ANTIMICROBIAL AGENTS AND CHEMOTHERAPY, Mar. 1986, p. 395–399
0066-4804/86/030395-05$02.00/0

Vol. 29, No. 3

Comparative Evaluation of Ciprofloxacin, Enoxacin, and Ofloxacin in Experimental *Pseudomonas aeruginosa* Pneumonia

BERND KEMMERICH, GLORIA J. SMALL, AND JAMES E. PENNINGTON*

Infectious Diseases Division, Department of Medicine, Brigham and Women's Hospital, and Harvard Medical School, Boston, Massachusetts 02115

Received 2 October 1985/Accepted 23 December 1985

The therapeutic activity of ciprofloxacin, enoxacin, and ofloxacin was evaluated in guinea pigs with acute and chronic experimental *Pseudomonas aeruginosa* pneumonia. Intratracheal instillations of *P. aeruginosa* resulted in fatal pneumonia in all untreated animals within 36 h. Among treatment groups (80 mg/kg [body weight] per day), cumulative survival rates were: 47%, ciprofloxacin; 55%, enoxacin; and 42%, ofloxacin. These rates were not significantly different. Intrapulmonary killing of *P. aeruginosa* was equivalent 3 h after a single dose of ciprofloxacin or ofloxacin (20 mg/kg) or enoxacin (40 mg/kg). The combination of ciprofloxacin with azlocillin, ceftazidime, or tobramycin did not increase the efficacy of intrapulmonary killing of *P. aeruginosa* over that of ciprofloxacin alone. A chronic, nonfatal bronchopneumonia was induced in guinea pigs by intratracheal instillation of microscopic agar beads impregnated with a mucoid strain of *P. aeruginosa*. Compared with no treatment, ciprofloxacin and enoxacin produced ≥99.9% intrapulmonary killing, and ofloxacin sterilized the lungs completely, after 4 days of treatment. In no quinolone-treated animal did resistant strains of *P. aeruginosa* emerge during 4-day treatment periods. In further studies with the chronic model, oral and parenteral ciprofloxacin treatment were found to be equivalent in efficacy. We conclude that several quinolone derivatives may be effective for the treatment of *P. aeruginosa* pneumonia and that combinations of quinolones with β-lactams or aminoglycosides may not increase efficacy against *P. aeruginosa* pneumonia.

Ciprofloxacin, enoxacin, and ofloxacin are three newly developed quinoline carboxylic acid derivatives with excellent in vitro activity against *Pseudomonas aeruginosa* (1, 2, 4, 7, 9, 15, 16, 29, 33). These compounds also offer interesting therapeutic alternatives for the treatment of *P. aeruginosa* infections because of their prolonged half-life and high tissue penetration (5, 6, 10, 14, 17, 28, 34, 35). The high rate of absorption after oral administration (6, 14, 17, 34) might be particularly useful in the treatment of chronic *P. aeruginosa* bronchopulmonary infections, such as those seen in patients with cystic fibrosis.

Ciprofloxacin was shown recently to be as active as tobramycin and superior to ticarcillin in experimental *P. aeruginosa* pneumonia in guinea pigs (31). No such information is available for enoxacin and ofloxacin. Also, information about the oral treatment of experimental lung infections and about combination therapy of quinolones with β-lactams or aminoglycosides for respiratory infection is not available. The objectives of the present study were (i) to investigate the comparative efficacy of ciprofloxacin, enoxacin, and ofloxacin in acute and chronic *P. aeruginosa* pneumonia, (ii) to provide an in vivo evaluation of monotherapy with ciprofloxacin versus combination with a *Pseudomonas*-active β-lactam or an aminoglycoside, and (iii) to compare oral and parenteral administration of ciprofloxacin in chronic *P. aeruginosa* bronchopneumonia.

MATERIALS AND METHODS

Animals. Disease-free Hartley strain guinea pigs weighing 350 to 400 g each were obtained from the Charles River Breeding Laboratories, Inc., Wilmington, Mass. The animals were housed in standard cages and fed guinea pig chow (Ralston-Purina, St. Louis, Mo.) and water.

Antibiotics. Antibiotics used in this study were ciprofloxacin (Bay o 9867) and azlocillin sodium (Miles Pharmaceuticals, West Haven, Conn.), enoxacin acetate (CI-919, AT-2266; Warner-Lambert Co., Ann Arbor, Mich.), ofloxacin (ORF 18489; Ortho Pharmaceutical Corp., Raritan, N.J.), ceftazidime (Glaxo Inc., Fort Lauderdale, Fla.), and tobramycin sulfate (Eli Lilly & Co., Indianapolis, Ind.). Ciprofloxacin, azlocillin, and ceftazidime were supplied as a powder and reconstituted in water to a final concentration of 20, 250, and 250 mg/ml, respectively. Powdered ofloxacin was dissolved in 0.1 N NaOH to a concentration of 10 mg/ml and thereafter titrated with concentrated HCl to a neutral pH. Enoxacin, supplied as a solution of 200 mg/ml, was diluted in water to a concentration of 40 mg/ml, and tobramycin was diluted with isotonic saline to a final concentration of 4 mg/ml.

Bacteria. Three clinical isolates of *P. aeruginosa* were used for infections. Strains 220 and A-5 were used in the acute pneumonia experiments, and the mucoid strain 2192M was used in the chronic model. The origins and characteristics of these strains were described previously (21, 22, 31). Maintenance and preparation of the bacteria for experimental infections were done as reported elsewhere (24). The MICs and MBCs of the antibiotics used in this study against the three challenge strains were determined by a microdilution method in Mueller-Hinton broth supplemented with calcium (50 μg/ml) and magnesium (25 μg/ml) by using a standard inoculum of 10^5 CFU/ml (32). The MBC was defined as the drug concentration killing ≥99.9% of the original inoculum. The checkerboard technique was used to test for synergism. Doubling dilutions of one drug were tested in combination with various concentrations of the other antibiotic. The interaction was defined as synergism if the sum of the fractional inhibitory concentrations (Σ FIC) of a checkerboard combination was ≤0.7, and it was defined as addition for Σ FIC of 0.71 to 1.29 and as antagonism for Σ FIC ≥1.3 (13).

* Corresponding author.

TABLE 1. Microbiological and pharmacokinetic studies for challenge strains and study drugs

Study drug (dose [mg/kg])	Concn (μg/ml) for strain						Peak concn in serum (μg/ml)[a]	Half-life (min)[b]
	220		A-5		2192M			
	MIC	MBC	MIC	MBC	MIC	MBC		
Ciprofloxacin (20)	0.25	0.25	0.25	0.25	0.125	0.125	2.37 ± 0.58	107
Enoxacin (20)	1.6	1.6	1.6	1.6	0.4	0.4	3.37 ± 0.23	115
Enoxacin (40)							7.90 ± 1.12	139
Ofloxacin (20)	0.78	0.78	0.78	0.78	0.4	0.4	10.1 ± 0.8	87

[a] Means ± standard deviation for three animals per group (30 to 60 min after injection).
[b] Mean half-lives in serum, calculated as described in the text.

Pharmacokinetic studies. To establish therapeutic regimens for the quinolones used in this study which would result in clinically relevant peak concentrations in serum exceeding the MICs and MBCs of the challenge strains and to determine their half-lives as a basis for dosing intervals, pharmacokinetic experiments were done in groups of three-to-four animals. A single dose of 20 mg of each study drug per kg of body weight was given subcutaneously (s.c.). For enoxacin, a 40-mg/kg dose was also used. Blood was obtained by cardiac puncture after 0.5, 1, 2, and 5 h. Sera were assayed by a high-pressure liquid chromatography assay for ofloxacin (kindly performed by Soledad Flor of Ortho), and microbiological assays (agar diffusion) (3) for ciprofloxacin and enoxacin were done with the assay organisms *Klebsiella pneumoniae* ATCC 10031 and *Escherichia coli* Kp (18), respectively.

Assuming an extravascular one-compartment model as sufficient to describe the kinetics of the quinolones for our purposes, data points of the postabsorptive phase were fit visually to a straight line on a semilog plot. Biological half-lives were then calculated from these monoexponential declining lines by using standard methodology (12, 26). Peak levels in serum and half-lives for tobramycin, azlocillin, and ceftazidime in guinea pigs were reported previously (23), and these measurements were not repeated.

Acute *Pseudomonas* pneumonia. The experimental model of acute *P. aeruginosa* pneumonia in guinea pigs was described in detail elsewhere (24). Briefly, the animals were inoculated intratracheally with either strain 220 or A-5 after intraperitoneal anesthesia with pentobarbital. Two different types of experiments, lung clearance studies and survival studies, were performed. The mean inocula for survival studies were 3.3×10^7 (range, 1.9×10^7 to 5.0×10^7) CFU of strain 220 and 6.1×10^7 (range, 4.5×10^7 to 8.6×10^7) CFU of strain A-5. Previous studies showed that these challenge doses result in a fatal bacteremic and hemorrhagic pneumonia in all untreated animals (23). Dosage regimens were based on measured concentrations in serum and calculated half-lives. Ciprofloxacin, enoxacin, and ofloxacin (20 mg/kg) and enoxacin (40 mg/kg) were given s.c. at 8 a.m. and 3 p.m. and at a double-strength dose at 10 p.m. Dose 1 was administered 1 h after infection. Control animals received three s.c. injections of 0.25 ml of isotonic saline daily. Because prior studies with this model showed that animals surviving 72 h will survive without further treatment, antibiotics were given for a total of 3 days. Experiments included equal numbers of animals in each treatment group. Survival times were recorded, and the percentage of survival after 3 days of treatment was calculated for each group. Because the influence of drug toxicity on survival could be excluded from studies with the nonlethal chronic pneumonia model, as well as from previous studies (31), further studies for the evaluation of drug toxicity in noninfected animals were not included.

Similar inocula were used to study the intrapulmonary killing of *P. aeruginosa*. Animals were treated 1 h after inoculation of the bacteria with a single dose of the above-described regimens or else with a combination of ciprofloxacin (20 mg/kg) with either azlocillin (125 mg/kg), ceftazidime (100 mg/kg), or tobramycin (1.7 mg/kg). β-Lactams and tobramycin were administered intramuscularly in hind flank sites. Animals were sacrificed 4 h after infection, and the lungs were removed and homogenized in 25 ml of sterile water (Waring blender; Dynamics Corp. of America, New Hartford, Conn.) and cultured quantitatively. Blood cultures were obtained at the same time by direct cardiac aspiration.

Chronic *Pseudomonas* pneumonia. The activity of the three quinolones in persistent, nonfatal bronchopulmonary infection by *P. aeruginosa* was evaluated in guinea pigs by using inocula of agar beads impregnated with a mucoid strain of *P. aeruginosa*, 2192 M, as described previously (20). Agar bead slurries containing approximately 1.5×10^7 CFU of *Pseudomonas* organisms per lung inoculum at a volume of 0.2 ml were instilled intratracheally. Treatment was started 3 days after infection and included ciprofloxacin, enoxacin, and ofloxacin (20 mg/kg) or enoxacin (40 mg/kg), given s.c. three times daily (8 a.m., 2 p.m., and 8 p.m.). Control groups received 0.9% saline, also thrice daily. Seven days after inoculation and 4 days after treatment was started, intrapulmonary killing of the challenge strain was determined with quantitative lung cultures, as described above. In addition, lung cultures were prepared in agar containing 4 μg of the quinolone used for treatment per ml. Blood cultures were obtained at the same time.

In separate experiments, the efficacy of parenteral (s.c.) versus oral (p.o.) ciprofloxacin was evaluated by using a dosage of 10 mg/kg (s.c.) or 40 mg/kg (p.o.) three times daily. Both regimens resulted in comparable peak concentrations in serum. The p.o. doses were prepared in 1 ml of water and could be swallowed easily by the animals from a syringe.

Statistical analysis. Student's two-tailed *t* test was used to analyze differences among lung clearance data and survival time for different regimens. Survival rates were compared by chi-square analysis with Yates's correction.

RESULTS

In vitro and pharmacokinetic studies. Ciprofloxacin displayed the highest in vitro activity against each of the three challenge strains of *P. aeruginosa* (Table 1). The MICs and MBCs of ofloxacin and enoxacin were three- to fivefold higher than those for ciprofloxacin. There was little difference between the MICs and MBCs of each study drug for the inoculum used (10^5 CFU/ml).

The combination of ciprofloxacin with a β-lactam showed

TABLE 2. In vitro synergy assays with challenge strains

Study drugs[a]	Σ FIC[b] for strain	
	220	A-5
Ciprofloxacin-azlocillin	0.5	1.0
Ciprofloxacin-ceftazidime	1.0	1.0
Ciprofloxacin-tobramycin	1.5	2.0

[a] MICs (μg/ml) of azlocillin, 16 (strain 220) and 8 (strain A-5). MICs of ceftazidime, 1.5 (strain 220) and 1.5 (strain A-5). MICs of tobramycin, 2 (strain 220) and 1 (strain A-5) (23, 30).
[b] For a definition, see Materials and Methods (13).

synergism for azlocillin against strain 220 (Σ FIC, 0.5) and additive activity in all other instances (Table 2). When tobramycin was combined with ciprofloxacin, antagonistic interaction could be observed (Σ FIC, 1.5 and 2.0).

The dosages of drug used in this study resulted for each antibiotic in concentrations in serum exceeding the MICs and MBCs of the challenge strains. Peak concentrations in serum occurred 30 to 60 min after s.c. administration (Table 1). Ofloxacin achieved higher concentrations in serum than enoxacin and ciprofloxacin at each sampling time. Mean concentrations in serum in guinea pigs receiving 20 mg of ofloxacin per kg were 9.3, 9.4, 2.58, and 1.55 μg/ml after 0.5, 1, 2, and 5 h. Comparable concentrations of ciprofloxacin in serum were 2.4, 1.9, 1.2, and 0.46 μg/ml, and concentrations of enoxacin were 3.0, 3.4, 2.5, and 0.8 μg/ml after the same dose. Concentrations of enoxacin in serum after a 40-mg/kg dose were 7.7, 7.4, 5.0, and 2.5 μg/ml for these time periods.

Acute *Pseudomonas* pneumonia. Ciprofloxacin and ofloxacin at 20 mg/kg were highly active in intrapulmonary killing of *P. aeruginosa* 4 h after infection (Table 3). Against strain 220, enoxacin was significantly less effective at the same dose. Doubling its dose resulted in a significant increase in its anti-*Pseudomonas* in vivo activity. Combination therapy with ciprofloxacin plus azlocillin, ceftazidime, or tobramycin did not show significantly increased intrapulmonary killing compared with treatment with ciprofloxacin alone. All regimens prevented bacteremia effectively (Table 3). Treatment with each of the quinolones improved survival in acute *P. aeruginosa* pneumonia significantly versus controls (Table 4). In spite of its relatively lower early intrapulmonary killing activity, the low-dose regimen of enoxacin showed excellent results for survival.

Chronic *Pseudomonas* pneumonia. Seven days after infection with mucoid strain 2192M-impregnated agar beads and 4 days after the start of therapy, all regimens had reduced the number of viable *P. aeruginosa* in the lungs significantly versus controls (Table 5). The most active compound was ofloxacin, which completely eliminated the pathogens from the lungs of infected guinea pigs after 4 days of treatment. Enoxacin showed a pronounced dose response in this model. Doubling the dose from 20 to 40 mg/kg three times daily resulted in a mean further reduction of about 2 $\log_{10}$ CFU per lung. Blood cultures were negative in all treated and control animals. The emergence of resistant bacteria was not observed when cultures were performed with the antibiotic-containing agar.

In additional studies with the chronic model, p.o. (40 mg/kg per dose) and s.c. (10 mg/kg per dose) ciprofloxacin administration were compared for efficacy. Mean concentrations (μg/ml ± standard deviation) in serum 1 h postdosing were 0.84 ± 0.15 (p.o.) and 1.28 ± 0.11 (s.c.). After 4 days of treatment, mean CFUs per ml of lung homogenate ($\log_{10}$; eight animals per group) were 4.70 ± 0.19, controls; 2.09 ± 0.29, p.o. (less than control; $P < 0.001$); and 3.08 ± 0.55, s.c. (less than control, $P < 0.01$).

DISCUSSION

There is increasing evidence for the in vivo activity of newly developed quinolones in experimental *P. aeruginosa* infections. Studies have included pneumonia in normal and neutropenic guinea pigs (11, 31) and osteomyelitis (19). Comparisons with β-lactams or aminoglycosides in these models demonstrated similar or even higher anti-*Pseudomonas* activity of the quinolones (19, 31). In our study of the comparative efficacy of three quinolones in acute and chronic *P. aeruginosa* pneumonia, ofloxacin and ciprofloxacin displayed greater intrapulmonary killing than enoxacin when the drugs were used at the same dosages. The higher in vitro activity of ciprofloxacin against the *P. aeruginosa* challenge strains and the higher peak levels of ofloxacin in serum may explain these results. Despite less early intrapulmonary killing of viable *P. aeruginosa* with enoxacin treatment, this agent produced excellent results in the survival studies with acute pneumonia.

Factors that may contribute to the favorable results with these agents in experimental *P. aeruginosa* infections are their microbiological (1, 2, 4, 7, 9, 15, 16, 29, 33) and pharmacokinetic (5, 10, 14, 17, 28, 34, 35) properties. In contrast to that of many β-lactam antibiotics, the bactericidal activity of quinolones is independent of the inoculum size for most *P. aeruginosa* isolates (4, 9, 29). This property of drug action appears to be particularly useful in treating life-threatening (high inoculum) *P. aeruginosa* infections of the lungs (23). The half-lives of the quinolones used in this study are considerably longer than those of most β-lactams with *P. aeruginosa* activity and of aminoglycosides, both in humans and in guinea pigs (23, 31). Thus, sustained levels of quinolones in serum could contribute to lung penetration, as shown by others (28). High tissue penetration of quinolones is also suggested by their large volume of distribution (14).

Dosages for our study drugs resulted in levels in serum above the MICs and MBCs for the challenge strains. Also, the levels in serum achieved with the 20-mg/kg doses were

TABLE 3. Bacteriological results of acute *P. aeruginosa* pneumonia

Treatment (single dose 1 h after infection [mg/kg])	Viable bacteria in lung 4 h after infection ($\log_{10}$ CFU)[a] for strain		Blood cultures (no. positive/total)
	220	A-5	
Saline	6.70 ± 0.50[b]	6.06 ± 0.39[b]	12/14
Ciprofloxacin (20)	3.03 ± 0.48	3.93 ± 0.52	1/14
Plus azlocillin (125)	2.85 ± 0.52	3.59 ± 1.39	0/14
Plus ceftazidime (100)	3.35 ± 0.38	3.83 ± 0.39	0/14
Plus tobramycin (1.7)	3.05 ± 0.52	4.39 ± 0.63	0/14
Enoxacin (20)	4.39 ± 0.32[c]	4.42 ± 0.55	2/16
Enoxacin (40)	3.60 ± 0.22	4.01 ± 0.66	1/8
Ofloxacin (20)	3.34 ± 0.63	3.42 ± 0.34	0/14

[a] Values are expressed as means ± standard deviation of $\log_{10}$ CFU of *P. aeruginosa* per ml of lung homogenate. Dilution factor was × 25. Eight to 16 animals per group.
[b] Significantly higher than all treatment groups ($P < 0.001$).
[c] Significantly higher than all other treatment groups ($P < 0.01$ to 0.001).

TABLE 4. Survival of animals with acute *P. aeruginosa* pneumonia

Treatment (mg/kg per day)	No. of survivors/no. of animals infected (%) with strain		Cumulative survival (%)	Survival time (h)[a] for strain		
	220	A-5		220	A-5	Cumulative
Saline	0/14 (0)[b]	0/12 (0)[b]	0[b]	15.4 ± 5.3[b]	16.6 ± 12.6[b]	15.9 ± 9.3[b]
Ciprofloxacin (80)	5/15 (33)	9/16 (60)	47	50.9 ± 20.5	58.2 ± 23.4	54.3 ± 21.8
Enoxacin (80)	8/15 (53)	9/16 (56)	55	52.6 ± 24.6	53.8 ± 24.3	53.2 ± 24.1
Enoxacin (160)	6/15 (40)	6/16 (38)	39	51.1 ± 20.4	51.7 ± 22.8	51.4 ± 21.3
Ofloxacin (80)	7/15 (47)	6/16 (38)	42	46.2 ± 24.6	45.3 ± 24.0	45.7 ± 23.9

[a] Values are expressed as means ± standard deviation. Long-term survivors were assigned a value of 72 h.
[b] Significantly different from all other treatment groups ($P < 0.05$ to 0.001).

similar to those recently reported for these agents in clinical trials and pharmacokinetic studies in humans (6, 8, 10, 17, 25, 34). Because of the shorter half-lives of the quinolones in guinea pigs, we decided to administer the daily dose in three divided doses and used a double-strength dose at night for the acute lung infections, whereas in reported clinical studies the dose interval has been 12 h (8, 25).

There is only limited information about the in vitro interaction of quinolones with other antibiotics. In one study, the highest rate of in vitro synergism for ciprofloxacin against *P. aeruginosa* was found in combination with azlocillin (27). Also, ceftazidime has shown synergism with ciprofloxacin against *P. aeruginosa* for 40% of strains tested, whereas aminoglycosides are rarely synergistic with ciprofloxacin against *P. aeruginosa* (27; J. A. Moody, L. R. Peterson, and D. N. Gerding, Program Abstr. 24th Intersci. Conf. Antimicrob. Agents Chemother., abstr. no. 393, 1984; R. Yarrish, L. Mack, J. Goldfarb, K. Van Horn, C. Gedris, G. Wormser, and A. Mascia, Program Abstr. 25th Intersci. Conf. Antimicrob. Agents Chemother., abstr. no. 403, 1985). Even less information is available regarding combination therapy with any of the three quinolones. Despite synergistic or additive in vitro interaction of ciprofloxacin with azlocillin or ceftazidime against our challenge strains, these combination regimens provided no significant increase in early intrapulmonary killing in this study.

Both the urine recovery of about 50% of p.o. administered ciprofloxacin and its absolute bioavailability of 63 to 77% (5, 14) indicate sufficient gastrointestinal absorption of this agent, but there are no reports about the efficacy of oral ciprofloxacin in experimental *P. aeruginosa* infections. Our comparative trial with both routes of administration demonstrated equivalent intrapulmonary killing in chronic *P. aeruginosa* bronchopneumonia in guinea pigs with dosages resulting in comparable peak concentrations in serum. Urine recovery data suggest an even better bioavailability for enoxacin (34) and ofloxacin (17; A Saito and M. Tomizawa, Proc. 13th Int. Congr. Chemother., SP 4.6/5–4, 1983). Thus, effective oral treatment of more chronic *P. aeruginosa* respiratory infections, such as bronchiectasis or chronic bronchitis in cystic fibrosis patients, should be possible with each of these quinolones. Comparative clinical studies of oral quinolones with conventional therapy in such patients would be useful.

In summary, our results suggest that ciprofloxacin, enoxacin, and ofloxacin may be highly active therapeutic alternatives for either acute or more chronic forms of *P. aeruginosa* lung infection and that combination therapy does not enhance ciprofloxacin activity against *P. aeruginosa* lung infection. Clinical evaluation of these quinolone agents for the treatment of *P. aeruginosa* pneumonia appears to be justified.

TABLE 5. Microbiological results of chronic *P. aeruginosa* pneumonia

Treatment (mg/kg three times daily)	Viable bacteria/ml of lung homogenate (log_{10} CFU)[a]
Saline	5.86 ± 0.61[b]
Ciprofloxacin (20)	2.34 ± 0.52
Enoxacin (20)	4.62 ± 0.62[c]
Enoxacin (40)	2.84 ± 0.65
Ofloxacin (20)	No growth

[a] Numbers are means ± standard deviation ($n = 8$ per group) 7 days after infection and 4 days after the start of treatment. Dilution factor was × 25.
[b] Higher than for all treatment groups ($P < 0.01$ to 0.001).
[c] Different from all other treatment groups ($P < 0.001$).

ACKNOWLEDGMENTS

This work was supported in part by research grants from Miles Pharmaceuticals, the Warner-Lambert Co., and the Ortho Pharmaceutical Corp.

We thank B. Painter, C. Heifetz, M. Cohen, and E. Tolman for their helpful advice and assistance. We also thank Lynn Sayers for preparing the manuscript.

LITERATURE CITED

1. **Barry, A. L., R. N. Jones, C. Thornsberry, L. W. Ayers, E. H. Gerlach, and H. M. Sommers.** 1984. Antibacterial activities of ciprofloxacin, norfloxacin, oxolinic acid, cinoxacin, and nalidixic acid. Antimicrob. Agents Chemother. **25**:633–637.
2. **Bassey, C. M., A. L. Baltch, R. P. Smith, and P. E. Conley.** 1984. Comparative in vitro activities of enoxacin (CI-919, AT-2266) and eleven antipseudomonal agents against aminoglycoside-susceptible and -resistant *Pseudomonas aeruginosa* strains. Antimicrob. Agent Chemother. **26**:417–418.
3. **Bennett, J. V., J. L. Brodie, E. J. Benner, and W. M. M. Kirby.** 1966. Simplified, accurate method for antibiotic assay of clinical specimens. Appl. Microbiol. **14**:170–177.
4. **Chartrand, S. A., R. K. Scribner, A. H. Weber, D. F. Welch, and M. I. Marks.** 1983. In vitro activity of CI-919 (AT-2266), an oral antipseudomonal compound. Antimicrob. Agents Chemother. **23**:658–663.
5. **Crump, B., R. Wise, and J. Dent.** 1983. Pharmacokinetics and tissue penetration of ciprofloxacin. Antimicrob. Agents Chemother. **24**:784–786.
6. **Davis, R. L., J. R. Koup, J. Williams-Warren, A. Weber, and A. L. Smith.** 1985. Pharmacokinetics of three oral formulations of ciprofloxacin. Antimicrob. Agents Chemother. **28**:74–77.
7. **Eliopoulos, G. M., A. Gardella, and R. C. Moellering, Jr.** 1984. In vitro activity of ciprofloxacin, a new carboxyquinoline antimicrobial agent. Antimicrob. Agents Chemother. **25**:331–335.
8. **Eron, L. J., L. Harvey, D. L. Hixon, and D. M. Poretz.** 1985. Ciprofloxacin therapy of infections caused by *Pseudomonas*

aeruginosa and other resistant bacteria. Antimicrob. Agents Chemother. **28**:308–310.
9. **Fass, R. J.** 1983. In vitro activity of ciprofloxacin (Bay o 9867). Antimicrob. Agents Chemother. **24**:568–574.
10. **Gonzalez, M. A., F. Uribe, S. D. Moisen, A. P. Fuster, A. Selen, P. G. Welling, and B. Painter.** 1984. Multiple-dose pharmacokinetics and safety of ciprofloxacin in normal volunteers. Antimicrob. Agents Chemother. **26**:741–744.
11. **Gordin, F. M., C. J. Hackbarth, K. G. Scott, and M. A. Sande.** 1985. Activities of pefloxacin and ciprofloxacin in experimentally induced *Pseudomonas* pneumonia in neutropenic guinea pigs. Antimicrob. Agents Chemother. **27**:452–454.
12. **Greenblatt, D. J., and J. Koch-Weser.** 1975. Clinical pharmacokinetics. N. Engl. J. Med. **297**:702–705.
13. **Hamilton-Miller, J. M. T.** 1985. Rationalization of terminology and methodology in the study of antibiotic interaction. J. Antimicrob. Chemother. **15**:655–657.
14. **Höffken, G., H. Lode, C. Prinzing, K. Borner, and P. Koeppe.** 1985. Pharmacokinetics of ciprofloxacin after oral and parenteral administration. Antimicrob. Agents Chemother. **27**:375–379.
15. **King, A., K. Shannon, and I. Phillips.** 1985. The *in-vitro* activities of enoxacin and ofloxacin compared with that of ciprofloxacin. J. Antimicrob. Chemother. **15**:551–558.
16. **Kouno, K., M. Inoue, and S. Mitsuhashi.** 1983. In vitro and in vivo antibacterial activity of AT-2266. Antimicrob. Agents Chemother. **24**:78–84.
17. **Lockley, M. R., R. Wise, and J. Dent.** 1984. The pharmacokinetics and tissue penetration of ofloxacin. J. Antimicrob. Chemother. **14**:647–652.
18. **Nakamura, S., N. Kurobe, S. Kashimoto, T. Ohue, Y. Takase, and M. Shimizu.** 1983. Pharmacokinetics of AT-2266 administered orally to mice, rats, dogs, and monkeys. Antimicrob. Agents Chemother. **24**:54–60.
19. **Norden, C. W., and E. Shinners.** 1985. Ciprofloxacin as therapy for experimental osteomyelitis caused by *Pseudomonas aeruginosa*. J. Infect. Dis. **151**:291–294.
20. **Pennington, J. E., W. F. Hickey, L. L. Blackwood, and M. A. Arnaut.** 1981. Active immunization with lipopolysaccharide *Pseudomonas* antigen for chronic *Pseudomonas* bronchopneumonia in guinea pigs. J. Clin. Invest. **68**:1140–1148.
21. **Pennington, J. E., and C. E. Johnson.** 1982. Comparative activities of *N*-formimidoyl thienamycin, ticarcillin, and tobramycin against experimental *Pseudomonas aeruginosa* pneumonia. Antimicrob. Agents Chemother. **22**:406–408.
22. **Pennington, J. E., and J. J. Miler.** 1979. Evaluation of a new polyvalent *Pseudomonas* vaccine in respiratory infections. Infect. Immun. **25**:1029–1034.
23. **Pennington, J. E., J. B. Schiff, and C. E. Johnson.** 1984. Therapeutic lessons learned from pneumonia models, p. 61–69. *In* H. Lode, B. Kemmerich, and J. Klastersky (ed.), Current aspects of bacterial and non-bacterial pneumonias. Georg Thieme Verlag, Stuttgart, Federal Republic of Germany.
24. **Pennington, J. E., and R. M. Stone.** 1979. Comparison of antibiotic regimens for treatment of experimental pneumonia due to *Pseudomonas*. J. Infect. Dis. **140**:881–889.
25. **Ramirez, C. A., J. L. Bran, C. R. Mejia, and J. F. Garcia.** 1985. Open, prospective study of the clinical efficacy of ciprofloxacin. Antimicrob. Agents Chemother. **28**:128–132.
26. **Ritschel, W. A.** 1980. Handbook of basic pharmacokinetics, 2nd ed., p. 230–248. Drug Intelligence Publications, Hamilton, Ill.
27. **Rudin, J. E., C. W. Norden, and E. M. Shinners.** 1984. In vitro activity of ciprofloxacin against aerobic gram-negative bacteria. Antimicrob. Agents Chemother. **26**:597–598.
28. **Saito, A., K. Sawatari, Y. Fukuda, M. Nagasawa, H. Koga, A. Tomonaga, H. Nakazato, K. Fujita, Y. Shigeno, Y. Suzuyama, K. Yamaguchi, K. Izumikawa, and K. Hara.** 1985. Susceptibility of *Legionella pneumophila* to ofloxacin in vitro and in experimental *Legionella* pneumonia in guinea pigs. Antimicrob. Agents Chemother. **28**:15–20.
29. **Sato, K., Y. Matsuura, M. Inoue, T. Une, Y. Osada, H. Ogawa, and S. Mitsuhashi.** 1982. In vitro and in vivo activity of DL-8280, a new oxazine derivative. Antimicrob. Agents Chemother. **22**:548–553.
30. **Schiff, J. B., and J. E. Pennington.** 1984. Comparative efficacies of piperacillin, azlocillin, ticarcillin, aztreonam, and tobramycin against experimental *Pseudomonas aeruginosa* pneumonia. Antimicrob. Agents Chemother. **25**:49–52.
31. **Schiff, J. B., G. J. Small, and J. E. Pennington.** 1984. Comparative activities of ciprofloxacin, ticarcillin, and tobramycin against experimental *Pseudomonas aeruginosa* pneumonia. Antimicrob. Agents Chemother. **26**:1–4.
32. **Washington, J. A., II, and V. L. Sutter.** 1980. Dilution susceptibility test: agar and macro-broth dilution procedures, p. 453–458. *In* E. H. Lennette, A. Balows, W. J. Hausler, Jr., and J. P. Truant (ed.), Manual of clinical microbiology, 3rd ed. American Society for Microbiology, Washington, D.C.
33. **Wise, R., J. M. Andrews, and L. J. Edwards.** 1983. In vitro activity of Bay 09867, a new quinoline derivative, compared with those of other antimicrobial agents. Antimicrob. Agents Chemother. **23**:559–564.
34. **Wise, R., R. Lockley, J. Dent, and M. Webberly.** 1984. Pharmacokinetics and tissue penetration of enoxacin. Antimicrob. Agents Chemother. **26**:17–19.
35. **Wise, R., R. M. Lockley, M. Webberly, and J. Dent.** 1984. Pharmacokinetics of intravenously administered ciprofloxacin. Antimicrob. Agents Chemother. **26**:208–210.