of the 4 patients for whom data were available excreting <1% of the administered dose in urine as unchanged (i.e., unmetabolized) polymyxin B (table 2). Consequently, the renal clearance of polymyxin B was very low (range, 0.00032–0.0039 mL/min/kg). In pooled plasma samples from 3 healthy human subjects (from the Australian Red Cross), the mean binding percentage (±SD) of polymyxin B was 55.9% ± 4.7%; the mean polymyxin B concentration (±SD) in plasma samples at the end of dialysis was 3.80 ± 0.27 mg/L. Binding of polymyxin B in the pooled plasma samples from the patients for whom pharmacokinetics were determined is shown in table 2; the mean polymyxin B concentration (±SD) in plasma samples at the end of dialysis had a range of 1.29–4.57 mg/L.

DISCUSSION

To our knowledge, this is the first study involving critically ill patients to report the plasma concentrations of polymyxin B (i.e., polymyxin B1 and polymyxin B2) that were achieved with currently used dosing regimens, as well as the corresponding key pharmacokinetic parameters, including total body clearance, urinary recovery, and the extent of plasma protein binding. An understanding of all 3 of these pharmacokinetic parameters is essential if we are to optimize the dosing regimen of polymyxin B on the basis of well-established pharmacokinetic and pharmacodynamic principles [27].

A recent study reported the pharmacokinetics of polymyxin B1 in the general patient population [22]. The study comprised 9 patients and involved collection of random blood samples (1–3 samples per patient were collected 3–22.5 h after receipt of polymyxin B). Because only serum concentrations of polymyxin B1 were measured, as noted by the authors [22], the concentrations of polymyxin B were underestimated.

We observed that concentrations of polymyxin B1 in plasma exceeded those of polymyxin B2 by ~4-fold, which is very similar to the relative proportion of these 2 components in the polymyxin B administered to the patients (data not shown). Thus, it is apparent that there are no major differences in key

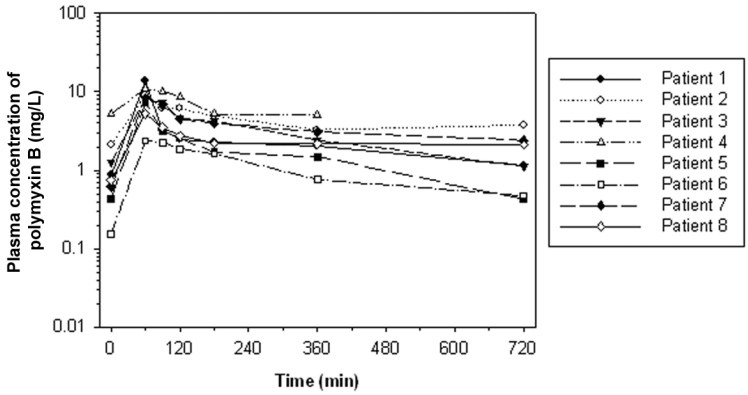

Figure 1. Concentrations of polymyxin B in plasma over time in 8 patients

Table 2. Pharmacokinetic parameters of polymyxin B in critically ill patients.

Patient	AUC_{0-12}, mg × h/L	Total body clearance, mL/min/kg	Urinary recovery, %	Volume of distribution, mL/kg	Plasma protein binding, %
1	NC	…	…	…	78.5
2	51.4	0.40	…	130	90.3
3	36.5	0.46	0.86	113	86.2
4	61.7	0.27	…	71	92.4
5	20.6	0.81	0.04	181	86.3
6	NC	…	0.06	…	…
7	NC	…	…	…	87.5
8	28.7	0.58	0.62	194	80.6

NOTE. AUC, area under the curve; NC, not calculated because samples were collected during only the first 12 h of the 48-h dosing interval.

pharmacokinetic parameters, such as total body clearance, between these 2 major components of polymyxin B. The plasma concentrations of polymyxin B shown in figure 1 should be considered in relation to the MICs of the pathogens (0.5–2 mg/L) (table 1). Although the total (i.e., bound plus unbound) plasma concentration of polymyxin B at the end of the 60-min infusion exceeded these MICs, it is important to also consider the extent of plasma protein binding. Because polymyxin B in the plasma of these patients was relatively highly bound (binding percentage, 78.5%–92.4%) (table 2), the unbound plasma concentrations were only a small proportion (7.6%–21.5%) of the total plasma concentrations shown in figure 1. Therefore, the maximal unbound plasma concentrations of polymyxin B were only in the vicinity of or lower than the MICs and the breakpoint (2 mg/L) against *P. aeruginosa* and *Acinetobacter* species [28]. It is likely that the suboptimal exposure to polymyxin B led to the poor clinical outcomes observed in this group of patients (A.P.Z., J.L, R.L.N., S.V.S., A.L.B., L.L., F.R., M.M.B., and L.Z.G., unpublished data). Clearly, systematic clinical pharmacokinetic and pharmacodynamic studies are urgently required to optimize clinical use of polymyxin B (used alone or in combination), to maximize antibacterial activity and minimize the potential for development of resistance.

It is interesting to note that the percentage of protein binding in plasma samples from critically ill patients (range, 78.5%–92.4%) was substantially higher than that observed in plasma samples from healthy human subjects (mean ± SD, 55.9% ± 4.7%). The plasma concentration of α_1-acid glycoprotein, an acute-phase reactant that is important for the binding of many basic drugs [29], is higher in critically ill patients [30] and in patients with sepsis [31], and this may explain the higher protein binding percentage observed in the patients. However, the protein(s) involved in the plasma binding of polymyxin B is currently unknown, and this is being investigated in our laboratory.

Although no clinical experimental data could be retrieved from the literature, a review in the 1970s [32] indicated that, within the first 12 h after injection of polymyxin B sulfate, the amount of drug recovered in urine was very low; thereafter, excretion increased, and with continued administration, ~60% of the dose was recovered in urine. It is not clear exactly how this percentage of urinary recovery was calculated and how it took account of the continued administration. In addition, it should be noted that nonspecific microbiological assays were used in those studies [32], which were conducted almost 50 years ago; such assays are problematic, especially when polymyxin B is coadministered with antibiotic(s) that is active against the test strain. In contrast, a specific HPLC assay was used in the present study. Less than 1% of the dose was recovered in unchanged form in the urine samples collected during a dosing interval at least 3 days after the first dose of intravenous polymyxin B. The low urinary recovery of unchanged polymyxin B occurred because the renal clearance was very low (mean ± SD, 0.0022 ± 0.0018 mL/min/kg; $n = 4$) in relation to total body clearance (table 2).

Although studies investigating urinary recovery of colistin in humans (after administration of its sulfate salt) are not available in the literature, in rats, the urinary recovery was <1% [18]; this value was similar to the one observed for polymyxin B in humans in the present study. Interestingly, the urinary recovery of colistin methanesulfonate sodium (a polyanion, nonactive prodrug of colistin) can be >60% after intravenous administration [13]; both colistin and polymyxin B are polycations, which probably explains the difference in renal handling between polymyxin B and/or colistin and colistin methanesulfonate sodium [4]. Thus, it is evident that nonrenal elimination is the predominant clearance pathway for both polymyxin B and colistin. Although renal clearance is only a very small contributor to the total body clearance of polymyxin B, it is important to recall that nephrotoxicity is arguably of the most

concern of the potential adverse effects. Therefore, it is important to understand the mechanisms involved in the renal handling of this drug. On the basis of knowledge of the glomerular filtration rate (creatinine clearance) and the renal clearance and plasma unbound fraction of polymyxin B, it is possible to determine whether net tubular secretion or reabsorption occurred. The renal clearance of polymyxin B in the patients was only 0.49%–2.33% of the anticipated clearance of polymyxin B by filtration at the glomerulus (calculated as the product of the unbound fraction of polymyxin B in plasma and creatinine clearance on the day of sample collection). Therefore, there must be very extensive net reabsorption of polymyxin B from tubular urine back into blood, which is a phenomenon that has also been observed for colistin in rats [18]. Extensive tubular reabsorption of polymyxin B (and colistin) may concentrate the drug inside tubular cells and, at least in part, explain the potential for nephrotoxicity.

The renal clearance of polymyxin B varied substantially (~12-fold) among patients, which was consistent with the very wide range of renal function in the patients in this study (range of creatinine clearance in patients for whom total body clearance and renal clearance of polymyxin B were determined, 20–240 mL/min). The total body clearance, however, was relatively similar in these patients (~3-fold the range). This is explained by our finding that the urinary recovery of polymyxin B was <1% of the administered dose. Under these circumstances, the total body clearance would be expected to be relatively insensitive to variations in renal function. Recommendation of polymyxin B dosing regimens for patients with various degrees of renal impairment must await additional pharmacokinetic data obtained from studies involving larger numbers of patients.

The limitations of this study need to be considered. As noted above, the number of patients was small, and additional studies involving critically ill patients are needed. In addition, clinical and ethical considerations precluded the collection of more blood samples during the dosing interval or extension of the collection period beyond 12 h. Notwithstanding these limitations, our study has yielded important new information on the plasma concentrations of polymyxin B that were achieved in critically ill patients who were receiving the currently used dosing regimens [23], the relatively low importance of renal versus nonrenal clearance and the resulting insensitivity of total body clearance to renal function, and higher plasma protein binding in critically ill patients.

In summary, our study indicates that the currently recommended dosing regimens of intravenous polymyxin B may lead to suboptimal antibiotic exposure against multidrug-resistant, gram-negative bacteria. This highlights the urgency to further investigate the clinical pharmacokinetics and pharmacodynamics of polymyxin B to optimize the use of this last-line antibiotic.

Acknowledgments

We thank Dr. Feda' EA Ali, Ms. Roxanne Owen, and Anima Poudayl for their excellent technical support.

Financial support. The National Council for Scientific and Technological Development, Ministry of Science and Technology, Brazil (152041/2006–0), and the Fundo de Incentivo à Pesquisa e Eventos, Hospital de Clínicas de Porto Alegre, Brazil. J.L. is an Australian National Health and Medical Research Council R. Douglas Wright Research Fellow.

Potential conflicts of interest. A.P.Z. has received speaking honoraria from Forest Laboratories. L.Z.G. and A.L.B. have received financial support from The National Council for Scientific and Technological Development, Ministry of Science and Technology, Brazil. All other authors: no conflicts.

References

1. Infectious Diseases Society of America. Bad bugs, no drugs. **2004**. Available at: http://www.idsociety.org/badbugsnodrugs.html. Accessed 3 October 2008.
2. Livermore DM. The need for new antibiotics. Clin Microbiol Infect **2004**; 10(Suppl 4):1–9.
3. Talbot GH, Bradley J, Edwards JE Jr, Gilbert D, Scheld M, Bartlett JG. Bad bugs need drugs: an update on the development pipeline from the Antimicrobial Availability Task Force of the Infectious Diseases Society of America. Clin Infect Dis **2006**; 42:657–68.
4. Li J, Nation RL, Turnidge JD, et al. Colistin: the re-emerging antibiotic for multidrug-resistant Gram-negative bacterial infections. Lancet Infect Dis **2006**; 6:589–601.
5. Zavascki AP, Goldani LZ, Li J, Nation RL. Polymyxin B for the treatment of multidrug-resistant pathogens: a critical review. J Antimicrob Chemother **2007**; 60:1206–15.
6. Evans ME, Feola DJ, Rapp RP. Polymyxin B sulfate and colistin: old antibiotics for emerging multiresistant gram-negative bacteria. Ann Pharmacother **1999**; 33:960–7.
7. Falagas ME, Kasiakou SK. Colistin: the revival of polymyxins for the management of multidrug-resistant gram-negative bacterial infections. Clin Infect Dis **2005**; 40:1333–41.
8. Li J, Nation RL, Milne RW, Turnidge JD, Coulthard K. Evaluation of colistin as an agent against multi-resistant gram-negative bacteria. Int J Antimicrob Agents **2005**; 25:11–25.
9. Goodwin NJ. Colistin sulfate versus sodium colistimethate. Ann Intern Med **1969**; 70:232–3.
10. Conway SP, Etherington C, Munday J, Goldman MH, Strong JJ, Wootton M. Safety and tolerability of bolus intravenous colistin in acute respiratory exacerbations in adults with cystic fibrosis. Ann Pharmacother **2000**; 34:1238–42.
11. Garnacho-Montero J, Ortiz-Leyba C, Jimenez-Jimenez FJ, et al. Treatment of multidrug-resistant *Acinetobacter baumannii* ventilator-associated pneumonia (VAP) with intravenous colistin: a comparison with imipenem-susceptible VAP. Clin Infect Dis **2003**; 36:1111–8.
12. Al-Aloul M, Miller H, Alapati S, Stockton PA, Ledson MJ, Walshaw MJ. Renal impairment in cystic fibrosis patients due to repeated intravenous aminoglycoside use. Pediatr Pulmonol **2005**; 39:15–20.
13. Reed MD, Stern RC, O'Riordan MA, Blumer JL. The pharmacokinetics of colistin in patients with cystic fibrosis. J Clin Pharmacol **2001**; 41: 645–54.
14. Li J, Turnidge J, Milne R, Nation RL, Coulthard K. In vitro pharmacodynamic properties of colistin and colistin methanesulfonate against *Pseudomonas aeruginosa* isolates from patients with cystic fibrosis. Antimicrob Agents Chemother **2001**; 45:781–5.
15. Owen RJ, Li J, Nation RL, Spelman D. In vitro pharmacodynamics of

colistin against *Acinetobacter baumannii* clinical isolates. J Antimicrob Chemother **2007**; 59:473–7.
16. Tam VH, Schilling AN, Vo G, et al. Pharmacodynamics of polymyxin B against *Pseudomonas aeruginosa.* Antimicrob Agents Chemother **2005**; 49:3624–30.
17. Li J, Coulthard K, Milne R, et al. Steady-state pharmacokinetics of intravenous colistin methanesulphonate in patients with cystic fibrosis. J Antimicrob Chemother **2003**; 52:987–92.
18. Li J, Milne RW, Nation RL, Turnidge JD, Smeaton TC, Coulthard K. Use of high-performance liquid chromatography to study the pharmacokinetics of colistin sulfate in rats following intravenous administration. Antimicrob Agents Chemother **2003**; 47:1766–70.
19. Li J, Milne RW, Nation RL, Turnidge JD, Smeaton TC, Coulthard K. Pharmacokinetics of colistin methanesulphonate and colistin in rats following an intravenous dose of colistin methanesulphonate. J Antimicrob Chemother **2004**; 53:837–40.
20. Li J, Rayner CR, Nation RL, et al. Pharmacokinetics of colistin methanesulfonate and colistin in a critically ill patient receiving continuous venovenous hemodiafiltration. Antimicrob Agents Chemother **2005**; 49:4814–5.
21. Le Brun PPH, de Boer AH, Mannes GPM, et al. Dry powder inhalation of antibiotics in cystic fibrosis therapy, part 2: inhalation of a novel colistin dry powder formulation—a feasibility study in healthy volunteers and patients. Eur J Pharm Biopharm **2002**; 54:25–32.
22. Kwa AL, Lim TP, Low JG, et al. Pharmacokinetics of polymyxin B1 in patients with multidrug-resistant Gram-negative bacterial infections. Diagn Microbiol Infect Dis **2008**; 60:163–7.
23. Polymyxin B for injection [package insert]. Bedford, OH: Bedford Laboratories, **2007**.
24. Gales AC, Jones RN, Sader HS. Global assessment of the antimicrobial activity of polymyxin B against 54731 clinical isolates of gram-negative bacilli: report from the SENTRY antimicrobial surveillance programme (2001–2004). Clin Microbiol Infect **2006**; 12:315–21.
25. Bratu S, Landman D, Haag R, et al. Rapid spread of carbapenem-resistant *Klebsiella pneumoniae* in New York City: a new threat to our antibiotic armamentarium. Arch Intern Med **2005**; 165:1430–5.
26. Li J, Nation RL. Old polymyxins are back, is resistance close? Clin Infect Dis **2006**; 43:663–4.
27. Nicolau DP, Quintiliani R, Nightingale CH. Antibiotic kinetics and dynamics for the clinician. Med Clin North Am **1995**; 79:477–95.
28. Clinical and Laboratory Standards Institute (CLSI). Performance standards for antimicrobial susceptibility testing: 18th informational supplement. CLSI document M100-S18. Wayne, PA: CLSI, **2008**.
29. Piafsky KM, Borga O. Plasma protein binding of basic drugs. II. Importance of α 1-acid glycoprotein for interindividual variation. Clin Pharmacol Ther **1977**; 22:545–9.
30. Zamacona MK, Suarez E, Aguilera L, Rodriguez S, Aguirre C, Calvo R. Serum protein binding of propofol in critically ill patients. Acta Anaesthesiol Scand **1997**; 41:1267–72.
31. De Paepe P, Belpaire FM, Buylaert WA. Pharmacokinetic and pharmacodynamic considerations when treating patients with sepsis and septic shock. Clin Pharmacokinet **2002**; 41:1135–51.
32. Hoeprich PD. The polymyxins. Med Clin North Am **1970**; 54:1257–65.