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In Vitro Activity of OPC-17116

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The in vitro activity of OPC-17116, a new C-5 methyl fluoroquinolone, was compared with the activities of other fluoroquinolones. OPC-17116 inhibited 50% of the members of the family *Enterobacteriaceae* tested and 90% of *Haemophilus influenzae*, *Neisseria* species, and *Moraxella catarrhalis* isolates at ≤0.25 μg/ml. At ≤2 μg/ml, 90% of the *Enterobacteriaceae* were inhibited, which was comparable to or better than the activities of fleroxacin, ofloxacin, and lomefloxacin but less than the activity of ciprofloxacin. OPC-17116 inhibited 90% of the staphylococci tested at ≤0.25 μg/ml, but it did not inhibit methicillin-resistant, ciprofloxacin-resistant *Staphylococcus aureus* or *Staphylococcus epidermidis*. Group A, B, C, F, and G streptococci and *Streptococcus pneumoniae* were inhibited by ≤0.5 μg/ml, being four-fold more active than ciprofloxacin and ofloxacin. Tosufloxacin was the most active agent tested against gram-positive cocci. OPC-17116 inhibited *Bacteroides fragilis* at 4 μg/ml. There was a minimal effect of inoculum size on MIC, and the MBCs were within 1 dilution of the MICs. The activity of OPC-17116 was decreased at pH 6 and in the presence of high Mg^{2+} concentrations, but it was unaffected by human serum. OPC-17116 showed a postantibiotic effect against *Pseudomonas aeruginosa* and *Staphylococcus aureus* similar to the postantibiotic effects reported for other fluoroquinolones. The frequency of spontaneous single-step resistance was low ($<10^{-9}$), but repeated passage of organisms in the presence of OPC-17116 resulted in the selection of resistant isolates.

There is continued interest in the development of new fluoroquinolone antimicrobial agents in order to improve antibacterial activity, overcome bacterial resistance, or improve the pharmacokinetics of these agents (15). OPC-17116 is a new fluoroquinolone which contains an N-1 cyclopropyl group, a C-5 methyl group, and a C-7 piperazinyl moiety to which a 3-methyl group is attached (Fig. 1).

We investigated the antibacterial activity of OPC-17116 and compared its activity with those of other fluoroquinolones. We also determined the effect of various assay conditions on the in vitro activity of OPC-17116.

MATERIALS AND METHODS

OPC-17116 was provided by Otsuka Pharmaceutical Co., Ltd., Osaka, Japan. Other agents were obtained from their respective manufacturers. Quinolones were prepared fresh daily by standard procedures, and all quinolones were tested simultaneously.

Bacterial isolates were obtained from patients who were hospitalized in the Columbia University hospital system. Only one isolate from each patient was tested to avoid testing of multiple copies of the same strain. Some of the isolates used in this study were collected during a clinical evaluation of new quinolones and other antimicrobial agents.

Antimicrobial susceptibilities of aerobic species were measured by an agar dilution method with Mueller-Hinton agar according to the guidelines of the National Committee for Clinical Laboratory Standards (10). Activity against streptococcal species was determined with Mueller-Hinton agar supplemented with 5% sheep blood. Activity against *Haemophilus*, *Moraxella*, and *Neisseria* species was determined with *Haemophilus* test medium (9). Activity against anaerobic species was determined with Wilkins-Chalgren medium (11). For aerobic species, a replicating spot device was used to apply 10^4 CFU to agar plates. An inoculum of 10^5 CFU was used for anaerobic species. Incubation took place at 35°C for 18 to 20 h; however, anaerobic organisms were incubated for 48 h in GasPak jars (BBL Microbiology Systems). Broth dilutions were performed with 5×10^5 CFU in tubes with a 1-ml volume. MBCs were determined by the method of Pearson et al. (13) by transferring 0.01 ml to antibiotic-free plates to yield a 99.9% reduction in CFU from the original count. The effect of serum, pH, and ion changes in the medium were determined as described previously (7).

Selection of resistant organisms was done by inoculating 5×10^5 CFU of an organism into Mueller-Hinton broth containing twofold increasing concentrations of the compound and transferring organisms from the highest concentration showing growth daily for 15 consecutive days.

Spontaneous single-step resistance to the compound was detected by plating an overnight culture, which was concentrated by centrifugation onto agar medium containing the compound at concentrations that were four and eight times the MIC. The actual number of colonies applied was determined by performing viability counts on drug-free medium.

The postantibiotic effect (PAE) was determined by previously published methods (2, 3).

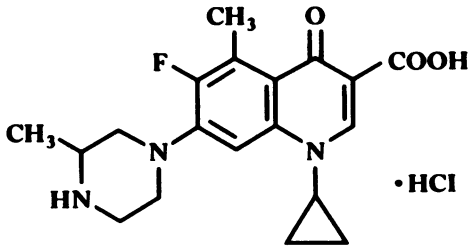

FIG. 1. Chemical structure of OPC-17116 [(±)-1-cyclopropyl-6-fluoro-1,4-dihydro-5-methyl-7-(3-methyl-1-piperazinyl)-carboxylic acid hydrochloride].

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TABLE 1. Activities of OPC-17116 compared with those of other quinolones

Organism (no. tested)	Antimicrobial agent	MIC (μg/ml)[a]		
		Range	50%	90%
Escherichia coli (30)	OPC-17116	0.015–0.12	0.06	0.12
	Ciprofloxacin	0.008–0.06	0.015	0.06
	Fleroxacin	0.015–0.25	0.06	0.12
	Ofloxacin	0.03–0.25	0.06	0.12
	Lomefloxacin	0.06–0.25	0.06	0.12
Enterobacter cloacae (20)	OPC-17116	0.06–0.25	0.06	0.25
	Ciprofloxacin	0.03–0.12	0.06	0.12
	Fleroxacin	0.06–0.5	0.06	0.25
	Ofloxacin	0.06–1	0.12	1
	Lomefloxacin	0.12–2	0.12	2
Enterobacter aerogenes (10)	OPC-17116	0.015–0.5	0.06	0.5
	Ciprofloxacin	0.015–0.25	0.015	0.12
	Fleroxacin	0.03–1	0.06	0.5
	Ofloxacin	0.03–1	0.12	0.5
	Lomefloxacin	0.06–2	0.12	0.5
Enterobacter agglomerans (10)	OPC-17116	0.008–0.12	0.03	0.12
	Ciprofloxacin	0.008–0.12	0.03	0.12
	Fleroxacin	0.015–0.5	0.06	0.12
	Ofloxacin	0.015–1	0.12	0.5
	Lomefloxacin	0.03–2	0.12	0.5
Klebsiella pneumoniae (30)	OPC-17116	0.06–0.5	0.12	0.25
	Ciprofloxacin	0.008–0.5	0.06	0.25
	Fleroxacin	0.06–2	0.25	1
	Ofloxacin	0.12–1	0.25	1
	Lomefloxacin	0.12–2	0.25	2
Klebsiella oxytoca (20)	OPC-17116	0.008–0.25	0.06	0.25
	Ciprofloxacin	0.008–0.12	0.015	0.03
	Fleroxacin	0.015–0.25	0.06	0.12
	Ofloxacin	0.03–0.5	0.06	0.25
	Lomefloxacin	0.06–0.5	0.06	0.25
Proteus mirabilis (30)	OPC-17116	0.015–0.5	0.25	0.25
	Ciprofloxacin	0.015–0.03	0.015	0.03
	Fleroxacin	0.06–0.5	0.06	0.12
	Ofloxacin	0.03–0.25	0.06	0.12
	Lomefloxacin	0.06–0.25	0.12	0.25
Proteus vulgaris (17)	OPC-17116	0.015–0.25	0.25	0.25
	Ciprofloxacin	0.008–0.12	0.015	0.06
	Fleroxacin	0.06–0.5	0.25	0.25
	Ofloxacin	0.06–0.25	0.12	0.25
	Lomefloxacin	0.06–0.25	0.12	0.25
Providencia rettgeri (15)	OPC-17116	0.06–2	0.25	2
	Ciprofloxacin	0.03–1	0.06	0.5
	Fleroxacin	0.06–1	0.12	1
	Ofloxacin	0.06–2	0.25	2
	Lomefloxacin	0.12–2	0.25	2
Providencia stuartii (15)	OPC-17116	0.06–2	0.25	2
	Ciprofloxacin	0.008–1	0.12	1
	Fleroxacin	0.06–2	0.5	2
	Ofloxacin	0.06–2	0.5	2
	Lomefloxacin	0.06–2	0.5	2
Morganella morganii (15)	OPC-17116	0.06–0.5	0.12	0.5
	Ciprofloxacin	0.008–0.06	0.015	0.03
	Fleroxacin	0.03–0.25	0.25	0.25
	Ofloxacin	0.03–0.25	0.12	0.5
	Lomefloxacin	0.03–0.25	0.12	0.5
Serratia marcescens (30)	OPC-17116	0.03–2	0.25	1
	Ciprofloxacin	0.015–2	0.03	0.5
	Fleroxacin	0.06–2	0.12	1
	Ofloxacin	0.12–2	0.25	1
	Lomefloxacin	0.12–2	0.25	1
Citrobacter freundii (30)	OPC-17116	0.015–1	0.06	0.5
	Ciprofloxacin	0.008–1	0.015	0.12
	Fleroxacin	0.015–1	0.03	1
	Ofloxacin	0.03–1	0.03	1
	Lomefloxacin	0.03–1	0.12	1
Citrobacter diversus (20)	OPC-17116	0.008–0.25	0.03	0.06
	Ciprofloxacin	0.008–0.03	0.008	0.015
	Fleroxacin	0.008–0.03	0.03	0.03
	Ofloxacin	0.008–0.25	0.03	0.25
	Lomefloxacin	0.015–1	0.03	0.25
Salmonella spp. (15)	OPC-17116	0.015–0.12	0.06	0.12
	Ciprofloxacin	0.008–0.03	0.008	0.03
	Fleroxacin	0.015–0.12	0.06	0.12
	Ofloxacin	0.015–0.12	0.06	0.12
	Lomefloxacin	0.015–0.25	0.12	0.25
Shigella spp. (16)	OPC-17116	0.008–0.12	0.03	0.12
	Ciprofloxacin	0.008–0.06	0.015	0.03
	Fleroxacin	0.015–0.12	0.06	0.12
	Ofloxacin	0.015–0.12	0.12	0.12
	Lomefloxacin	0.06–0.12	0.12	0.12
Yersinia enterocolitica (10)	OPC-17116	0.008–0.05	0.06	0.25
	Ciprofloxacin	0.008–0.12	0.03	0.06
	Fleroxacin	0.008–1	0.12	0.25
	Ofloxacin	0.008–0.25	0.12	0.25
	Lomefloxacin	0.008–0.25	0.12	0.25
Pseudomonas aeruginosa (40)	OPC-17116	0.25–16	0.5	4
	Ciprofloxacin	0.12–16	0.25	1
	Fleroxacin	0.25–16	2	4
	Ofloxacin	0.25–32	4	8
	Lomefloxacin	0.25–32	4	8
Pseudomonas cepacia (10)	OPC-17116	0.25–1	0.25	0.5
	Ciprofloxacin	0.5–8	0.5	2
	Fleroxacin	0.25–8	0.5	2
	Ofloxacin	0.5–8	2	4
	Lomefloxacin	0.5–8	1	4
Xanthomonas maltophilia (10)	OPC-17116	0.25–1	0.5	1
	Ciprofloxacin	0.5–8	4	8
	Fleroxacin	0.5–4	4	4
	Ofloxacin	1–8	4	8
	Lomefloxacin	4–16	4	16
Acinetobacter anitratus (30)	OPC-17116	0.03–64	0.12	16
	Ciprofloxacin	0.25–16	0.5	16
	Fleroxacin	0.25–64	1	16
	Ofloxacin	0.25–16	2	16
	Lomefloxacin	0.25–16	2	16
Haemophilus influenzae (27)	OPC-17116	0.008–0.06	0.008	0.06
	Ciprofloxacin	0.008–0.06	0.015	0.03
	Fleroxacin	0.008–0.12	0.015	0.06
	Ofloxacin	0.008–0.12	0.03	0.06
	Lomefloxacin	0.008–0.12	0.03	0.12

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TABLE 1—*Continued*

Organism (no. tested)	Antimicrobial agent	MIC (μg/ml)[a]		
		Range	50%	90%
Moraxella catarrhalis (15)	OPC-17116	0.008–0.06	0.03	0.06
	Ciprofloxacin	0.008–0.06	0.015	0.06
	Fleroxacin	0.015–0.12	0.12	0.12
	Ofloxacin	0.03–0.06	0.03	0.06
	Lomefloxacin	0.03–0.12	0.03	0.012
Neisseria gonorrhoeae (20)	OPC-17116	0.03–0.12	0.03	0.06
	Ciprofloxacin	0.015–0.12	0.03	0.06
	Fleroxacin	0.03–0.25	0.12	0.12
	Ofloxacin	0.03–0.25	0.12	0.12
	Lomefloxacin	0.03–0.12	0.12	0.12
Staphylococcus aureus, methicillin susceptible (31)	OPC-17116	0.06–0.25	0.12	0.25
	Ciprofloxacin	0.25–4	0.5	1
	Fleroxacin	0.25–4	0.5	1
	Ofloxacin	0.25–4	0.5	2
	Lomefloxacin	0.25–4	1	2
	Temafloxacin	0.12–0.5	0.25	0.5
	Tosufloxacin	0.015–0.25	0.06	0.12
Staphylococcus aureus, methicillin resistant (31)	OPC-17116	0.03–16	0.25	16
	Ciprofloxacin	0.5–>16	1	>16
	Fleroxacin	0.5–>16	4	>16
	Ofloxacin	0.5–>16	2	>16
	Lomefloxacin	1–>16	4	>16
	Temafloxacin	0.12–>16	0.6	>16
	Tosufloxacin	0.06–8	0.12	8
Staphylococcus epidermidis, methicillin susceptible (30)	OPC-17116	0.03–0.25	0.12	0.25
	Ciprofloxacin	0.12–1	0.25	0.5
	Fleroxacin	0.25–2	1	1
	Ofloxacin	0.25–2	0.25	0.5
	Lomefloxacin	0.5–2	1	1
	Temafloxacin	0.25–1	0.25	0.25
	Tosufloxacin	0.03–1	0.25	1
Staphylococcus epidermidis, methicillin resistant (30)	OPC-17116	0.03–8	0.12	8
	Ciprofloxacin	0.12–>16	0.5	>16
	Fleroxacin	0.25–>16	1	>16
	Ofloxacin	0.12–>16	0.5	>16
	Lomefloxacin	0.25–>16	1	>16
	Temafloxacin	0.03–16	0.12	4
	Tosufloxacin	0.12–>16	0.5	16
Streptococcus pyogenes (15)	OPC-17116	0.25–1	0.25	0.5
	Ciprofloxacin	0.25–2	0.5	2
	Fleroxacin	1–8	8	8
	Ofloxacin	0.5–2	0.5	2
	Lomefloxacin	1–8	8	8
	Temafloxacin	0.25–2	0.5	0.5
	Tosufloxacin	0.06–0.5	0.12	0.12
Streptococcus agalactiae (16)	OPC-17116	0.25–0.5	0.25	0.5
	Ciprofloxacin	0.25–4	1	2
	Fleroxacin	1–8	8	8
	Ofloxacin	0.25–4	1	2
	Lomefloxacin	1–8	8	8
Group C, F, and G streptococci (31)	OPC-17116	0.25–0.5	0.25	0.5
	Ciprofloxacin	0.5–8	1	2
	Fleroxacin	1–>8	4	>8
	Ofloxacin	0.25–8	1	2
	Lomefloxacin	1–>8	4	>8
Streptococcus pneumoniae (20)	OPC-17116	0.12–1	0.25	0.5
	Ciprofloxacin	0.5–4	1	2
	Fleroxacin	2–>8	8	8
	Ofloxacin	0.5–4	1	2
	Lomefloxacin	2–>8	8	8
	Temafloxacin	0.25–2	0.5	1
	Tosufloxacin	0.03–0.5	0.12	0.5
Viridans group streptococci (16)	OPC-17116	0.25–1	0.5	0.5
	Ciprofloxacin	0.5–8	2	4
	Fleroxacin	1–>8	4	>8
	Ofloxacin	0.5–8	2	4
	Lomefloxacin	1–>8	4	>8
Enterococcus faecalis (29)	OPC-17116	0.25–4	1	4
	Ciprofloxacin	0.5–>8	2	4
	Fleroxacin	>8	>8	>8
	Ofloxacin	0.5–>8	2	8
	Lomefloxacin	>8	>8	>8
Clostridium perfringens (20)	OPC-17116	0.25–1	0.5	1
	Ciprofloxacin	0.25–4	0.5	2
	Fleroxacin	0.5–4	0.5	2
	Ofloxacin	0.5–4	1	2
	Lomefloxacin	0.5–4	1	4
Bacteroides fragilis (20)	OPC-17116	0.25–4	0.5	4
	Ciprofloxacin	1–32	16	32
	Fleroxacin	1–32	32	32
	Ofloxacin	0.5–16	8	16
	Lomefloxacin	1–32	16	32
	Temafloxacin	2–>16	2	8
	Tosufloxacin	1–8	1	2

[a] 50% and 90%, MICs for 50 and 90% of isolates tested, respectively.

RESULTS

The in vitro activity of OPC-17116 is shown in Table 1. OPC-17116 inhibited 50% of members of the family *Enterobacteriaceae* tested at ≤0.25 μg/ml and 90% of them at ≤2 μg/ml. The activity of OPC-17116 in general was comparable to the activities of the other fluoroquinolones tested (fleroxacin, ofloxacin, and lomefloxacin). The differences in MICs usually were within a two- to fourfold dilution range. For a number of species, ciprofloxacin MICs were two- or fourfold lower than the MICs of OPC-17116. Organisms such as *Escherichia coli* and *Klebsiella*, *Enterobacter*, *Salmonella*, and *Shigella* species were inhibited by ≤0.5 μg/ml, whereas

TABLE 2. Effect of inoculum size on MICs of OPC-17116

Organism[a]	Geometric mean (range) MIC (μg/ml) at CFU of:	
	10^5	10^7
Escherichia coli	0.04 (0.015–0.06)	0.04 (0.03–0.06)
Enterobacter cloacae	0.16 (0.06–0.5)	0.31 (0.06–1)
Klebsiella pneumoniae	0.18 (0.12–0.25)	0.23 (0.12–0.5)
Serratia marcescens	0.45 (0.25–0.5)	0.80 (0.5–2)
Pseudomonas aeruginosa	0.35 (0.12–0.5)	0.80 (0.5–2)
Staphylococcus aureus, methicillin resistant	0.21 (0.06–0.25)	0.80 (0.06–2)

[a] Five isolates of each species were tested.

TABLE 3. Effect of pH on MICs of OPC-17116

Organism[a]	Geometric mean (range) MIC (μg/ml) at pH:		
	6	7	8
Escherichia coli	0.09 (0.06–0.12)	0.04 (0.015–0.06)	0.02 (0.008–0.03)
Enterobacter cloacae	0.78 (0.12–2)	0.18 (0.06–0.5)	0.06 (0.015–0.12)
Klebsiella pneumoniae	0.50 (0.25–1)	0.19 (0.06–0.25)	0.06 (0.03–0.12)
Serratia marcescens	2.95 (0.5–4)	0.63 (0.12–1)	0.16 (0.06–0.25)
Pseudomonas aeruginosa	2 (1–4)	0.38 (0.25–0.5)	0.25 (0.12–0.5)
Staphylococcus aureus, methicillin resistant	2 (0.5–4)	0.20 (0.06–0.25)	0.22 (0.12–0.25)

[a] Five isolates of each species were tested.

for some of the *Providencia* isolates OPC-17116 MICs were higher (1 to 2 μg/ml). OPC-17116 inhibited 90% of *Pseudomonas aeruginosa* isolates tested at 4 μg/ml. It was less active than ciprofloxacin but had activity comparable to or greater than those of the other three fluoroquinolones. The activities of OPC-17116 against *Haemophilus influenzae*, *Moraxella catarrhalis*, and *Neisseria gonorrhoeae* were similar to those of the other quinolones, with 90% of isolates inhibited by 0.06 μg/ml.

Against gram-positive species, OPC-17116 showed greater activity than did all the other agents except tosufloxacin. It inhibited 90% of methicillin-susceptible *Staphylococcus aureus* and *Staphylococcus epidermidis* isolates at ≤0.25 μg/ml, which was greater than the activities of ciprofloxacin and ofloxacin. Methicillin-resistant *Staphylococcus aureus* isolates that were susceptible to ciprofloxacin were inhibited by 0.25 μg/ml, but the methicillin-resistant, ciprofloxacin-resistant isolates were resistant to all of the fluoroquinolones, including OPC-17116. OPC-17116 inhibited 90% of group A and group B streptococci at ≤0.5 μg/ml, making it two- to fourfold more active than ciprofloxacin and ofloxacin but less active than tosufloxacin. It inhibited 90% of the *Streptococcus pneumoniae* isolates at 0.5 μg/ml, whereas the values were 8 μg/ml for fleroxacin and lomefloxacin and 2 μg/ml for ciprofloxacin and ofloxacin. OPC-17116 was twofold more active than ciprofloxacin against *Enterococcus faecalis*, with an MIC for 90% of isolates tested of 2 μg/ml, but for the *Enterococcus faecium* isolates (eight isolates), MICs were 8 μg/ml (data not shown).

OPC-17116 was more active against *Clostridium perfringens* than were the other quinolones tested, inhibiting 90% of these isolates at 1 μg/ml and inhibiting 10 other clostridial isolates at 4 μg/ml (data not shown). Most *Bacteroides fragilis* isolates were inhibited by 4 μg of OPC-17116 per ml, whereas the values were 32 μg/ml for ciprofloxacin and 2 μg/ml for tosufloxacin.

Effects of assay conditions. The effect of inoculum size is shown in Table 2. There was, in general, only a twofold increase in the MICs at 10^7 CFU compared with the MICs at 10^5 CFU for the members of the family *Enterobacteriaceae*, *Pseudomonas aeruginosa*, and *Staphylococcus aureus* tested. The MBCs for *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Serratia marcescens*, *Staphylococcus aureus*, *Streptococcus pyogenes*, and *Moraxella catarrhalis* (five isolates each) were identical or within a twofold dilution of the MICs (data not shown). As shown in Table 3, the activity of OPC-17116 was decreased at pH 6 compared with that at pH 7, and activity at pH 8 was, in general, similar to that at pH 7.

The effect of increased concentrations of magnesium on the activity of OPC-17116 was determined for several organisms. At 9 mM Mg^{2+}, there was an appreciable increase in the MICs and MBCs (Table 4). However, the MICs remained below ≤2 μg/ml even at a high concentration of magnesium (9 mM). There was no decrease in the in vitro activity of OPC-17116 when the in vitro activity was determined in 50% normal human serum. The MBCs in serum compared with those in Mueller-Hinton broth were unchanged for the two isolates each of *Escherichia coli*, *Serratia marcescens*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Streptococcus pyogenes*, and *Streptococcus pneumoniae* tested (Table 5).

PAE. The PAE of OPC-17116 was determined for a single isolate each of *Pseudomonas aeruginosa* and *Staphylococcus aureus*. A PAE of 2 h was obtained after a 2-h exposure to a concentration one time the MBC and a PAE of 4.2 h after a 2-h exposure at a concentration that was twice the MBC for *Pseudomonas aeruginosa*. A PAE of 4 h was achieved for a methicillin-resistant, ciprofloxacin-susceptible *Staphylococcus aureus* isolate after a 2-h exposure to OPC-17116 at a concentration of twice the MBC.

Development of resistance. As shown in Table 6, the frequency of spontaneous single-step resistance to OPC-17116 by the four organisms tested was $<10^{-9}$. Serial daily transfer of these organisms in the presence of OPC-17116 for 14 days produced an increase in the OPC-17116 MIC for *Escherichia coli* from 0.004 to 0.12 μg/ml, for *Serratia marcescens* from 0.03 to 1 μg/ml, for *Pseudomonas aerugi-*

TABLE 4. Effect of magnesium on activity of OPC-17116

Organism	MIC (μg/ml) in M-H containing Mg^{2+} at[a]:							
	M-H alone		3 mM		6 mM		9 mM	
	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC
Escherichia coli	0.004	0.004	0.015	0.015	0.015	0.03	0.03	0.03
Serratia marcescens	0.03	0.06	0.06	0.06	0.06	0.12	0.06	0.12
Pseudomonas aeruginosa	0.06	0.5	0.12	0.5	0.25	1	0.5	2
Staphylococcus aureus, methicillin resistant	0.03	0.12	0.06	0.12	0.12	0.25	0.25	0.5

[a] M-H, Mueller-Hinton broth.

TABLE 5. Comparison of OPC-17116 MBCs in broth and 50% human serum

Organism	MBC (μg/ml) in:	
	Broth[a]	50% Serum
Escherichia coli	0.015	0.015
Serratia marcescens	0.06	0.12
Pseudomonas aeruginosa	0.5	0.25
Staphylococcus aureus	0.5	0.5
Streptococcus pyogenes	0.12	0.06
Streptococcus pneumoniae	0.12	0.06

[a] Mueller-Hinton broth was used for *Escherichia coli*, *Serratia marcescens*, *Pseudomonas aeruginosa*, and *Staphylococcus aureus*; Todd-Hewitt broth was used for *Streptococcus pyogenes* and *Streptococcus pneumoniae*.

nosa from 0.06 to >32 μg/ml, and for *Staphylococcus aureus* from 0.03 to 1 μg/ml.

DISCUSSION

In the past few years, a large number of fluoroquinolone compounds have been synthesized; and a number of agents are in clinical use in the United States, Europe, and Asia (8). There has been a concerted effort to synthesize new agents with improved activities against gram-positive species (15). OPC-17116 contains an N-1 cyclopropyl group similar to that in ciprofloxacin, but it has a C-5 methyl and a 3-methyl group on the C-7 piperazinyl moiety. Results of this study demonstrated that OPC-17116 inhibits gram-positive species such as staphylococci at concentrations of ≤0.25 μg/ml and inhibits streptococci and *Streptococcus pneumoniae* at concentrations of ≤0.5 μg/ml; this inhibition is improved over those of ciprofloxacin and ofloxacin, both of which have been used to treat infections caused by the organisms mentioned above (8). OPC-17116, in general, was more active than temafloxacin against gram-positive cocci, but it was two- to fourfold less active than tosufloxacin. Resistance of *Staphylococcus aureus* to fluoroquinolones has increased at a remarkable rate (1). Unfortunately, OPC-17116 did not inhibit methicillin-resistant *Staphylococcus aureus* isolates which were resistant to ciprofloxacin.

The activities of OPC-17116 against members of the family *Enterobacteriaceae* and against *Haemophilus*, *Neisseria*, and *Moraxella* species were comparable to the activities of the other agents tested, although ciprofloxacin was the most active agent overall. Ciprofloxacin was severalfold more active than OPC-17116 against *Pseudomonas aeruginosa*.

OPC-17116 is less active at a low pH, and there is a decrease in activity in the presence of high concentrations of magnesium, as has been demonstrated for other fluoroquinolones (7). Similar to the results found for other quinolones, resistant mutants can be selected by repeated passage in the presence of OPC-17116, even though the frequency of single-step spontaneous resistance is low.

TABLE 6. Frequency of spontaneous resistance to OPC-17116

Organism	MIC (μg/ml)	Resistance frequency to:	
		4× the MIC	8× the MIC
Escherichia coli	0.004	$<3.97 \times 10^{-9}$	$<3.97 \times 10^{-9}$
Serratia marcescens	0.03	$<6.16 \times 10^{-9}$	$<6.16 \times 10^{-9}$
Pseudomonas aeruginosa	0.06	$<2.79 \times 10^{-9}$	$<2.79 \times 10^{-9}$
Staphylococcus aureus, methicillin resistant	0.03	$<6.47 \times 10^{-9}$	$<6.47 \times 10^{-9}$

Hara and colleagues (6) have demonstrated that OPC-17116 inhibits *Mycoplasma pneumoniae* and achieves high concentrations in sputum and polymorphonuclear cells. OPC-17116 has also been shown to be very active in vitro against *Legionella* species and to cure guinea pigs infected with *Legionella* species (4). OPC-17116 has been shown to be effective in treating animals systemically infected with methicillin-resistant *Staphylococcus aureus* and with *Streptococcus pneumoniae* (12). Preliminary pharmacokinetic studies have shown that the maximal concentration in plasma after a 400-mg dose is 1.5 to 1.8 μg/ml, with a half-life of 10 to 13 h (14). We have shown that oral doses of 400 mg of OPC-17116 produce bactericidal activity in serum against *Escherichia coli*, *Klebsiella pneumoniae*, *Haemophilus influenzae*, *Streptococcus pyogenes*, *Streptococcus pneumoniae*, and *Pseudomonas aeruginosa* (5). In view of the in vitro activity, efficacy in animal infection experiments, and preliminary pharmacokinetic data, further clinical studies are indicated to evaluate the ultimate role of OPC-17116 in the therapy of clinical infections caused by susceptible microorganisms.

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