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Prulifloxacin: clinical studies of a broad-spectrum quinolone agent

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Prulifloxacin, the lipophilic prodrug of ulifloxacin, is a new oral fluoroquinolone with a broad spectrum of *in vitro* activity against various Gram-positive and Gram-negative microorganisms. Currently, it is the most potent *in vitro* fluoroquinolone against *Escherichia coli* and *Pseudomonas aeruginosa*, and also has the lowest potential of inducing the emergence of resistant strains for these bacteria. It exhibits good penetration in target tissues and fluids, and possesses a long half-life, thus allowing for once-daily administration. Prulifloxacin has been successfully tested in Phase III randomized, controlled trials including patients with acute exacerbations of chronic bronchitis, uncomplicated and complicated urinary tract infections, and chronic bacterial prostatitis. Results are awaited from recently completed and ongoing Phase III randomized, placebo-controlled studies testing prulifloxacin for the treatment of traveler's diarrhea. Prulifloxacin has an acceptable toxicity profile, comparable to that of other fluoroquinolones, with gastric disturbances, diarrhea, nausea and skin rash of mild-to-moderate severity being the most frequent adverse events. Additional research is needed to further elucidate the promising role of prulifloxacin in the treatment of infections sustained by multidrug-resistant pathogens and to consolidate the wide spectrum of activity from a clinical standpoint.

Overview of the current market

In the last decade, the efficacy and relative safety of fluoroquinolones administered orally or intravenously have made them attractive for the management of a wide range of infectious diseases [1].

Fluoroquinolones are currently considered first-line agents for the management of respiratory tract bacterial infections, namely community-acquired and nosocomial pneumonia, complicated or exacerbating chronic bronchitis in immunocompetent adults, and sinusitis [2]. Some molecules of this class are also recommended as the therapy of choice against severe infections localized in different organs and systems in outpatients with multiple comorbidities and inpatients of both intensive and nonintensive care units, especially when *Pseudomonas aeruginosa* is the isolated or suspected causative microorganism [2].

Moreover, fluoroquinolones also represent an established treatment option against uncomplicated urinary tract infections (UTIs), especially when resistance to first-line agents is high, complicated UTIs, acute and chronic bacterial prostatitis, sexually transmitted diseases and pelvic infections [1]. Finally, they have documented efficacy in the management of skin and other soft tissue infections, bone and joint infections, and gastrointestinal infections caused by a variety of bacteria [1].

The emergence of multidrug resistance (MDR) among Gram-negative bacteria, especially *P. aeruginosa*, both in community-acquired, mainly chronic infections, and healthcare-associated infections is a matter of great concern [3–5], and this has prompted the search for drugs with a potent antibacterial activity against these serious pathogens. Consequently, the recent entry of prulifloxacin, a new fluoroquinolone with a promising pharmacokinetic and pharmacodynamic profile, into this field has been welcomed with enthusiasm.

In the present article, the available data on the efficacy and tolerability of prulifloxacin extensively examined in previous publications [6–9] are reviewed and the recent clinical applications of the drug with new indications are presented.

Introduction to prulifloxacin

Prulifloxacin, a new oral antibacterial agent of the fluoroquinolone class, is the lipophilic prodrug of the thiazeto-quinoline carboxylic acid derivative ulifloxacin, which has a broad spectrum of *in vitro* activity against various Gram-negative and Gram-positive bacteria. The molecule was synthesized in 1987 and patented in 1989 in Japan [101]. The molecular formula is $C_{21}H_{20}FN_3O_6S$ and the molecular weight 461.46 g/mol. The chemical structure of prulifloxacin contains the skeletal quinolone with a four-member ring in the 1,2-position including a sulfur atom to increase antibacterial activity and an oxodioxolenylmethyl group in the 7-piperazine ring to improve its bioavailability (Figure 1).

Keywords

chronic bronchitis ■ clinical trials ■ fluoroquinolones ■ gastroenteritis ■ multiple drug resistance ■ prostatitis ■ prulifloxacin ■ *Pseudomonas aeruginosa* ■ ulifloxacin ■ urinary tract infection

After absorption from the gastrointestinal tract, prulifloxacin undergoes immediate first-pass metabolism, being hydrolyzed by serum esterases, mainly paraoxonase [10], and quantitatively transformed into the active compound ulifloxacin (Figure 1). Similarly to other quinolones, ulifloxacin exerts its antibacterial action via the inhibition of DNA gyrase and topoisomerase IV enzymes, thereby preventing DNA replication, transcription, repair and recombination [11,12]. Prulifloxacin has a long elimination half-life, thus allowing for once-daily administration [Mignot *et al.*: A repeat dose of NM441 in healthy volunteers. Rome, Italy, 7th May 2001. Data on file].

Pharmacokinetics & metabolism

The pharmacokinetic properties of prulifloxacin were studied in animal models. In rats, after oral administration the drug was mainly absorbed in the upper intestine and quantitatively metabolized by serum esterases to the active metabolite ulifloxacin [10]. The maximum tissue distribution was achieved 1 h after oral administration, with high levels in the liver and kidneys, moderate levels in the lung, spleen, pancreas and mandible glands, and negligible levels in the cerebrum and cerebellum [13]. In lactating rats, ulifloxacin concentration in milk was higher than in plasma [14]. A similar metabolism was demonstrated in dogs and monkeys [15].

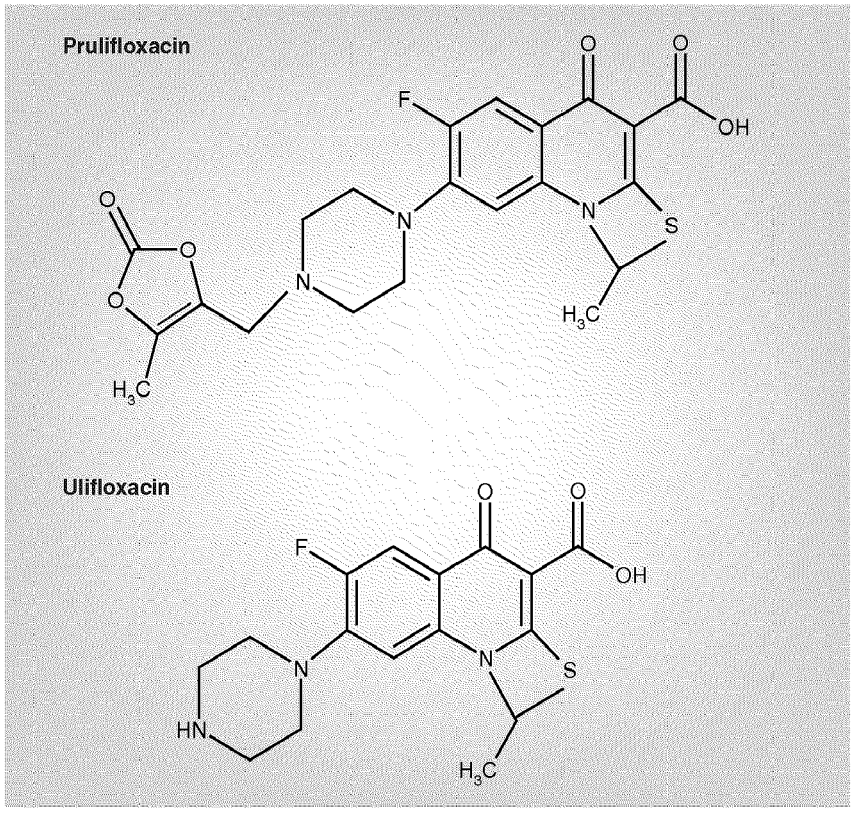

Figure 1. The prodrug prulifloxacin and its active metabolite ulifloxacin.

Prulifloxacin in humans was first investigated in Japan [16–19] and subsequently in Europe [Mignot *et al.*: A repeat dose of NM441 in healthy volunteers. Rome, Italy, 7 May 2001. Data on file; Villa *et al.*: Safety, tolerability and pharmacokinetic evaluation of prulifloxacin (AF3012) 600 mg single dose in patients with mild or moderate renal impairment. Rome, Italy, 28th August 1998. Data on file] [20,21] in young and elderly healthy subjects in several dose-ranging studies after single- or repeat-dose administration. Also in humans, following oral administration the prodrug is rapidly and extensively metabolized to ulifloxacin by first-pass metabolism, so that there are no detectable concentrations of prulifloxacin in plasma. Prulifloxacin absorption is reduced when the drug is taken with milk. Pharmacokinetic parameters of prulifloxacin and ulifloxacin are detailed in Table 1.

The protein binding of ulifloxacin assessed *in vitro* and *in vivo* is approximately 50% in a concentration range of 0.1–10 µg/ml [Angelini ACRAF SpA investigator's brochure. Rome, Italy, 20th April 2004. Data on file] [13]. With a mean apparent volume of distribution of 1231 l after a single 600-mg prulifloxacin dose, ulifloxacin shows good penetration into many tissues, except for the CNS [22,23]. Its levels detected in some tissues and fluids (i.e., urine) are greater and longer lasting than plasma concentrations [13,14,21].

After administration of a single 600-mg dose of prulifloxacin to patients undergoing lobectomy for lung cancer, Concia *et al.* found that the overall mean corrected lung/plasma ratio for ulifloxacin over the 24-h period was 6.9 (range: 1.2–14.1) [24]. When sampling intervals were assessed, the corrected lung/plasma ratios were 7.5 (2 h after dosing), 6.3 (4 h), 4.3 (6 h), 7.0 (12 h) and 9.2 (24 h). The mean corrected lung/plasma area under the concentration–time curve ratio was 6.3. Gorlero *et al.* evaluated the penetration of prulifloxacin, administered as a single or repeat 3-day 600-mg dose, into gynecological tissues [22]. Mean ulifloxacin concentrations in plasma and tissues were higher in all cases for repeat than for single dosing. Specifically, 3 h after repeat and single administration, mean ulifloxacin concentrations were 1.38 and 0.81 µg/g in fallopian tubes, 1.48 and 1.05 µg/g in the posterior fornix, 1.46 and 1.45 µg/g in the portio vaginalis, and 2.20 and 1.39 µg/g in the endometrium, respectively. Mean tissue/plasma ratios for ulifloxacin ranged between 1.5 and 3 in all cases. Ulifloxacin tissue levels observed after repeat administration were generally higher than minimum inhibitory concentration (MIC) values for the bacteria most frequently involved in gynecological infections.

Table 1. Pharmacokinetic parameters of the active metabolite ulifloxacin after oral administration of single or repeat 600-mg dose of the prodrug prulifloxacin*.

Dose	C_{max} (mg/l)‡	T_{max} (h)§	$T_{½}$ (h)‡	AUC (mgh/l)‡
Single dose	1.6	1	10.7	7.3
Steady state after 12-day administration	2	1	7.6	7.6

Data taken from Mignot et al. *A repeat dose study of NM441 in healthy volunteers. Rome, Italy, 7 May 2001 and* [22].
‡*Mean value.*
§*Median value.*
AUC: Area under the curve; C_{max}: Maximum concentration; $T_{1/2}$: Half life; T_{max}: Time to maximum concentration.

The salivary concentrations of ulifloxacin are approximately 20% of plasma concentrations, while in the bile ulifloxacin reached concentrations of approximately 42 mg/l, which are significantly higher than MIC values for the most common pathogens [23].

Ulifloxacin also has a good intracellular penetration. It actively enters polymorphonuclear leukocytes [25,26] and shows bactericidal activity against phagocytosed bacteria, such as *P. aeruginosa* and *Klebsiella pneumoniae* [27]. Specifically, in an *in vitro* study with human polymorphonuclear leukocytes, the intracellular/extracellular concentration ratio for ulifloxacin was 12.3, with an extracellular concentration of 20 µg/ml, slightly higher than ciprofloxacin [25]. In a mouse model, ulifloxacin uptake into peritoneal macrophages was high, with an intracellular/extracellular concentration ratio ranging from 5 to 8, depending on concentration and incubation time [26].

At 48 h after administration, 17–23% of a single 300–600-mg dose of prulifloxacin was excreted as ulifloxacin in the urine and 17–29% in the feces [Angelini ACRAF SpA investigator's brochure. Rome, Italy, 20th April 2004. Data on file] [21]. Renal clearance of ulifloxacin (approximately 170 ml/min) is not dose-dependent, and is higher than creatinine clearance, most likely due to the presence of an additional, active tubular secretion [21].

In healthy elderly (aged >65 years) Caucasian subjects receiving a single 600-mg dose of prulifloxacin, the area under the curve (11.44 µg/h/ml) and the elimination half-life (13.4 h) of ulifloxacin were increased by 57 and 23%, respectively, compared with the values observed in young healthy volunteers. However, both renal clearance (168 ml/min) and urinary excretion ratio (21%) of ulifloxacin were similar to those seen in younger individuals [Angelini ACRAF SpA investigator's brochure. Rome, Italy, 20th April 2004. Data on file].

In patients with mild or moderate renal failure (creatinine clearance 20–60 ml/min), changes in pharmacokinetic parameters of ulifloxacin (i.e., increased time to peak concentration, elimination half-life and area under the curve, and decreased renal clearance and urinary excretion) were correlated to the severity of renal impairment, therefore dosage adjustment in patients with any degree of renal impairment is recommended [Villa *et al.*: Safety, tolerability and pharmacokinetic evaluation of prulifloxacin (AF3012) 600 mg single dose in patients with mild or moderate renal impairment. Rome, Italy, 28th August 1998. Data on file].

Pharmacodynamics

In vitro activity

The *in vitro* antimicrobial activity of ulifloxacin was evaluated using MIC tests performed with standard broth or agar dilution methods recommended by the National Committee for Clinical Laboratory Standards guidelines [28,29]. MIC_{50} and MIC_{90} are the minimum concentrations of the antibacterial agent required to inhibit the growth of 50 and 90% of the tested microorganisms, respectively.

Besides data from two earlier Japanese *in vitro* studies [11,30], more recent data are available from two European studies including clinical isolates obtained between 1998 and 2000 [31,32]. Based on early pharmacokinetic and pharmacodynamic data, the following MIC breakpoints have tentatively been proposed: susceptible: 1 mg/l or less; intermediate: equal to 2 mg/l; and resistant: 4 mg/l or more. In the study by Montanari *et al.* [33], ulifloxacin MIC versus zone diameter scattergrams and discrepancy rates were analyzed in 461 freshly isolated clinical strains (237 Enterobacteriaceae, 101 nonfermenters and 123 Gram-positive bacteria). The following zone diameter breakpoints were chosen and are proposed for the interpretation of ulifloxacin disc (5 µg) test results: 15 mm or less and 19 mm or more for Enterobacteriaceae, 16 mm or less and 20 mm or more for nonfermenters, and 14 mm or less and 18 mm or more for Gram-positive bacteria. By applying these breakpoint values, no major errors were detected, while minor errors were largely below the accepted discrepancy rates.

Representative studies in Italy and Spain [31,32] indicate that ulifloxacin has an *in vitro* activity against a variety of clinical isolates of Gram-negative and Gram-positive bacteria commonly associated with chronic bronchitis, pneumonia and UTI (Table 2). The *in vitro* activity of ulifloxacin was generally greater than that of other fluoroquinolones, especially ciprofloxacin, against clinical isolates of *Escherichia coli*, *Klebsiella* spp., *Proteus* spp., *Providencia* spp., *Morganella* spp., *P. aeruginosa*, *Moraxella catarrhalis* and *Haemophilus influenzae* collected from Italian and Spanish healthcare facilities. Ulifloxacin showed good activity against clinical isolates of ciprofloxacin-susceptible and gentamicin-resistant *P. aeruginosa* strains (MIC_{90} <1 mg/l) [32]. Like other fluoroquinolones, ulifloxacin had scarce activity against ciprofloxacin-resistant *P. aeruginosa* strains isolated in Italy and Spain. Ulifloxacin demonstrated lower capability than ciprofloxacin to select resistant strains after repeated exposure against Gram-negative pathogens [Angelini ACRAF SpA investigator's brochure. Rome, Italy, 20th April 2004. Data on file]. After seven passages at subinhibitory concentrations of either agent in *in vitro* studies, MIC values for ulifloxacin and ciprofloxacin were unchanged and twofold higher, respectively, against *E. coli*, and four- and sixfold higher, respectively, against *P. aeruginosa*. Roveta *et al.* [34] investigated the activity of ulifloxacin compared with ciprofloxacin, levofloxacin and moxifloxacin against a large (n = 300) collection of *P. aeruginosa* strains characterized according to the National Committee for Clinical Laboratory Standards microdilution method [28]. These strains were isolated from Italian patients with cystic fibrosis, pneumonia, bloodstream infections and UTIs, and the MDR phenotype of *P. aeruginosa* (resistance to more than three primary antimicrobial agents) represented 46% of them. Ulifloxacin emerged as the most potent antipseudomonal drug among the fluoroquinolones tested, with 72% of susceptible strains versus 65, 61 and 23% for ciprofloxacin, levofloxacin and moxifloxacin, respectively. MIC_{50} values were 0.25 mg/l for ulifloxacin, and 0.5, 2 and 4 mg/l for ciprofloxacin, levofloxacin and moxifloxacin, respectively. Similar results were obtained by Ceccarini *et al.* [35], who evaluated the activity of ulifloxacin versus other fluoroquinolones against *P. aeruginosa* by assessing the bactericidal effect on pseudomonal biofilm disrupted by sonication and measuring bacterial regrowth. Although a high biofilm eradication rate was observed at concentrations fourfold higher than MIC for all tested drugs, a 5 log_{10} reduction in biofilm population was obtained at concentrations of 1–2 mg/l (equivalent to those found in plasma and tissues after oral treatment) for ulifloxacin and at higher concentrations for ciprofloxacin (2 mg/l), levofloxacin (4–8 mg/l) and moxifloxacin (8–16 mg/l).

The *in vitro* activity of ulifloxacin against Gram-positive bacteria including *Streptococcus* spp., *S. aureus*, *Enterococcus* spp. and coagulase-negative staphylococci was similar or greater than that of ciprofloxacin, but lower than that of moxifloxacin [31]. Susceptibility of *Streptococcus pneumoniae* isolated in Italy and *Streptococcus pyogenes* collected in Spain was similar to other fluoroquinolones, against *S. pneumoniae*, irrespective to penicillin susceptibility, ulifloxacin and ciprofloxacin were less active than moxifloxacin [32]. Against oxacillin- or methicillin-susceptible *S. aureus*, ulifloxacin was highly effective (MIC_{90} ≤0.5 mg/l) [31,32]. Ulifloxacin was active against *Enterococcus faecalis* isolated from the urinary tract, except against vancomycin-resistant isolates collected from Spanish hospitals (MIC_{90} >4 mg/l) [32]. Methicillin-resistant *S. aureus* and coagulase-negative staphylococci strains were resistant to ulifloxacin as well as to other fluoroquinolones.

In the study by Gemignani *et al.* including 647 clinical isolates collected in a single Italian tertiary referral hospital, overall susceptibility rate assessed by disc diffusion assay for ulifloxacin was similar to that for ciprofloxacin and levofloxacin against Gram-negative pathogens (65 vs 64 and 59%), and to that for moxifloxacin against Gram-positive bacteria (56 vs 64%) [36]. When only strains isolated from intensive care units or diabetic foot clinics (that are typically MDR) were considered, the rate of quinolone resistance was lower for ulifloxacin than for the comparators, although significance was not reported.

Ulifloxacin activity was tested *in vitro* against 582 strains isolated from worldwide patients with traveler's diarrhea and compared with that of ciprofloxacin, nalidixic acid, rifaximin, ampicillin, doxycycline and cotrimoxazole [37]. The common causative agents were *E. coli*, *Aeromonas* spp., *Campylobacter* spp., *Salmonella* spp., *Shigella* spp., *Yersinia* spp. and *Vibrio* spp. Ulifloxacin was the most active antibacterial against all species tested (MIC_{50} ≤0.03 mg/l, MIC_{90} ≤0.06 mg/l), except for *Campylobacter* spp. (MIC_{50} ≤0.03 mg/l, MIC_{90} ≥4 mg/l), for which the resistance rate was similar to that of ciprofloxacin. Its potency was two- to fourfold higher than that of ciprofloxacin.

Table 2. *In vitro* activity of ulifloxacin compared with reference fluoroquinolone agents against common Gram-negative and Gram-positive clinical isolates*.

Microorganisms (no. isolates), origin	Fluoroquinolone agent	MIC range (mg/l)	MIC_{50} (mg/l)	MIC_{90} (mg/l)
Gram-negative bacteria				
Escherichia coli (37) community, Italy	Ulifloxacin	≤0.015 to 1	≤0.015	0.12
	Ciprofloxacin	≤0.015 to 8	≤0.015	0.5
	Levofloxacin	≤0.015 to 4	0.03	0.25
	Moxifloxacin	≤0.015 to 16	0.03	0.5
Klebsiella spp. (15) community, Italy	Ulifloxacin	≤0.015 to 0.25	≤0.015	0.12
	Ciprofloxacin	≤0.015 to 0.5	0.03	0.25
	Levofloxacin	0.03 to 0.5	0.06	0.25
	Moxifloxacin	0.03 to 1	0.12	0.25
Haemophilus spp. (24) community, Italy	Ulifloxacin	≤0.015 to 0.03	≤0.015	≤0.015
	Ciprofloxacin	≤0.015	≤0.015	≤0.015
	Levofloxacin	≤0.015 to 0.06	≤0.015	0.03
	Moxifloxacin	≤0.015 to 0.06	≤0.015	0.03
Proteus, Providencia, Morganella spp. (23) community, Italy	Ulifloxacin	≤0.015 to 2	≤0.015	0.5
	Ciprofloxacin	≤0.015 to 2	0.03	1
	Levofloxacin	≤0.015 to 2	0.06	0.5
	Moxifloxacin	0.06 to 8	0.25	4
Pseudomonas aeruginosa (16) community, Italy	Ulifloxacin	0.06 to 64	1	32
	Ciprofloxacin	0.06 to 128	2	64
	Levofloxacin	0.5 to 128	4	64
	Moxifloxacin	0.8 to >128	64	128
Pseudomonas aeruginosa (75) ciprofloxacin-sensitive, Spain	Ulifloxacin	≤0.015 to 1	0.25	1
	Ciprofloxacin	0.06 to 1	0.5	1
	Levofloxacin	0.25 to 4	1	2
	Moxifloxacin	0.5 to >4	4	>4
Gram-positive bacteria				
Enterococcus faecalis (26) nosocomial, Italy	Ulifloxacin	0.12 to 64	0.5	2
	Ciprofloxacin	0.25 to 64	1	2
	Levofloxacin	0.25 to 32	0.5	2
	Moxifloxacin	≤0.015 to 16	0.25	0.5
Enterococcus faecium (18) nosocomial, Italy	Ulifloxacin	0.03 to >128	0.5	16
	Ciprofloxacin	0.5 to >128	1	16
	Levofloxacin	0.12 to >128	0.5	16
	Moxifloxacin	0.03 to 64	0.25	4
Staphylococcus aureus (26) community, oxacillin-sensitive, Italy	Ulifloxacin	0.12 to 0.5	0.25	0.25
	Ciprofloxacin	0.12 to 0.5	0.25	0.5
	Levofloxacin	0.06 to 0.25	0.12	0.25
	Moxifloxacin	≤0.015 to 0.25	0.03	0.06
Streptococcus pneumoniae (36) community, Italy	Ulifloxacin	≤0.015 to 2	0.5	1
	Ciprofloxacin	≤0.015 to 2	0.5	1
	Levofloxacin	≤0.015 to 1	0.5	1
	Moxifloxacin	≤0.015 to 0.25	0.12	0.12
Streptococcus pyogenes (21) community, Italy	Ulifloxacin	0.12 to 2	0.25	1
	Ciprofloxacin	0.25 to 2	0.25	1
	Levofloxacin	0.25 to 2	0.25	1
	Moxifloxacin	0.06 to 0.5	0.12	0.25
Streptococcus agalactiae (16) Spain	Ulifloxacin	0.25 to 1	0.25	1
	Ciprofloxacin	0.5 to 2	1	1
	Levofloxacin	1 to 2	1	1
	Moxifloxacin	0.25	0.25	0.25

Data taken from [31,32]. MIC_x: Minimum concentration required to inhibit the growth of x% of the tested microorganisms.

The antimicrobial activity of ulifloxacin was also assessed with minimum bactericidal concentration (MBC) tests. MBC_{90} is the minimum concentration required to kill 90% of the tested organisms. If the ratio MBC/MIC exceeds 32, the strain is considered tolerant and the drug bacteriostatic. Ulifloxacin showed bactericidal activity at concentrations easily reachable *in vivo* against community-associated *E. coli* and *Klebsiella* strains, all *Proteus* spp., *H. influenzae*, *Providencia* and *Morganella* strains [31]. Conversely, against hospital-associated *E. coli* and *Klebsiella* strains, the bactericidal activity of ulifloxacin (MBC/MIC = 2–4) was exhibited at a higher concentration than susceptibility breakpoint (MBC_{90} = 16 and 4 mg/l, respectively). Against *P. aeruginosa* strains isolated in Italy, ulifloxacin was not bactericidal. As for Gram-positive bacteria, ulifloxacin was bactericidal against methicillin-susceptible *S. aureus* strains and *S. pneumoniae* and *S. pyogenes* strains isolated in Italy [31].

To study the bactericidal activity in correlation with time, time–kill curve assays have been developed. Montanari *et al.* assessed ulifloxacin compared with ciprofloxacin against ciprofloxacin-susceptible and -resistant *E. coli* and *Proteus mirabilis* [31]. At concentrations two- and fourfold higher than MIC, ulifloxacin showed a bactericidal activity, defined as a reduction by 3 log_{10} or more, in the inoculum concentration, at 4 and 8 h of incubation. The same bactericidal potency could be demonstrated for ciprofloxacin against susceptible strains. Roveta *et al.* found that ulifloxacin tested against strains of *P. aeruginosa* had a faster and more potent bactericidal activity compared with ciprofloxacin and levofloxacin [34]. In time–kill curve experiments, the bactericidal effect for ulifloxacin occurred as early as 2 h after incubation and was maintained for the entire duration of the test (i.e., 24 h), with a reduction in the inoculum concentration constantly exceeding 3 and reaching 5 log_{10}.

Mutant prevention concentration (MPC) is the minimum concentration capable preventing the growth of the least susceptible microorganism in a high-density bacterial population, such as inocula in excess of 108 colony forming unit/ml [38]. From a theoretical standpoint, the most potent molecule (i.e., the one with the lowest MIC) is also the least likely to determine selection of resistant mutants. A high MPC represents a high probability to select resistant mutant during monotherapy of severe infections. Notably, when tested against *P. aeruginosa*, ulifloxacin produced the lowest MPC values (≤4 mg/l) compared with ciprofloxacin and levofloxacin [35]. Ulifloxacin produced lower or equal MPC values than ciprofloxacin and levofloxacin also when tested against 30 *E. coli* strains isolated from patients with a UTI [39].

In vivo activity

In representative mouse models of systemic infection [30,40], respiratory tract infection [30] and UTI [30,41], prulifloxacin showed similar or superior efficacy against Gram-negative or Gram-positive bacteria compared with reference quinolones.

Specifically, in a model of systemic infection [40], the dose required to protect 50% of the infected mice (effective dose 50% [ED_{50}]) for prulifloxacin was similar to the ED_{50} for ciprofloxacin and ofloxacin against *S. aureus*, *E. coli* and *K. pneumoniae*, while it was approximately two- to fivefold lower than the ED_{50} for ciprofloxacin and ofloxacin against *S. pneumoniae* and *P. aeruginosa*. In a model of respiratory tract infection, ED_{50} for prulifloxacin was up to twofold lower than ED_{50} for ciprofloxacin and ofloxacin against *K. pneumoniae* [30]. In models of UTI, the efficacy of prulifloxacin, measured as bacterial count reduction, was superior to that of ofloxacin and levofloxacin against *E. coli* and *P. aeruginosa* [30,41].

Clinical efficacy

The clinical efficacy of prulifloxacin has been demonstrated in several Phase III trials, of which the results are either fully published or available as data on file provided from the manufacturing or licensee companies.

Acute exacerbation of chronic bronchitis

Prulifloxacin 600 mg once daily was compared with ciprofloxacin 500 mg twice daily, both administered for 10 days, in 235 adult patients with acute exacerbation of chronic bronchitis (AECB) in a multicenter, randomized, double-blind, double-dummy study [42]. Clinical success assessed at the end-of-treatment visit was observed in 84.7% (95% confidence interval [CI]: 78–91.4) and 85% (95% CI: 78.4–91.5) of the patients in prulifloxacin and ciprofloxacin groups, respectively. Similar proportions of patients in the two groups achieved clinical cure (15.3 vs 11.5%) or improvement (69.4 vs 73.5%). The microbiological activity of the two drugs was similar compared with the most frequently isolated strains including *H. influenzae*, *S. pneumoniae*, *K. pneumoniae* and *P. aeruginosa*. In this trial, patients with *S. pneumoniae* infections treated with prulifloxacin had a higher

eradication rate than ciprofloxacin (8/9, 89% vs 9/12, 75%), but the small sample does not allow definitive conclusions to be drawn.

In an additional multicenter, randomized, double-blind, double-dummy study, prulifloxacin 600 mg once daily was compared with 1000 mg twice-daily amoxicillin/clavulanic acid, both administered for 10 days, in 214 adult patients with AECB [Grassi *et al.*: Evaluation of efficacy and tolerability versus amoxi clavulanic acid in the treatment of acute exacerbations of chronic bronchitis. Rome, Italy, 27th May 1998. Data on file]. No significant difference was found for both clinical success and microbiological eradication rate between the two groups at the end-of-treatment visit: 92.5% (95% CI: 87.4–97.5) for prulifloxacin versus 93.4% (95% CI: 88.6–98.1) for amoxicillin/clavulanic acid.

Acute uncomplicated lower UTIs

Prulifloxacin 600 mg was compared with pefloxacin 800 mg single doses in a multicenter, randomized, open-label trial including 239 women with acute simple cystitis [43]. Microbiological eradication rate was similar in the groups (97.4 vs 92.2%, respectively), as well as clinical efficacy rate at 5–7 days after treatment, with only 7.8 and 15.7% failures in the prulifloxacin and pefloxacin-treated patients, respectively. These results were sustained at follow-up visit on post-treatment week 4.

Prulifloxacin 600 mg was also tested against ciprofloxacin 500 mg single doses in a multicenter, randomized, double-blind, double-dummy trial including 251 women with uncomplicated lower UTI [Zagni *et al.*: Double-blind, controlled study of prulifloxacin in comparison with ciprofloxacin in the treatment of acute uncomplicated lower urinary tract infection in women. Rome, Italy, 27th June 1998. Data on file]. Microbiological eradication at 5–7 days and at 30 days after treatment was achieved in 97.2 versus 97.2% (95% CI: 94–100 for both) and in 95.2% (95% CI: 91.2–99.3) versus 95.4% (95% CI: 91.4–99.3) of patients treated with prulifloxacin and ciprofloxacin, respectively. Clinical success rate at first and second follow-up visit was similar: 98.1 (95% CI: 95.5–100) and 97.1% (95% CI: 94–100) for prulifloxacin versus 98.2 (95% CI: 95.6–100) and 96.3% (95% CI: 92.7–99.9) for ciprofloxacin, respectively.

Complicated lower UTIs

Prulifloxacin 600 mg once daily was compared with ciprofloxacin 500 mg twice daily, both administered for 10 days, in a multicenter, randomized, double-blind, double-dummy study including 257 patients with complicated lower UTIs [44]. Complicated UTI was defined as the presence of an indwelling catheter, intermittent catheterization, post-void residual urine more than 50 ml, benign prostate hyperplasia, ureteral obstruction, or other urological conditions characterized by any combination of dysuria, urgency, frequency or fever. Microbiological eradication rate was significantly higher with prulifloxacin than with ciprofloxacin at 5–7 days after end of treatment (90.8 vs 77.8%; p = 0.008), whereas it was similar (75.3 vs 72.2%) at post-treatment week 4. At the first follow-up visit, clinical success rate was 94.8% for prulifloxacin and 93.3% for ciprofloxacin.

Microbiological and clinical results of a multicenter, randomized, open-label study comparing prulifloxacin 600 mg once daily with amoxicillin/clavulanic acid 1000 mg twice daily, both administered for 10 days, in 225 patients with complicated lower UTI were similar in both treatment groups [Defidio *et al.*: Efficacy and tolerability of prulifloxacin versus amoxi clavulanic acid in the treatment of complicated urinary tract infections, Rome, Italy, 27th May 1998. Data on file]. The rate of microbiological eradication on post-treatment week 1 and 4 were 93.1 (95% CI: 88.2–98) and 95.1% (95% CI: 90.9–99.3) for prulifloxacin, and 93.5 and 93.7% for amoxicillin/clavulanic acid, respectively, with no significant difference. The clinical success rate with either treatment at first follow-up visit was comparable: 96.1% (95% CI; 92.3–99.8) for prulifloxacin versus 97.1% (95% CI: 93.8–100) for amoxicillin/clavulanic acid.

Chronic bacterial prostatitis

In a recent single-center, randomized, double-blind trial, 96 patients with chronic bacterial prostatitis (CBP) and evidence of ongoing infection were randomized to receive a 4-week oral course of either prulifloxacin 600 mg or levofloxacin 500 mg once daily [45]. The rationale for testing prulifloxacin in CBP patients was based on the above mentioned *in vitro* data showing that ulifloxacin has a greater penetration within bacteria than other fluoroquinolones [27] and within macrophages and polymorphonuclear neutrophils, thereby killing bacteria directly or rendering them more prone to phagocytic action [26]. Both are highly desirable properties in the CBP setting, since most antibiotics are known to have a low diffusion capability into chronically inflamed/infected prostate ducts. In addition, ulifloxacin has been found to stimulate *in vitro* the synthesis of several cytokines, such

as IL-6, IL-8 and TNF-α [46], which may play a key role in mediating and possibly antagonizing the infective process in the prostate *in vivo* by enhancing the local immune response.

In this study [45], all men diagnosed with CBP were evaluated with the Meares-Stamey test and the National Institutes of Health Chronic Prostatitis Symptom Index at baseline and 1 week after therapy completion. Patients with microbiological eradication were evaluated again with the Meares-Stamey test 6 months after end of treatment. Microbiologic eradication rate was 72.73% for prulifloxacin and 71.11% for levofloxacin ($p = 0.86$), and the reduction in the post-treatment questionnaire score was 10.75 and 10.73, respectively ($p = 0.98$). Furthermore, a lower, albeit not significant ($p = 0.1$), proportion of patients treated with prulifloxacin had recurrent prostate infection at 6-month follow-up visit compared with levofloxacin. Based on this trend, one may speculate that the active metabolite, by means of its putative direct action on the chemotaxis of neutrophils and other phagocytes, is able to create an unfavorable milieu against persisting or re-infecting bacteria.

Diabetic foot infection

In a nonrandomized, noncomparative, open-label pilot study including 60 patients with diabetic foot infection of variable severity [47], prulifloxacin 600 mg once daily administered either alone for at least 15 days or in combination with teicoplanin (against methicillin-resistant *S. aureus*) or metronidazole (against anaerobes) for at least 40 days was able to cure all cases with skin and/or soft tissue infection and 86% of those with osteomyelitis, respectively. Randomized controlled trials of adequate power are eagerly awaited to confirm these encouraging preliminary results.

Food & drug interactions

Similarly to other fluoroquinolones, but to a lesser degree, prulifloxacin affects the disposition of theophylline. Fattore *et al.* [20] found that the area under the curve and elimination half-life of theophylline increased by approximately 15%, and the apparent oral clearance decreased by approximately 15%, when prulifloxacin 600 mg was administered once daily for 8 days with coadministration of theophylline 6 mg/kg on days 1 and 7 in young healthy volunteers. The remaining pharmacokinetic characteristics were unaltered.

Although the interaction between the two drugs appears to be clinically negligible, monitoring of theophylline levels is recommended during concomitant administration of prulifloxacin, especially in subjects with metabolic disorders [Angelini ACRAF SpA investigator's brochure. Rome, Italy, 20 April 2004. Data on file].

The absorption of prulifloxacin is reduced when milk is taken [17], or cimetidine, aluminum-, magnesium- and calcium-containing antacids and iron-containing supplements are coadministered or administered up to 3 h before or up to 2 h after prulifloxacin [18].

Concurrent administration of probenecid and prulifloxacin increased systemic exposure to ulifloxacin by 46%, prolonged its elimination half-life by 60% and reduced its apparent total clearance and urinary excretion by 30 and 57%, respectively, suggesting that renal excretion of ulifloxacin occurs via an active tubular secretion mechanism, besides glomerular filtration [19].

Safety & tolerability

Fluoroquinolones are commonly used worldwide, even in children and elderly people, and have a relatively favorable side-effect profile [48]. Typically, adverse events involve the gastrointestinal tract, CNS and skin, and are generally of mild-to-moderate severity and reversible after treatment withdrawal [48]. Nevertheless, patients receiving fluoroquinolones have to be closely monitored, because some compounds of this class have been associated with some form of toxicity. Sparfloxacin was associated with a high rate of phototoxicity, temafloxacin and grepafloxacin were withdrawn from the market due to cardiotoxicity, and trovafloxacin was restricted because of unexpected severe hepatotoxicity [49]. Furthermore, with the use of ciprofloxacin, gemifloxacin, moxifloxacin and levofloxacin, there remain some concerns regarding the increased risk of developing tendinopathy and tendon rupture [50]. Finally, a recent alert has been given regarding acute fulminant hepatitis and Stevens–Johnson syndrome caused by moxifloxacin [51].

Prulifloxacin was generally well-tolerated in all clinical trials, even when administered with long-course schedules, and there were no significant differences in the incidence, type and severity of treatment-related adverse events in single- or repeat-dose comparisons with the reference antimicrobials. The most frequently reported side effects were gastric disturbances, diarrhea, nausea and skin rash of mild-to-moderate severity. Adverse events noted in clinical trials are detailed in Table 3.

With regard to potential cardiotoxicity, results of *in vitro* [52] and *in vivo* [52,53] studies suggest that the QT interval is unlikely to be prolonged during continued therapy with prulifloxacin.

Specifically, like ciprofloxacin and unlike moxifloxacin, application of ulifloxacin produced only a minor decrease in current amplitude of the human ether-a-go-go-related gene potassium channel in stably transfected human embryonic kidney cells [52]. Furthermore, oral administration of prulifloxacin 150 mg/kg once daily for 5 days to conscious dogs monitored by telemetry had no effect on PR, QT and corrected QT intervals [52]. Similarly to levofloxacin, a continuous intravenous infusion of ulifloxacin 4 mg/kg/min in anesthetized rabbits did not prolong the QT interval or induce cardiac arrhythmias [53].

The phototoxic potential of prulifloxacin appeared to be low and similar to that of ciprofloxacin. In an *ad hoc* randomized, single-blind, crossover trial including 20 healthy Caucasian volunteers exposed to ultraviolet A irradiation before and after oral administration of prulifloxacin 600 mg once daily or ciprofloxacin 500 mg twice daily for 8 days, no phototoxic reaction was observed in seven out of ten subjects in each treatment group [Angelini ACRAF SpA investigator's brochure. Rome, Italy, 20th April 2004. Data on file].

Similarly to ciprofloxacin, but to a lesser degree, prulifloxacin induced convulsions in mice only when fenbufen, a nonsteroidal anti-inflammatory drug, was coadministered [54].

Prulifloxacin is devoid of side effects on the central, peripheral and autonomic nervous system, and on the genitourinary tract [55,56]. It was the possible cause of acute renal failure in a single reported case [57].

No hepatotoxicity, chondrotoxicity and *Clostridium difficile*-associated enteritis have been reported with its use so far.

Regulatory affairs

Prulifloxacin was first approved in 2002 in Japan to treat a wide range of bacterial infections, including infectious diarrhea [58].

In 2005, after application by the licensee company Angelini (Angelini ACRAF S.p.A., Rome, Italy), it was approved through a Mutual Recognition Procedure for the treatment of acute uncomplicated lower UTIs as a single 600 mg dose, and of AECB and complicated lower UTIs as a once daily 600 mg dosage for a maximum of 10 days [102]. It is currently licensed in Italy, Portugal, Greece, Austria, Hungary, Czech Republic, Slovak Republic and Poland, and marketed in Italy, Portugal and Greece. The licensee company Optimer (Optimer Pharmaceuticals Inc., CA, USA) has promoted two Phase III randomized, double-blind, placebo-controlled trials in adult travelers with acute bacterial gastroenteritis. The first trial was conducted in Mexico and Peru, the second trial is being conducted in India, Guatemala and Mexico, in order to apply for licensing for infectious diarrhea by the US FDA [103].

Conclusion

Prulifloxacin is a new oral fluoroquinolone that has high bactericidal activity against a wide spectrum of Gram-negative and Gram-positive pathogens. It is apparently the most potent *in vitro* fluoroquinolone agent against *P. aeruginosa* to date, but this appealing activity remains to be confirmed by *in vivo* studies.

In Phase III randomized clinical trials, prulifloxacin appeared as effective as other fluoroquinolones (ciprofloxacin, pefloxacin and levofloxacin) and amoxicillin/clavulanic acid in the treatment of AECB, acute uncomplicated and complicated lower UTIs and CBP.

Prulifloxacin is well-tolerated, with a similar safety profile to that of other reference drugs of the quinolone class. Adverse events are generally of mild-to-moderate severity, with gastric pain, diarrhea, nausea and skin rash being the most frequent.

Table 3. Safety data reported in clinical trials testing once-daily prulifloxacin 600 mg.

Study	No. patients receiving prulifloxacin	Duration of treatment (days)	Serious adverse events (%)	Nonserious adverse events (%)	Withdrawal rate (%)	Ref.
Grassi *et al.*	117	10	0	16.2	0.9	[42]
Grassi *et al.*	107	10	0	16.8	0.9	[Data on file]
Cervigni *et al.*	121	1	0	1.6	0	[43]
Zagni *et al.*	126	1	0	4.8	0	[Data on file]
Carmignani *et al.*	127	10	0	10.2	0.8	[44]
Defidio *et al.*	113	10	0	7.9	0	[Data on file]
Giannarini *et al.*	48	28	0	16.7	4	[45]
Cavani and Paura	60*	15–40	0	NR	0	[47]

Note that only treatment-related adverse events and withdrawal events occurred in the intention-to-treat population are counted.
**Either alone or in combination with teicoplanin or metronidazole.*
NR: Not reported.

New indications are currently being planned to be explored in additional well-designed and adequately powered clinical trials.

Future perspective

Whilst a number of studies have proven the efficacy of fluoroquinolones against a variety of pathogens causing gastrointestinal, respiratory and urinary tract infections, there is growing interest in the development of new molecules of the class in order to further widen the spectrum of activity, increase the antimicrobial potency and overcome the emergence of bacterial resistance. However, large-scale marketing of new fluoroquinolones might be hampered by the potentially unfavorable safety profile, thus the currently available, more tolerable drugs are likely to remain the mainstay of cure. Prulifloxacin is one such compound, holding a promising role in the treatment of both community-acquired, nosocomial and critical infections. *In vitro* studies have indicated prulifloxacin as the most potent quinolone agent and the one with the lowest potential of inducing the emergence of mutant resistant strains, when tested against *E. coli* and *P. aeruginosa* and compared with ciprofloxacin and levofloxacin [34,35,39]. Since antibiotic resistance is expected to further increase in the next few years, this molecule will keep its present prominent role in preventing the emergence of resistance among Gram-negative microorganisms and will, possibly, hold a new position in the growing antimicrobial armamentarium against the most problematic MDR pathogens. In addition, the possibility for once-daily administration and the good tolerability are further elements of appeal, which may contribute to a higher patient compliance.

Although several clinical trials have been successfully accomplished, prulifloxacin should be further investigated in the field of respiratory infections (community-acquired and nosocomial pneumonia, where *P. aeruginosa* is most probable), recurrent lower UTIs (such as chronic prostatitis), diabetic foot infections, and skin and soft tissue infections. Finally, the preliminary successful results obtained in the ongoing trials on infectious diarrhea let one hypothesize that prulifloxacin will play a leading role even in the treatment of acute bacterial gastroenteritis, which remains one of the leading causes of morbidity and mortality worldwide [103].

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Executive summary

Mechanism of action

- Like other fluoroquinolones, ulifloxacin inhibits DNA gyrase and topoisomerase IV enzymes, thereby preventing DNA replication, transcription, repair and recombination.

Pharmacokinetic properties

- Prulifloxacin is rapidly and quantitatively metabolized to ulifloxacin by first-pass metabolism, shows good penetration into several body tissues and fluids, and possesses a long elimination half-life (approximately 10 h).

Clinical efficacy

- In Phase III randomized clinical trials, prulifloxacin appeared as effective as other fluoroquinolones (ciprofloxacin, pefloxacin and levofloxacin) and amoxicillin/clavulanic acid in the treatment of acute exacerbation of chronic bronchitis, acute uncomplicated and complicated lower urinary tract infections, and chronic bacterial prostatitis.

Safety & tolerability

- Prulifloxacin has a good safety profile. Adverse events are generally of mild-to-moderate severity, with gastric pain, diarrhea, nausea and skin rash being the most frequent.

Drug interactions

- Prulifloxacin causes only a weak alteration in the disposition of theophylline. Its absorption is reduced when milk is taken, or cimetidine, antacids and iron-containing supplements are coadministered.

Dosage & administration

- Prulifloxacin is available in 600-mg oral tablets. A single dose is recommended to treat uncomplicated lower urinary tract infections, whereas a once-daily 10-day course is to be given in complicated lower urinary tract infections and acute exacerbation of chronic bronchitis. In chronic bacterial prostatitis, a 4-week regimen is suggested.

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