

## Comparison of the In Vitro Activities of Bay 12-8039, a New Quinolone, and Other Antimicrobials against Clinically Important Anaerobes

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Received 12 July 1996/Returned for modification 10 October 1996/Accepted 2 January 1997

**Bay 12-8039, a new 8-methoxy quinolone, was compared with other agents for activity against clinically relevant anaerobes. Bay 12-8039 inhibited 91 and 96% of the 410 test isolates at 2 and 4  $\mu\text{g/ml}$ , respectively. Bay 12-8039 had activity comparable to that of metronidazole and overall was at least 16-fold more active than ciprofloxacin, ofloxacin, and cefoxitin, 32-fold more active than cefotetan, and at least 128-fold more active than penicillin G.**

The role of anaerobic bacteria in human infections is well established (5). These organisms contribute to abscess formation, produce enzymes which may contribute to the spread of the infection, release toxins that can produce extensive tissue damage, and have been isolated from patients with fatal bacteremia (3). Of concern with regard to these anaerobes is their increasing resistance to a variety of antimicrobial agents, particularly  $\beta$ -lactam agents. The majority of  $\beta$ -lactamases produced by anaerobes possess cephalosporinase activity, while others hydrolyze penicillins and carbapenems (1, 4, 11). A variety of species of anaerobes has been shown to produce  $\beta$ -lactamases, including the *Bacteroides fragilis* group, *Prevotella* spp., *Porphyromonas* spp., *Fusobacterium* spp., and *Clostridium* spp. (1, 9). Resistance to clindamycin has also increased sharply in the last decade (2). Thus, other classes of antimicrobials with antianaerobic activity are needed.

The newly developed fluoroquinolones, such as norfloxacin, ciprofloxacin, and ofloxacin, have exhibited broad-spectrum activity against aerobic gram-negative bacilli, including strains multiresistant to a variety of antimicrobial agents. However, the activity against anaerobic bacteria has remained poor by comparison, although newer, experimental fluoroquinolones such as WIN 57273 and DU-6859a have extended antianaerobe activity (6, 13). This study compares the in vitro activity of Bay 12-8039, a new 8-methoxy quinolone (10), to those of other antimicrobial agents against clinically significant anaerobes.

(This research was presented at the 36th Interscience Conference on Antimicrobial Agents and Chemotherapy, New Orleans, La., 15 to 18 September 1996.)

A total of 410 nonduplicate, clinical isolates were tested, and the group of isolates was comprised of the following: *Bacteroides fragilis* group, 249 strains; *Prevotella bivia*, 21 strains; *Prevotella disiens*, 19 strains; *Fusobacterium* spp., 23 strains; *Clostridium* spp., 40 strains; *Eubacterium* spp., 14 strains; *Peptostreptococcus* spp., 25 strains; and *Veillonella parvula*, 18 strains. All isolates were identified with selective media, biochemical profiles, and gas-liquid chromatography (7, 12).

Each of the following antimicrobials was provided by the manufacturer: Bay 12-8039 and ciprofloxacin (Bayer Corp.),

ofloxacin (Ortho-McNeil Pharmaceuticals), cefoxitin (Merck & Co.), cefotetan (Zeneca Pharmaceuticals), clindamycin (The Upjohn Co.), metronidazole (G. D. Searle), and penicillin G (Pfizer Pharmaceuticals). All laboratory standard powders were stored at  $-20^{\circ}\text{C}$  until used.

Susceptibility testing was performed with each strain by a National Committee for Clinical Laboratory Standards recommended broth microdilution method (8). Serial twofold dilutions of each antimicrobial were prepared in Anaerobe broth MIC (Difco) within a dilution scheme of 0.015 to 128  $\mu\text{g/ml}$ . For fastidious strains, 3% lysed horse blood was added to the test medium. The inoculum was prepared by suspension of growth from anaerobic sheep blood agar plates to a density equal to that of a no. 1 McFarland standard and further diluted to give a final inoculum size of  $10^5$  CFU/100- $\mu\text{l}$  well. All susceptibility plates were incubated at  $35^{\circ}\text{C}$  anaerobically for 48 h and then read. The MIC was defined as the lowest concentration of each antimicrobial that inhibited the visible growth of the test isolate. Quality assurance testing was performed with *Bacteroides fragilis* ATCC 25285, *Bacteroides thetaiotaomicron* ATCC 29741, *Eubacterium lentum* ATCC 43055, and *Clostridium perfringens* ATCC 13124.

Figure 1 compares the distributions of MICs of the three

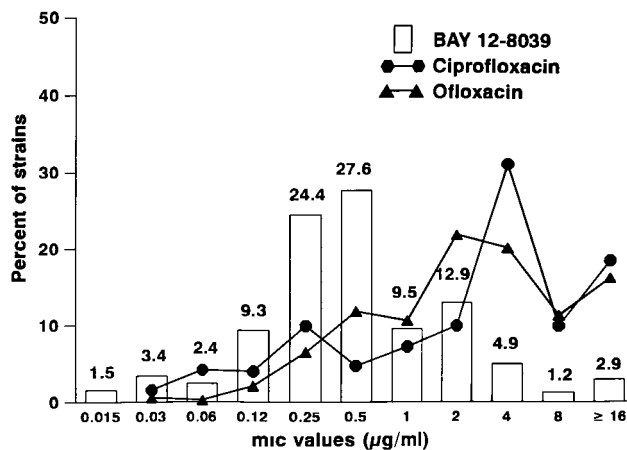


FIG. 1. Comparison of the distributions of the MICs of Bay 12-8039, ciprofloxacin, and ofloxacin for 410 clinical anaerobe isolates.

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TABLE 1. Comparison of the in vitro activities of Bay 12-8039 and other antimicrobials against clinical strains of anaerobes

Organism (no. of strains tested) and antimicrobial	MIC ( $\mu\text{g/ml}$ ) <sup>a</sup>		
	Range	50%	90%
<b>All anaerobes (410)</b>			
Bay 12-8039	0.015– $\geq 32$	0.5	2
Ciprofloxacin	0.03– $\geq 32$	4	$\geq 32$
Ofloxacin	0.03– $\geq 32$	2	$\geq 32$
Cefoxitin	0.06–128	4	32
Cefotetan	0.06– $\geq 256$	4	64
Clindamycin	0.12–64	0.5	4
Metronidazole	0.12–64	0.5	1
Penicillin G	0.06– $\geq 256$	8	$\geq 256$
<b><i>Bacteroides fragilis</i> (126)</b>			
Bay 12-8039	0.12–16	0.5	2
Ciprofloxacin	0.25–16	4	16
Ofloxacin	0.5–16	4	16
Cefoxitin	0.5–128	4	32
Cefotetan	0.25–128	4	32
Clindamycin	0.015–16	0.25	2
Metronidazole	0.12–1	0.5	1
Penicillin G	4–128	8	128
<b><i>Bacteroides thetaiotaomicron</i> (35)</b>			
Bay 12-8039	0.25– $\geq 32$	2	16
Ciprofloxacin	1– $\geq 32$	8	$\geq 32$
Ofloxacin	1– $\geq 32$	16	$\geq 32$
Cefoxitin	0.5–64	16	32
Cefotetan	1– $\geq 256$	32	64
Clindamycin	0.015– $\geq 32$	2	$\geq 32$
Metronidazole	0.12–1	0.5	1
Penicillin G	2– $\geq 256$	16	$\geq 256$
<b><i>Bacteroides distasonis</i> (25)</b>			
Bay 12-8039	0.12–4	0.5	2
Ciprofloxacin	0.5– $\geq 32$	4	8
Ofloxacin	0.5–16	2	16
Cefoxitin	2–128	16	32
Cefotetan	2– $\geq 32$	32	128
Clindamycin	0.015– $\geq 32$	0.5	16
Metronidazole	0.12–1	0.5	1
Penicillin G	4– $\geq 256$	32	128
<b><i>Bacteroides ovatus</i> (25)</b>			
Bay 12-8039	0.25– $\geq 32$	2	4
Ciprofloxacin	4– $\geq 32$	16	$\geq 32$
Ofloxacin	2– $\geq 32$	8	16
Cefoxitin	1–128	16	32
Cefotetan	2– $\geq 256$	32	128
Clindamycin	0.015– $\geq 32$	1	$\geq 32$
Metronidazole	0.25–1	1	1
Penicillin G	0.25– $\geq 256$	16	$\geq 256$
<b><i>Bacteroides vulgatus</i> (25)</b>			
Bay 12-8039	0.25– $\geq 16$	0.5	2
Ciprofloxacin	4– $\geq 32$	8	$\geq 32$
Ofloxacin	1– $\geq 32$	4	16
Cefoxitin	0.25–128	2	32
Cefotetan	0.25–128	4	64
Clindamycin	0.015– $\geq 32$	0.12	$\geq 32$
Metronidazole	0.12–1	0.25	1
Penicillin G	0.25– $\geq 256$	32	128
<b><i>Bacteroides uniformis</i> (14)</b>			
Bay 12-8039	0.25– $\geq 32$	2	16
Ciprofloxacin	2– $\geq 32$	$\geq 32$	$\geq 32$
Ofloxacin	2– $\geq 32$	8	$\geq 32$
Cefoxitin	0.5–32	1	32
Cefotetan	2–64	4	64
Clindamycin	0.015– $\geq 32$	0.5	2
Metronidazole	0.12–1	0.5	0.5
Penicillin G	2– $\geq 256$	8	$\geq 256$

Continued

TABLE 1—Continued

Organism (no. of strains tested) and antimicrobial	MIC ( $\mu\text{g/ml}$ ) <sup>a</sup>		
	Range	50%	90%
<b><i>Prevotella bivia</i> (21)</b>			
Bay 12-8039	0.25–4	1	2
Ciprofloxacin	0.5–16	4	16
Ofloxacin	0.25–16	4	8
Cefoxitin	0.12–4	0.5	4
Cefotetan	0.12–16	2	8
Clindamycin	0.015–0.12	0.015	0.03
Metronidazole	0.25–4	1	2
Penicillin G	0.06–32	2	16
<b><i>Prevotella disiens</i> (19)</b>			
Bay 12-8039	0.12–4	0.5	0.5
Ciprofloxacin	0.25–16	1	1
Ofloxacin	0.5–16	1	2
Cefoxitin	0.12–4	0.5	2
Cefotetan	0.12–8	2	4
Clindamycin	0.015–0.06	0.015	0.03
Metronidazole	0.5–2	1	2
Penicillin G	0.06–16	1	16
<b><i>Fusobacterium</i> spp. (23)<sup>b</sup></b>			
Bay 12-8039	0.03–1	0.12	0.5
Ciprofloxacin	0.06–8	0.5	4
Ofloxacin	0.25–4	1	4
Cefoxitin	0.06–4	0.06	4
Cefotetan	0.06–8	0.06	4
Clindamycin	0.015– $\geq 32$	0.03	1
Metronidazole	0.12–1	0.25	1
Penicillin G	0.06–64	0.06	16
<b><i>Clostridium perfringens</i> (20)</b>			
Bay 12-8039	0.12–0.5	0.25	0.5
Ciprofloxacin	0.06–0.5	0.25	0.5
Ofloxacin	0.12–1	0.5	1
Cefoxitin	0.25–2	0.5	1
Cefotetan	0.06–8	0.06	0.5
Clindamycin	0.03–8	0.5	2
Metronidazole	0.12–64	1	2
Penicillin G	0.06–0.12	0.06	0.12
<b><i>Clostridium</i> spp. (20)<sup>c</sup></b>			
Bay 12-8039	0.12–4	0.25	1
Ciprofloxacin	0.06–16	1	4
Ofloxacin	0.25–16	0.5	8
Cefoxitin	0.06–128	4	64
Cefotetan	0.06–128	4	128
Clindamycin	0.015–8	0.12	4
Metronidazole	0.12–64	0.5	4
Penicillin G	0.06–128	0.25	32
<b><i>Eubacterium</i> spp. (14)<sup>d</sup></b>			
Bay 12-8039	0.015–0.5	0.12	0.25
Ciprofloxacin	0.06–2	0.25	1
Ofloxacin	0.12–2	0.5	1
Cefoxitin	0.12–8	4	8
Cefotetan	0.12–32	16	32
Clindamycin	0.015–16	0.25	2
Metronidazole	0.12–8	0.25	0.25
Penicillin G	0.06–1	0.5	1
<b><i>Peptostreptococcus</i> spp. (25)<sup>e</sup></b>			
Bay 12-8039	0.015–4	0.06	0.25
Ciprofloxacin	0.03–8	0.12	0.5
Ofloxacin	0.03–16	0.25	1
Cefoxitin	0.06–4	0.12	0.5
Cefotetan	0.06–2	0.25	1
Clindamycin	0.015–1	0.03	0.25
Metronidazole	0.12–1	0.12	0.5
Penicillin G	0.06–0.12	0.06	0.06

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TABLE 1—Continued

Organism (no. of strains tested) and antimicrobial	MIC ( $\mu\text{g/ml}$ ) <sup>a</sup>		
	Range	50%	90%
<i>Veillonella parvula</i> (18)			
Bay 12-8039	0.015–1	0.06	0.25
Ciprofloxacin	0.03–2	0.06	0.5
Ofloxacin	0.12–4	0.25	1
Cefoxitin	0.06–4	0.25	1
Cefotetan	0.06–2	0.5	1
Clindamycin	0.015–1	0.015	0.5
Metronidazole	0.25–1	0.5	1
Penicillin G	0.06–4	0.12	2

<sup>a</sup> 50% and 90% MIC<sub>50</sub> and MIC<sub>90</sub>, respectively.

<sup>b</sup> *Fusobacterium* spp. consist of *F. nucleatum* (10 strains), *F. necrophorum* (2 strains), and *Fusobacterium* spp. (11 strains).

<sup>c</sup> *Clostridium* spp. consist of *C. septicum* (one strain), *C. butyricum* (three strains), *C. ramosum* (three strains), *C. innocuum* (one strain), *C. sphenoides* (two strains), *C. sporogenes* (three strains), *C. clostridioforme* (one strain), *C. cadaveris* (one strain), *C. difficile* (one strain), and *Clostridium* spp. (four strains).

<sup>d</sup> *Eubacterium* spp. consist of *E. lentum* (12 strains), *E. limosum* (1 strain), and *E. alactolyticum* (1 strain).

<sup>e</sup> *Peptostreptococcus* spp. consist of *P. asaccharolyticus* (5 strains) and *Peptostreptococcus* spp. (20 strains).

fluoroquinolones tested for all the anaerobic isolates. Bay 12-8039 had a mode MIC of 0.5  $\mu\text{g/ml}$  compared to 2  $\mu\text{g/ml}$  for ofloxacin and 4  $\mu\text{g/ml}$  for ciprofloxacin. Table 1 compares the in vitro activities of Bay 12-8039 and the other antimicrobials. The overall activity of Bay 12-8039 was good against all the anaerobes tested, with a MIC at which 50% of the isolates are inhibited (MIC<sub>50</sub>) of 0.5  $\mu\text{g/ml}$  and a MIC<sub>90</sub> of 2  $\mu\text{g/ml}$ , and 96% of all strains were inhibited at 4  $\mu\text{g/ml}$ . The in vitro activity of Bay 12-8039 was comparable overall to that of metronidazole. Only 17 strains of the *B. fragilis* group had Bay 12-8039 MICs of  $\geq 8 \mu\text{g/ml}$ , whereas all other anaerobe groups were inhibited by  $\leq 4 \mu\text{g}$  of Bay 12-8039 per ml. In a comparison of the activities (MIC<sub>90</sub>) of the agents, Bay 12-8039 was 2- to 16-fold more active than both ciprofloxacin and ofloxacin against *B. fragilis* group strains, 2- to 8-fold more active against *Prevotella* spp., and 2- to 8-fold more active against strains of *Fusobacterium*, *Clostridium*, *Eubacterium*, *Peptostreptococcus*, and *V. parvula* than were ciprofloxacin and ofloxacin. Compared to cefoxitin and cefotetan, Bay 12-8039 was 2- to 16-fold and 4- to 64-fold more active, respectively, against the *B. fragilis* group species, while Bay 12-8039 was 16- to  $\geq 64$ -fold more active than penicillin G against the same strains. Bay 12-8039 was 2- to 64-fold more active than cefoxitin and 4- to 128-fold more active than cefotetan against the remaining groups of anaerobes. Bay 12-8039 exhibited good activity against strains of *Clostridium* and *Peptostreptococcus*; however, penicillin G was the most active antimicrobial tested. Compared to clindamycin, Bay 12-8039 was slightly more active overall; however, exceptions included clindamycin, which was more active than Bay 12-8039 against strains of *Bacteroides uniformis* and *Prevotella*, although all strains of *Prevotella* were inhibited at  $\leq 4 \mu\text{g}$  of Bay 12-8039 per ml. The following values were noted for

Bay 12-8039 when the quality control isolates were tested in 10 separate susceptibility runs: *B. fragilis* ATCC 25285, 0.12 to 0.25  $\mu\text{g/ml}$ ; *B. thetaiotaomicron* ATCC 29741, 2  $\mu\text{g/ml}$ ; *E. lentum* ATCC 43055, 0.12 to 0.25  $\mu\text{g/ml}$ ; and *C. perfringens* ATCC 13124, 0.25  $\mu\text{g/ml}$ .

Wise et al. (14) recently reported increased activity of Bay 12-8039 compared with those of other fluoroquinolones against aerobes, including members of the family *Enterobacteriaceae*, *Pseudomonas* spp., and staphylococci. In addition, in combination with the present antianaerobe data, it can be suggested that Bay 12-8039 may be useful in mixed aerobe-anaerobe infections. However, firm conclusions in this regard await the results of pharmacologic, animal model, and human safety studies.

This study was supported in part by a grant from Bayer Corporation.

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