ANTIMICROBIAL AGENTS AND CHEMOTHERAPY, Nov. 1999, p. 2806–2807 Vol. 43, No. 11
0066-4804/99/$04.00+0

In Vitro Activities of Gemifloxacin (SB 265805, LB20304) against Recent Clinical Isolates of *Chlamydia pneumoniae*

PATRICIA M. ROBLIN, TAMARA REZNIK, ANDREI KUTLIN, AND MARGARET R. HAMMERSCHLAG*

Division of Infectious Diseases, Department of Pediatrics, Health Science Center at Brooklyn, State University of New York, Brooklyn, New York 11203

Received 11 June 1999/Returned for modification 29 July 1999/Accepted 31 August 1999

We compared the in vitro activity of gemifloxacin, a new quinolone antibiotic, to the activities of levofloxacin, moxifloxacin, trovafloxacin, erythromycin, and doxycycline against 20 isolates of *Chlamydia pneumoniae*. Gemifloxacin was the most active quinolone tested, with a MIC at which 90% of the isolates are inhibited and a minimal bactericidal concentration at which 90% of strains tested are killed of 0.25 μg/ml, but this activity was less than those of doxycycline and erythromycin.

Chlamydia pneumoniae is a frequent cause of community-acquired respiratory tract infections, including pneumonia and bronchitis in adults and children (1, 4). Quinolones have attracted interest as a potential therapy for community-acquired respiratory tract infections because they are active against a wide range of pathogens responsible for these infections, such as *Mycoplasma pneumoniae*, *Streptococcus pneumoniae* (including penicillin-resistant strains), and *C. pneumoniae* (6). We compared the in vitro activity of gemifloxacin (SB 265805, LB20304), a novel C-7 pyrrolidine-substituted naphthyridone antibiotic, to the in vitro activities of levofloxacin, moxifloxacin (BAY 12-8039), trovafloxacin, erythromycin, and doxycycline against 20 isolates of *C. pneumoniae*, including recent clinical isolates.

Gemifloxacin (SB 265805, LB20304) (SmithKline Beecham, Collegeville, Pa.), levofloxacin (Ortho Pharmaceuticals, Raritan, N.J.), moxifloxacin (BAY 12-8039) (Bayer Corp., West Haven, Conn.), trovafloxacin (Pfizer, New York, N.Y.), erythromycin, and doxycycline were supplied as powders and solubilized according to instructions from the manufacturers. Twenty isolates of *C. pneumoniae* were tested: TW 183 (Washington Research Foundation, Seattle, Wash.), CM-1 (ATCC 1360), J21 (from Japan; ATCC VR1435), W6805 and T2219 (isolated from two patients with pneumonia from Wisconsin and Brooklyn, N.Y., respectively), and 15 recent clinical isolates from adults enrolled in a U.S. multicenter community-acquired pneumonia treatment study. This study was conducted in 20 states during 1997 and 1998. All isolates were obtained prior to patient treatment.

Susceptibility testing of *C. pneumoniae* was performed in cell culture using HEp-2 cells grown in 96-well microtiter plates (12, 13). Each well was inoculated with 0.1 ml of the test strain diluted to yield 10^3 to 10^4 inclusion-forming units per ml, was centrifuged at 1,700 × *g* for 1 h, and was incubated at 35°C for 1 h. Wells were then aspirated and overlaid with 0.2 ml of Iscove's minimal essential medium containing 1 μg of cycloheximide per ml and serial twofold dilutions of the test drug. After incubation at 35°C for 72 h, cultures were fixed and stained for inclusions with fluorescein-conjugated antibody to the lipopolysaccharide genus antigen (Pathfinder; Kallestad Diagnostics, Chaska, Minn.). The MIC was considered to be the lowest antibiotic concentration at which no inclusions were seen. The minimal bactericidal concentration (MBC) was determined by aspirating the antibiotic-containing medium and then washing wells twice with phosphate-buffered saline and adding antibiotic-free medium. Cultures were frozen at −70°C, thawed, passed onto new cells and incubated for 72 h and then fixed and stained as described above. The MBC was considered to be the lowest antibiotic concentration which resulted in no inclusions after passage. All tests were run in triplicate.

The MICs and MBCs for *C. pneumoniae* are given in Table 1. The MIC at which 90% of the isolates are inhibited (MIC_{90}) and MBC at which 90% of strains tested are killed (MBC_{90}) of gemifloxacin were 0.25 μg/ml, compared to 1 μg/ml for the other quinolones tested. Gemifloxacin was less active than erythromycin and doxycycline, which both displayed MIC_{90}s and MBC_{90}s of 0.06 μg/ml.

Gemifloxacin was the most active quinolone antibiotic tested. The MIC obtained for gemifloxacin against *C. pneumoniae* in the present study were very consistent from isolate to isolate, especially in view of the wide geographic distribution of the isolates tested. Gemifloxacin has been demonstrated to have a broad spectrum with excellent activity against the most common bacterial pathogens causing respiratory infection: *S. pneumoniae* (including penicillin- and ciprofloxacin-resistant isolates), *Haemophilus influenzae*, and *Moraxella catarrhalis* (2, 8, 9). Gemifloxacin also has excellent in vitro activity against *Legionella* species and *M. pneumoniae* (3, 7, 10). Similar to other quinolones, gemifloxacin is concentrated in tissue and macrophages at concentrations 17- to 28-fold higher than in serum (3).

Data on the activity of gemifloxacin against *C. pneumoniae* are limited, in part due to the relatively small number of clinical isolates that have been available for testing. Ridgway et al. (10) tested five isolates of *C. pneumoniae*: TW 183, IOL 207, VR 1355 (2043), TW 27.9, and VR 1310 (CWL-029). The MIC range was 0.06 to 0.12 μg/ml, which is one to two dilutions lower than the results we obtained. This discrepancy may be within the standard error of the test or secondary to the methods used. Ridgway et al. used McCoy cells, which are 10- to 100-fold less susceptible to *C. pneumoniae* infection than HEp-2 cells, possibly leading to lower endpoints (11). The activities of the other quinolones tested in this study, levofloxacin, moxifloxacin, and trovafloxacin, were the same as previously reported, using different selections of isolates (5, 12, 13).

* Corresponding author. Mailing address: Department of Pediatrics, Box 49, SUNY Health Science Center at Brooklyn, 450 Clarkson Ave., Brooklyn, NY 11203-2098. Phone: (718) 245-4075. Fax: (718) 245-2118. E-mail: mhammerschlag@pol.net.

TABLE 1. Activities of gemifloxacin and other antibiotics against 20 isolates of *C. pneumoniae*

Antimicrobial agent	MIC (μg/ml)[a]			MBC (μg/ml)[b]	
	Range	50%	90%	Range	90%
Gemifloxacin	0.125–0.25	0.25	0.25	0.125–0.25	0.25
Levofloxacin	0.25–1	0.5	1	0.25–1	1
Moxifloxacin	0.125–1	0.5	1	0.125–1	1
Trovafloxacin	0.5–1	0.5	1	0.5–1	1
Erythromycin	0.008–0.06	0.06	0.06	0.008–0.06	0.06
Doxycycline	0.016–0.06	0.03	0.06	0.016–0.06	0.06

[a] 50% and 90%, MIC_{50} and MIC_{90}, respectively.
[b] 90%, MBC_{90}.

The broad-spectrum activity of gemifloxacin suggests that it may have a role in the treatment of respiratory infections. However, in vitro activity and good tissue penetration may not always predict microbiologic efficacy in vivo, especially for *C. pneumoniae* (1, 6). The use of gemifloxacin in the treatment of *C. pneumoniae* infections will need to be confirmed in clinical studies using culture to assess microbiologic efficacy.

REFERENCES

1. **Block, S., J. Hedrick, M. R. Hammerschlag, G. H. Cassell, and J. C. Craft.** 1995. *Mycoplasma pneumoniae* and *Chlamydia pneumoniae* in community acquired pneumonia in children: comparative safety and efficacy of clarithromycin and erythromycin suspensions. Pediatr. Infect. Dis. J. **14:**471–477.
2. **Cormican, M. G., and R. N. Jones.** 1997. Antimicrobial activity and spectrum of LB20304, a novel fluoronaphthyridone. Antimicrob. Agents Chemother. **41:**204–211.
3. **Critchley, I. A., J. Broskey, and K. Coleman.** 1998. SB-265805 (LB20304a): in vitro activity, intracellular accumulation and killing of *Legionella pneumophila* in human macrophage, abstr. F-100, p. 257. *In* Abstracts of the 38th Interscience Conference on Antimicrobial Agents and Chemotherapy. American Society for Microbiology, Washington, D.C.
4. **Grayston, J. T., L. A. Campbell, C.-C. Kuo, C. H. Mordhorst, P. Saikku, D. H. Thom, and S. P. Wang.** 1990. A new respiratory tract pathogen: *Chlamydia pneumoniae* strain TWAR. J. Infect. Dis. **161:**618–625.
5. **Hammerschlag, M. R., K. K. Qumei, and P. M. Roblin.** 1992. In vitro activity of azithromycin, clarithromycin, L-ofloxacin, and other antibiotics against *Chlamydia pneumoniae*. Antimicrob. Agents Chemother. **36:**1573–1574.
6. **Hammerschlag, M. R.** Community-acquired pneumonia due to atypical organisms in adults: diagnosis and treatment. Infect. Dis. Clin. Pract., in press.
7. **Hannan, P., and G. Woodnutt.** 1998. SB-265805 (LB20304a): susceptibility of human mycoplasmas in vitro, abstr. F-101, p. 257. *In* Abstracts of the 38th Interscience Conference on Antimicrobial Agents and Chemotherapy. American Society for Microbiology, Washington, D.C.
8. **Moore, T., N. Niconovich, and K. Coleman.** 1998. SB-265805 (LB20304a): in vitro antibacterial activity against the common respiratory tract pathogens *Streptococcus pneumoniae*, *Haemophilus influenzae*, and *Moraxella catarrhalis*, abstr. F-98, p. 257. *In* Abstracts of the 38th Interscience Conference on Antimicrobial Agents and Chemotherapy. American Society for Microbiology, Washington, D.C.
9. **Oh, J.-I., K.-S. Paek, M.-J. Ahn, M.-Y. Kim, C. H. Hong, I.-C. Kim, and J.-H. Kwak.** 1996. In vitro and in vivo evaluation of LB20304, a new fluoronaphthyridone. Antimicrob. Agents Chemother. **40:**1564–1568.
10. **Ridgway, G. L., N. Salman, S. Clark, I. Mathias, and D. Felmingham.** 1998. SB-265805 (LB20304a): comparative in vitro activity against *Legionella pneumophila* and *Chlamydia* spp., abstr. F-97, p. 256. *In* Abstracts of the 38th Interscience Conference on Antimicrobial Agents and Chemotherapy. American Society for Microbiology, Washington, D.C.
11. **Roblin, P. M., W. Dumornay, and M. R. Hammerschlag.** 1992. Use of HEp-2 cells for improved isolation and passage of *Chlamydia pneumoniae*. J. Clin. Microbiol. **30:**1968–1971.
12. **Roblin, P. M., A. Kutlin, and M. R. Hammerschlag.** 1997. In vitro activity of trovafloxacin against *Chlamydia pneumoniae*. Antimicrob. Agents Chemother. **41:**2033–2034.
13. **Roblin, P. M., and M. R. Hammerschlag.** 1998. In vitro activity of a new 8-methoxyquinolone, BAY 12-8039, against *Chlamydia pneumoniae*. Antimicrob. Agents Chemother. **42:**951–952.