In Vitro Activities of Irloxacin and E-3846, Two New Quinolones

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Irloxacin and E-3846 are two new fluorinated quinolones. We evaluated the activities of these antimicrobial agents, ciprofloxacin, ofloxacin, enoxacin, pefloxacin, norfloxacin, and nalidixic acid against 1,161 bacterial strains. Ciprofloxacin was the most active quinolone. Irloxacin did not show great activity. The activity of E-3846 against gram-negative bacteria was similar to those of ofloxacin and pefloxacin, and E-3846 was the most active quinolone against gram-positive bacteria and anaerobes.

In the past few decades, a number of compounds containing a 1,4-dihydro-4-oxo-pyridine-3 carboxylic acid moiety and collectively known as quinolones have been developed. Modifications at positions 1, 6, 7, and 8 have altered both the antibacterial spectrum and potency of the quinolones. Irloxacin and E-3846 are two new quinolones developed in Spain (Laboratorios Esteve) that were shown previously to possess good activity against a broad spectrum of microorganisms (2, 4). In this work, we compare the in vitro activities of irloxacin and E-3846 with those of six reference quinolones (norfloxacin, ciprofloxacin, ofloxacin, pefloxacin, enoxacin, and nalidixic acid). A total of 1,161 clinical isolates were studied (see Table 1).

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The following antibiotics were kindly supplied by their manufacturers: irloxacin and E-3846 (Laboratorios Esteve), norfloxacin (Merck Sharp & Dohme), ciprofloxacin (Bayer), ofloxacin (Glaxo Pharmaceuticals, Ltd.), pefloxacin (Rhône-Poulenc), enoxacin (Dainippon), and nalidixic acid (Sterling-Winthrop). Antimicrobial susceptibility testing was performed by the agar dilution method in accordance with guidelines of the National Committee for Clinical Laboratory Standards (5, 6). The medium used for testing enterobacteria and *Pseudomonas*, *Acinetobacter*, *Branhamella*, and *Staphylococcus* spp. was Mueller-Hinton agar (Difco Laboratories, Detroit, Mich.). The same medium supplemented with 5% sheep blood was used when *Streptococcus* and *Enterococcus* spp. were tested. Wilkins-Chalgren agar supplemented with 5% sheep blood was utilized for testing anaerobes, and Mueller-Hinton agar supplemented with 1% hemoglobin (bioMerieux, Charbonnières les Bains, France) and 1% PoliVitex (bioMerieux) was used when *Brucella*, *Neisseria*, and *Haemophilus* spp. were tested. BCYE- α (Oxoid Ltd., Basingstoke, United Kingdom) was used for testing *Legionella pneumophila*. The inoculum was prepared from a 24-h broth growth. The final inoculum was 10^4 CFU per spot, and results were noted after overnight incubation at 37°C in an aerobic atmosphere, except for *Haemophilus*, *Neisseria*, *Legionella*, and *Branhamella* spp., which were incubated in 5% CO₂, and anaerobes, which were incubated in an atmosphere of 10% CO₂-90% nitrogen. *Campylobacter*

(*Helicobacter*) *pylori* susceptibility studies were performed as previously described (3). Plates were inoculated with a Steers replicator. *Staphylococcus aureus* ATCC 25923, *Enterococcus faecalis* ATCC 29212, *Escherichia coli* ATCC 25922, *Klebsiella pneumoniae* ATCC 13883, *Pseudomonas aeruginosa* ATCC 27853, *Bacteroides fragilis* ATCC 25285, and *Bacteroides thetaiotaomicron* ATCC 29741 were used for quality control.

The susceptibilities of the 1,161 clinical isolates to the quinolones tested are presented in Table 1. The most active quinolone against all species and genera of enterobacteria and *Pseudomonas* was ciprofloxacin. Against *Acinetobacter calcoaceticus*, ofloxacin and pefloxacin were slightly more active than ciprofloxacin. E-3846 showed activity similar to or slightly lower than those of ofloxacin, pefloxacin, and norfloxacin. Irloxacin was more active than nalidixic acid only. Against *Haemophilus influenzae*, *Neisseria gonorrhoeae*, and *Moraxella (Branhamella) catarrhalis*, both β -lactamase producers and non- β -lactamase producers, all fluorinated quinolones were highly active. E-3846 showed activity similar to those of the most active quinolones, while irloxacin was among the less active agents. As a group, the fluorinated quinolones showed moderate activity against *Brucella* spp. and *L. pneumophila*, ciprofloxacin and E-3846 also being the most active. E-3846 and ciprofloxacin were the most active quinolones against *C. pylori*. However, the in vitro activity of quinolones against this organism does not always correlate with in vivo activity. Against gram-positive bacteria, E-3846 was most active while enoxacin, norfloxacin, and irloxacin were the least active fluorinated quinolones. E-3846 also showed the best overall activity against *Bacteroides* spp. Irloxacin had activity similar to those of ciprofloxacin and ofloxacin.

Irloxacin is structurally similar to norfloxacin and pefloxacin. The C-7 substituent of irloxacin is a pyrrolyl, in contrast to the piperazin-1-yl found in norfloxacin and the 4-methyl-piperazin-1-yl found in pefloxacin. Nevertheless, the inclusion of this five-membered heterocyclic ring instead of the six-membered rings of norfloxacin and pefloxacin did not improve activity overall, since irloxacin was less active against gram-negative bacteria and only slightly more active against gram-positive bacteria and anaerobes.

E-3846 is structurally related to ciprofloxacin in the same way that irloxacin is related to norfloxacin, i.e., the only difference between E-3846 and ciprofloxacin is the pyrrolyl C-7 substituent instead of the piperazin-1-yl C-7 substituent of ciprofloxacin. In contrast to irloxacin, E-3846 showed good activity against gram-negative bacteria, likely due to the optimal steric, spatial, and electronic interactions of the

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TABLE 1. In vitro activities of irloxacin, E-3846, and six other antibiotics

Microorganism (no. of strains)	Antibiotic	MIC ($\mu\text{g/ml}$) ^a		
		Range	50%	90%
<i>Escherichia coli</i> (126)	Irloxacin	0.25–4	1	1
	E-3846	≤ 0.008 –1	0.12	0.25
	Norfloxacin	≤ 0.008 –0.5	0.06	0.06
	Ciprofloxacin	≤ 0.008 –0.06	≤ 0.008	0.01
	Ofloxacin	≤ 0.008 –0.5	0.06	0.06
	Pefloxacin	0.03–2	0.06	0.125
	Enoxacin	0.06–2	0.12	0.5
	Nalidixic acid	0.5–8	2	4
<i>Klebsiella pneumoniae</i> (63)	Irloxacin	1– ≥ 128	2	16
	E-3846	0.25–16	0.5	4
	Norfloxacin	0.06–2	0.125	1
	Ciprofloxacin	≤ 0.008 –0.5	≤ 0.008	0.12
	Ofloxacin	0.06–1	0.12	0.5
	Pefloxacin	0.12–4	0.25	2
	Enoxacin	0.06–4	0.25	2
	Nalidixic acid	2– ≥ 128	4	≥ 128
<i>K. oxytoca</i> (42)	Irloxacin	0.25–4	1	2
	E-3846	0.06–1	0.25	0.5
	Norfloxacin	0.03–0.25	0.05	0.25
	Ciprofloxacin	≤ 0.008 –0.06	≤ 0.008	≤ 0.008
	Ofloxacin	0.03–0.5	0.03	0.12
	Pefloxacin	0.06–0.5	0.12	0.25
	Enoxacin	0.06–1	0.12	0.5
	Nalidixic acid	2–32	2	8
<i>Enterobacter</i> spp. (42)	Irloxacin	0.5–64	2	8
	E-3846	0.12–8	0.25	1
	Norfloxacin	0.06–8	0.12	8
	Ciprofloxacin	≤ 0.008 –0.25	≤ 0.008	0.12
	Ofloxacin	0.03–1	0.06	0.5
	Pefloxacin	0.06–4	0.12	1
	Enoxacin	0.06–4	0.12	0.5
	Nalidixic acid	2– ≥ 128	4	16
<i>Serratia</i> spp. (42)	Irloxacin	0.5–32	2	8
	E-3846	0.12–8	0.25	4
	Norfloxacin	0.06–32	0.25	8
	Ciprofloxacin	≤ 0.008 –8	0.01	2
	Ofloxacin	0.06–8	0.5	4
	Pefloxacin	0.06–32	1	16
	Enoxacin	0.12–32	2	8
	Nalidixic acid	1– ≥ 128	4	≥ 128
<i>Citrobacter</i> spp. (42)	Irloxacin	1– ≥ 128	4	8
	E-3846	0.25–8	0.5	1
	Norfloxacin	0.06–4	0.12	0.5
	Ciprofloxacin	≤ 0.008 –1	0.01	0.12
	Ofloxacin	0.06–1	0.25	0.5
	Pefloxacin	0.12–4	0.25	1
	Enoxacin	0.06–16	0.25	1
	Nalidixic acid	4– ≥ 128	8	16
<i>Proteus mirabilis</i> (126)	Irloxacin	0.25–4	1	2
	E-3846	0.12–1	0.5	0.5
	Norfloxacin	0.03–0.5	0.06	0.12
	Ciprofloxacin	≤ 0.008 –0.12	≤ 0.008	0.06
	Ofloxacin	0.06–0.25	0.06	0.12
	Pefloxacin	0.12–0.5	0.12	0.25
	Enoxacin	0.25–0.5	0.25	0.5
	Nalidixic acid	2–16	4	8

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TABLE 1—Continued

Microorganism (no. of strains)	Antibiotic	MIC ($\mu\text{g/ml}$) ^a		
		Range	50%	90%
<i>P. vulgaris</i> (21)	Irloxacin	0.5–8	2	8
	E-3846	0.25–8	1	4
	Norfloxacin	0.03–0.25	0.06	0.12
	Ciprofloxacin	0.01–0.06	0.03	0.06
	Ofloxacin	0.03–1	0.12	0.5
	Pefloxacin	0.06–0.5	0.25	0.5
	Enoxacin	0.06–0.5	0.25	0.5
	Nalidixic acid	1–64	4	32
<i>Morganella morganii</i> (42)	Irloxacin	0.5–1	0.5	1
	E-3846	0.25–0.5	0.25	0.5
	Norfloxacin	0.03–0.25	0.03	0.12
	Ciprofloxacin	≤ 0.008 –0.03	≤ 0.008	0.03
	Ofloxacin	0.06–0.12	0.06	0.12
	Pefloxacin	0.12–0.5	0.12	0.5
	Enoxacin	0.06–0.25	0.12	0.25
	Nalidixic acid	1–2	1	2
<i>Salmonella</i> spp. (21)	Irloxacin	0.5–1	0.5	1
	E-3846	≤ 0.008 –0.25	0.12	0.25
	Norfloxacin	≤ 0.008 –0.12	0.06	0.06
	Ciprofloxacin	≤ 0.008 –0.01	≤ 0.008	0.01
	Ofloxacin	0.06–0.12	0.12	0.12
	Pefloxacin	0.06–0.12	0.12	0.12
	Enoxacin	0.12–0.5	0.25	0.2
	Nalidixic acid	2–8	4	4
<i>Shigella</i> spp. (21)	Irloxacin	0.25–0.5	0.5	0.5
	E-3846	0.01–0.12	0.03	0.06
	Norfloxacin	0.01–0.06	0.03	0.06
	Ciprofloxacin	≤ 0.008	≤ 0.008	≤ 0.008
	Ofloxacin	0.03–0.06	0.06	0.06
	Pefloxacin	0.03–0.06	0.03	0.06
	Enoxacin	0.06–0.12	0.06	0.12
	Nalidixic acid	2–4	2	2
<i>Yersinia enterocolitica</i> (27)	Irloxacin	0.12–0.5	0.5	0.5
	E-3846	0.01–0.25	0.12	0.12
	Norfloxacin	0.01–0.12	0.06	0.06
	Ciprofloxacin	≤ 0.008 –0.01	≤ 0.008	0.01
	Ofloxacin	0.01–0.12	0.06	0.12
	Pefloxacin	0.01–0.12	0.06	0.12
	Enoxacin	0.03–0.25	0.12	0.12
	Nalidixic acid	0.5–4	2	2
<i>Pseudomonas aeruginosa</i> (42)	Irloxacin	4– ≥ 128	8	≥ 128
	E-3846	1– ≥ 128	4	64
	Norfloxacin	0.5– ≥ 128	1	16
	Ciprofloxacin	0.12–32	0.25	8
	Ofloxacin	0.5–64	2	32
	Pefloxacin	1– ≥ 128	4	64
	Enoxacin	0.5– ≥ 128	2	32
	Nalidixic acid	64– ≥ 128	≥ 128	≥ 128
<i>Haemophilus influenzae</i> (32)	Irloxacin	0.01–0.5	0.03	0.12
	E-3846	≤ 0.008 –0.25	0.01	0.03
	Norfloxacin	0.01–0.06	0.03	0.06
	Ciprofloxacin	≤ 0.008 –0.01	0.01	0.01
	Ofloxacin	0.01–0.06	0.03	0.06
	Pefloxacin	0.01–0.06	0.03	0.06
	Enoxacin	0.03–0.25	0.03	0.06
	Nalidixic acid	0.01–1	0.12	0.5
<i>Neisseria gonorrhoeae</i> (21)	Irloxacin	≤ 0.008 –0.25	0.03	0.06
	E-3846	≤ 0.008 –0.03	≤ 0.008	≤ 0.008
	Norfloxacin	≤ 0.008 –0.06	0.01	0.03

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TABLE 1—Continued

Microorganism (no. of strains)	Antibiotic	MIC ($\mu\text{g/ml}$) ^a		
		Range	50%	90%
	Ciprofloxacin	≤ 0.008	≤ 0.008	≤ 0.008
	Ofloxacin	$\leq 0.008-0.06$	≤ 0.008	0.03
	Pefloxacin	$\leq 0.008-0.12$	≤ 0.008	0.06
	Enoxacin	0.03-0.12	0.03	0.12
	Nalidixic acid	0.25-1	0.25	0.5
<i>Moraxella catarrhalis</i> (27)	Irloxacin	0.12-0.25	0.12	0.12
	E-3846	0.01-0.03	0.03	0.03
	Norfloracin	0.12	0.12	0.12
	Ciprofloxacin	0.03	0.03	0.03
	Ofloxacin	0.06	0.06	0.06
	Pefloxacin	0.12	0.12	0.12
	Enoxacin	0.12-0.25	0.25	0.25
	Nalidixic acid	2-4	4	4
<i>Brucella</i> spp. (64)	Irloxacin	1-2	1	1
	E-3846	0.25-0.5	0.25	0.25
	Norfloracin	1-2	1	2
	Ciprofloxacin	0.25	0.25	0.25
	Ofloxacin	0.5-1	0.5	0.5
	Pefloxacin	2-4	2	2
	Enoxacin	4-8	4	4
	Nalidixic acid	16	16	16
<i>Legionella pneumophila</i> (31)	Irloxacin	1-4	2	4
	E-3846	0.5-1	0.5	1
	Norfloracin	2	2	2
	Ciprofloxacin	0.25-0.5	0.5	0.5
	Ofloxacin	0.5-1	0.5	1
	Pefloxacin	1-2	1	2
	Enoxacin	2-4	4	4
	Nalidixic acid	2-8	2	4
<i>Acinetobacter calcoaceticus</i> (11)	Irloxacin	1-64	16	64
	E-3846	0.25-16	2	8
	Norfloracin	4-64	4	32
	Ciprofloxacin	0.5-32	0.5	8
	Ofloxacin	0.5-16	0.5	4
	Pefloxacin	0.5-16	2	4
	Enoxacin	0.5-16	2	8
	Nalidixic acid	4- ≥ 128	8	≥ 128
<i>Staphylococcus aureus</i> (32)	Irloxacin	0.25-0.5	0.25	0.5
	E-3846	0.06-0.12	0.12	0.12
	Norfloracin	1-4	2	4
	Ciprofloxacin	0.25-1	0.5	0.5
	Ofloxacin	0.25-0.5	0.25	0.5
	Pefloxacin	0.25-0.5	0.25	0.5
	Enoxacin	0.5-2	0.5	1
	Nalidixic acid	64- ≥ 128	≥ 128	≥ 128
<i>S. epidermidis</i> (32)	Irloxacin	0.5-1	1	1
	E-3846	0.12-0.25	0.25	0.25
	Norfloracin	1-2	1	1
	Ciprofloxacin	0.12-0.5	0.25	0.25
	Ofloxacin	0.12-0.5	0.25	0.5
	Pefloxacin	0.25-1	0.5	0.5
	Enoxacin	0.25-1	0.5	0.5
	Nalidixic acid	64- ≥ 128	≥ 128	≥ 128
<i>Enterococcus faecalis</i> (15)	Irloxacin	1-8	2	8
	E-3846	0.5-1	0.5	1
	Norfloracin	2-64	8	16
	Ciprofloxacin	0.5-8	1	2
	Ofloxacin	0.5-8	1	2
	Pefloxacin	2-16	2	4
	Enoxacin	4-64	8	16
	Nalidixic acid	2- ≥ 128	8	≥ 128

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TABLE 1—Continued

Microorganism (no. of strains)	Antibiotic	MIC ($\mu\text{g/ml}$) ^a		
		Range	50%	90%
<i>Streptococcus pneumoniae</i> (17)	Irloxacin	0.5–8	4	8
	E-3846	0.06–1	0.5	1
	Norfloxacin	1–16	8	16
	Ciprofloxacin	0.25–4	2	2
	Ofloxacin	0.25–4	2	2
	Pefloxacin	2–16	8	8
	Enoxacin	8–32	8	16
	Nalidixic acid	16– ≥ 128	≥ 128	≥ 128
<i>Bacteroides fragilis</i> (64)	Irloxacin	0.5–32	8	16
	E-3846	0.12–4	4	4
	Norfloxacin	1– ≥ 128	32	≥ 128
	Ciprofloxacin	0.5–32	4	16
	Ofloxacin	0.25–16	4	8
	Pefloxacin	1–64	16	32
	Enoxacin	1–64	32	64
	Nalidixic acid	32– ≥ 128	≥ 128	≥ 128
<i>B. thetaiotaomicron</i> (32)	Irloxacin	0.5–32	8	16
	E-3846	0.12–4	2	4
	Norfloxacin	1– ≥ 128	32	≥ 128
	Ciprofloxacin	0.5–32	4	16
	Ofloxacin	0.25–16	2	8
	Pefloxacin	1–64	16	32
	Enoxacin	1–64	32	64
	Nalidixic acid	32– ≥ 128	≥ 128	≥ 128
<i>B. ovatus</i> (32)	Irloxacin	1–16	8	16
	E-3846	0.25–4	4	4
	Norfloxacin	2–64	16	64
	Ciprofloxacin	1–16	4	16
	Ofloxacin	0.5–8	4	8
	Pefloxacin	2–32	16	32
	Enoxacin	4–64	16	64
	Nalidixic acid	64– ≥ 128	≥ 128	≥ 128
<i>B. vulgatus</i> (32)	Irloxacin	1–8	4	8
	E-3846	0.25–4	2	4
	Norfloxacin	4– ≥ 128	32	64
	Ciprofloxacin	1–8	2	8
	Ofloxacin	0.5–8	2	4
	Pefloxacin	2–32	16	32
	Enoxacin	4–64	32	64
	Nalidixic acid	64– ≥ 128	≥ 128	≥ 128
<i>B. distasonis</i> (32)	Irloxacin	0.5–16	4	16
	E-3846	0.25–4	2	4
	Norfloxacin	1– ≥ 128	32	≥ 128
	Ciprofloxacin	0.5–16	4	16
	Ofloxacin	0.25–16	4	8
	Pefloxacin	1–64	16	32
	Enoxacin	2–64	32	64
	Nalidixic acid	32– ≥ 128	≥ 128	≥ 128
<i>Campylobacter pylori</i> (30)	Irloxacin	1–16	2	16
	E-3846	≤ 0.008 –0.03	≤ 0.008	0.01
	Norfloxacin	0.5–2	1	2
	Ciprofloxacin	≤ 0.008 –0.25	0.03	0.125
	Ofloxacin	0.25–2	1	2
	Pefloxacin	0.5–8	4	4
	Enoxacin	1–32	8	8
	Nalidixic acid	16– ≥ 128	≥ 128	≥ 128

^a 50% and 90%, MIC for 50 and 90% of strains tested, respectively.

cyclopropyl group (1). It was also the most active of the quinolones tested against gram-positive bacteria and anaerobes. If pharmacokinetics studies show a favorable profile, its broad spectrum may make E-3846 a promising antimicrobial agent.

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