Future MICROBIOLOGY

DRUG EVALUATION

Delafloxacin, a non-zwitterionic fluoroquinolone in Phase III of clinical development: evaluation of its pharmacology, pharmacokinetics, pharmacodynamics and clinical efficacy

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ABSTRACT Delafloxacin is a fluoroquinolone lacking a basic substituent in position 7. It shows MICs remarkably low against Gram-positive organisms and anaerobes and similar to those of ciprofloxacin against Gram-negative bacteria. It remains active against most fluoroquinolone-resistant strains, except enterococci. Its potency is further increased in acidic environments (found in many infection sites). Delafloxacin is active on staphylococci growing intracellularly or in biofilms. It is currently evaluated as an intravenous and intravenous/oral stepdown therapy in Phase III trials for the treatment of complicated skin/skin structure infections. It was also granted as Qualified Infectious Disease Product for the treatment of acute bacterial skin and skin structure infections and community-acquired bacterial pneumonia, due to its high activity on pneumococci and atypical pathogens.

KEYWORDS
• anaerobes • delafloxacin
• fluoroquinolone
• gonorrhea • MRSA
• pneumonia • skin and skin structure infection
• *Staphylococcus aureus*
• *Streptococcus pneumoniae*

In an era of increasing bacterial resistance, clinicians start facing situations in which therapeutic options become worryingly scarce. The so-called ESKAPE pathogens (*Enterococcus faecium*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Acinetobacter baumanii*, *Pseudomonas aeruginosa*, *Enterobacter* spp.), pose specific problems as they escape the activity of most antibiotics [1]. Multidrug resistant strains in these species become highly prevalent in some regions such as Southeast Asia [2] and start to spread all over the world [3]. In this context, clinicians are forced to resort to old drugs for which resistance is still low because they were long neglected [4]. Yet, some of these molecules, like polymyxins, are toxic [5]. Moreover, most of them have been registered before modern pharmacokinetics/pharmacodynamics concepts were developed, so that optimal dosing is not established, making probably resistance development ineluctable [6]. Thus, finding new therapeutic options is urgently needed. A few novel molecules acting on still unexploited targets are now in preclinical or clinical development [7] but most successful efforts were oriented to the search of new molecules with optimized pharmacological profile in existing classes of drugs [8–10].

Quinolones represent one of the few totally synthetic antibiotic classes. For this reason a lot of chemical modifications have been performed on the quinolone core in order to modulate their spectrum of activity, pharmacokinetic profile, or toxicity [11,12]. Delafloxacin is one of the quinolones emerging from these studies. It is currently in Phase III of clinical development for the treatment of complicated acute bacterial skin and skin structure infections. This review describes the pharmacological profile of this drug and summarizes the clinical data available so far.

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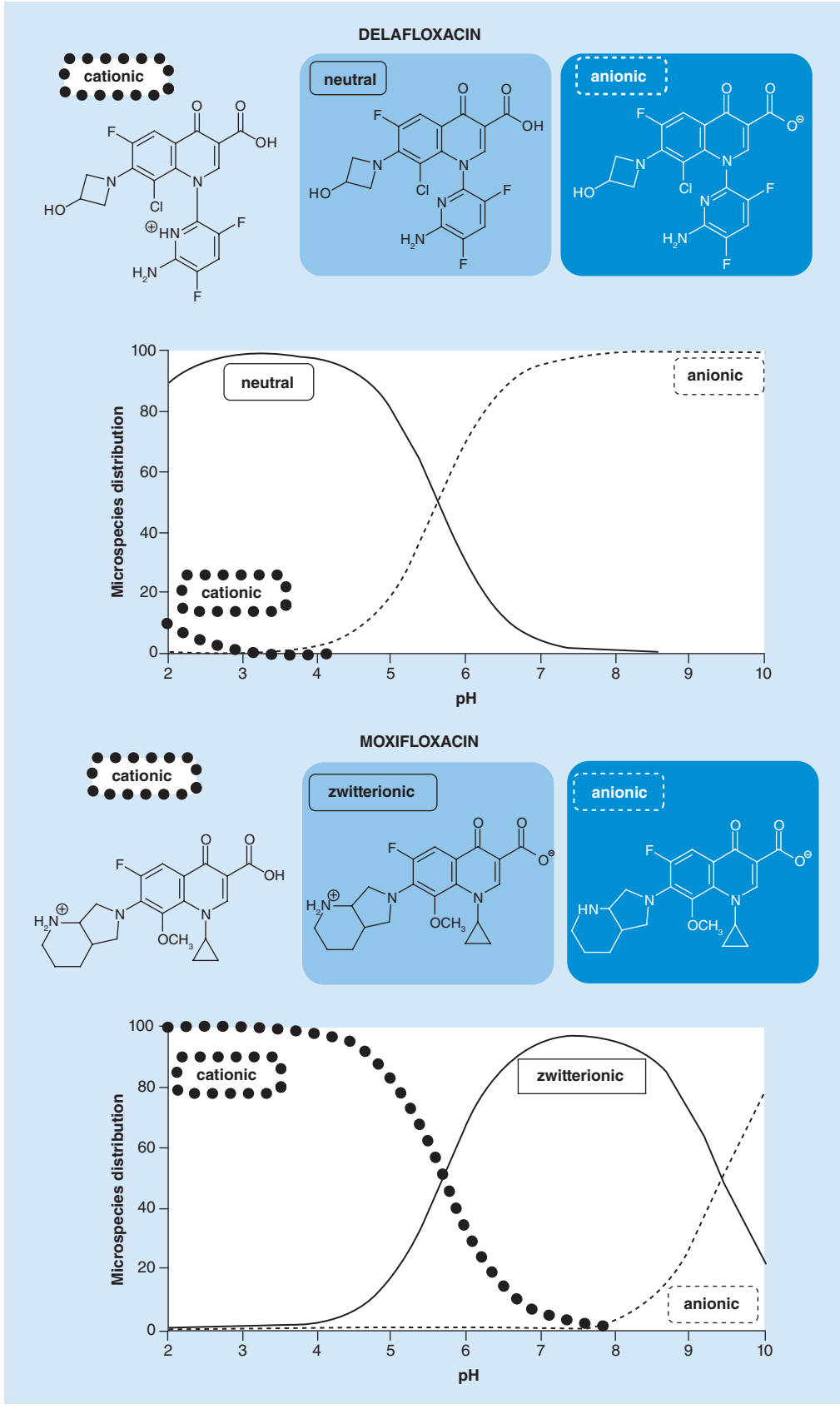
DELAFLOXACIN
cationic
neutral
anionic
Microspecies distribution
pH
MOXIFLOXACIN
cationic
zwitterionic
anionic
Microspecies distribution
pH

Figure 1. Proportions of molecular microspecies for delafloxacin (top) and moxifloxacin (bottom) as a function of the pH (see facing page). Due to the absence of protonable substituant in position 7, delafloxacin is neutral at acidic pH and anionic at physiological pH. To the opposite, moxifloxacin, is found in its protonated form at acidic pH but is zwitterionic at physiological pH. Calculations were made using Reaxys software; Elsevier (2014).

History of delafloxacin

Delafloxacin (CAS registry number 189279-58-1) was described as WQ-3034 by Wakunaga Pharmaceutical Co., Ltd., Osaka & Hiroshima, Japan [13]. It was first licensed in 1999 to Abbott Park, IL, and further developed as ABT-492. Its excellent activity was documented at that time against *Staphylococcus aureus* [14], *Streptococcus pneumoniae* [14–16], β-hemolytic streptococci [16], some strains of *Enterococcus faecalis* and *faecium* [16], *Haemophilus influenzae* [14,16,17], *Moraxella catarrhalis* [14,16], *Neisseria gonorrhoeae* [16,18], *Neisseria meningitidis* [16], *Legionella* spp [18], *Chlamydia pneumoniae* [19], mycoplasmas and ureaplasmas [20], *Helicobacter pylori* [18], enterobacteriaceae [14,16], *Pseudomonas aeruginosa* to some extent [16,18], *Yersinia pestis* [21], anaerobes [14,16] and *Mycobacterium tuberculosis* including its intracellular forms [22]. Globally, it was considered as more potent against fluoroquinolone-susceptible or resistant Gram-positive organisms than other fluoroquinolones with a spectrum oriented towards Gram-positive organisms (trovafloxacin, moxifloxacin, gatifloxacin and gemifloxacin) and as less potent against Gram-negative organisms than ciprofloxacin [23]. In 2006, the compound was licensed to Rib-X Pharmaceuticals Inc. (now Melinta Therapeutics, CT, USA), where it received the code name RX-3341.

Pharmacology of delafloxacin

Delafloxacin is a dual-targeting fluoroquinolone, capable of forming cleavable complexes with DNA and topoisomerase IV or DNA gyrase and of inhibiting the activity of these enzymes in both Gram-positive and Gram-negative bacteria [18]. In contrast to other molecules like ciprofloxacin, however, it shows a similar affinity for both types of enzymes [18], which may explain its broad spectrum of activity. Intriguingly, delafloxacin is not a more potent inhibitor of topoisomerases than other fluoroquinolones [18] though it shows greater potency, especially against Gram-positive organisms. It has, therefore, been suggested that this improved activity was due to the conjunction of three unique features in this molecule, namely the presence of a heteroaromatic substituent at position 1, a weak polarity associated to the presence of a chlorine in position 8 and a lack of basic group in position 7 [24]. The absence of protonable substituent in position 7 gives to delafloxacin an anionic character at neutral pH, which is unusual for a fluoroquinolone. **Figure 1** shows the ionization status of the molecule according the pH in comparison with that of moxifloxacin. At physiological pH (~7–7.4), delafloxacin is mainly found as an anion, but at slightly acidic pH (≤5.5), the majority form is uncharged, the calculated pKa of the carboxylic function being 5.6 (using Reaxys software; Elsevier, 2014). On the contrary, moxifloxacin is present mainly as a cation at pH lower than 5.5 and as a zwitterion at higher values. This specific characteristic may explain why delafloxacin accumulates much more in bacteria at acidic pH [25], as the nonionized form of a drug is considered as more diffusible though biological membranes. This feature, therefore, contributes to justify why delafloxacin potency is highly improved in acidic environments, in contrast with what is observed for other fluoroquinolones. For *S. aureus,* MICs decrease of up to 5–7 dilutions when measured at pH 5.5 versus 7.4, reaching values as low as 0.00003 mg/l. On the same pH range, moxifloxacin MICs increase of 2–3 dilutions [25,26]. For *E. coli*, *K. pneumoniae* and *P. aeruginosa*, delafloxacin MICs are 4–6 dilutions lower at pH 5.5 than at pH 7.4, while those of ciprofloxacin are 3–8 dilutions higher [26,27].

Resistance to fluoroquinolones is primarily caused by target mutations, which occur in general in the primary target enzyme (thus more often in GyrA subunit of DNA gyrase in Gram-negative bacteria; ParC subunit of topoisomerase IV in Gram-positive bacteria, except *S. pneumoniae* [12,28]). Active efflux also contributes to decrease susceptibility, with narrow spectrum transporters expressed in Gram-positive pathogens and conferring resistance to fluoroquinolones only (like NorA, NorB, NorC, MdeA or the plasmid-encoded QacA and QacB [29–31] in *S. aureus,* or PmrA and the heterodimer PatA/

Table 1. Susceptibility of relevant Gram-positive pathogens to delafloxacin and other commercially available fluoroquinolones.

Species	Phenotype	Number of strains	Antibiotic	MIC_{50} (mg/l)	MIC_{90} (mg/l)	MIC range (mg/l)	Ref.[†]
S. aureus	All	681	Levofloxacin	0.12	>32	0.03–>32	[41]
		681	Delafloxacin	0.12	0.5	≤0.004–16	[41]
	FQ-S	70	Levofloxacin	0.25	0.5	0.06–0.5	[23]
		88		0.12	0.25	0.06–1	[42]
		70	Moxifloxacin	0.06	0.1	0.015–0.5	[23]
		70	Delafloxacin	0.004	0.008	0.002–0.008	[23]
		88		0.002	0.008	≤0.001–0.06	[42]
	FQ-R	71	Levofloxacin	16	32	4–64	[23]
		100		4	8	2–32	[42]
		71	Moxifloxacin	4	8	0.25–16	[23]
		71	Delafloxacin	0.25	1	0.015–1	[23]
		100		0.006	0.12	0.015–2	[42]
S. epidermidis	FQ-S	9	Levofloxacin		0.25	0.12–0.5	[23]
		9	Moxifloxacin		0.12	0.03–0.12	[23]
		9	Delafloxacin		0.008	0.002–0.08	[23]
	FQ-R	10	Levofloxacin	16	16	4–128	[23]
		10	Moxifloxacin	2	2	1–>128	[23]
		10	Delafloxacin	0.5	0.5	0.12–1	[23]
Coagulase-negative staphylococci	All	19	Levofloxacin	0.12	>32	0.06–>32	[42]
		19	Delafloxacin	0.004	1	0.001–2	[42]
	FQ-R	10	Levofloxacin	8	64	4–128	[18]
		10	Delafloxacin	0.25	0.5	0.03–0.5	[18]
β-hemolytic staphylococci	All	17	Levofloxacin	0.5	2	0.03–2	[42]
		17	Delafloxacin	0.008	0.015	≤0.002–0.015	[42]
S. pneumoniae	FQ-S	69	Levofloxacin	1	1	0.5–2	[23]
		69	Moxifloxacin	0.12	0.12	0.06–0.25	[23]
		69	Delafloxacin	0.008	0.015	0.004–0.015	[23]
	FQ-R	33	Levofloxacin	16	32	2–32	[23]
		33	Moxifloxacin	2	4	0.25–8	[23]
		33	Delafloxacin	0.12	0.5	0.015–0.5	[23]
E. faecalis	FQ-S	18	Levofloxacin	1	1	0.5–2	[18,23]
		18	Moxifloxacin	0.25	0.5	0.12–0.5	[23]
		18	Delafloxacin	0.06	0.06	0.03–0.12	[23]
	FQ-R	26	Levofloxacin	32	128	16–128	[23]
		26	Moxifloxacin	8	32	2–64	[23]
		26	Delafloxacin	0.25	8	0.06–32	[23]
E. faecium	FQ-S	14	Levofloxacin	1	4	0.5–4	[23]
		14	Moxifloxacin	1	2	0.12–4	[23]
		14	Delafloxacin	0.12	1	0.06–2	[23]
	FQ-R	28	Levofloxacin	32	64	8–>128	[23]
		28	Moxifloxacin	16	16	1–32	[23]
		28	Delafloxacin	4	8	0.25–16	[23]
C. difficile	All	12	Levofloxacin	2	4	2–4	[18]
		12	Delafloxacin	≤0.015	≤0.015	≤0.015	[18]

[†]Comparison of MIC distributions among antibiotics should be performed using data from a same bibliographic reference.
FQ-S: Fluoroquinolone susceptible; FQ-R: Fluoroquinolone resistant (based in most cases on CLSI susceptibility breakpoints for marketed comparators).

PatB in *S. pneumoniae* [32,33]), and broad-spectrum transporters expressed in Gram-negative pathogens and conferring cross-resistance to several antibiotic classes [34]. More anecdotal resistance mechanisms include the production of the protein Qnr, which impairs the binding of fluoroquinolones to DNA [35], or of a AAC(6')-Ib-cr enzyme originally inactivating

aminoglycosides, which N-acetylates fluoroquinolones with a piperidine substituent in position 7 (norfloxacin and ciprofloxacin) [36].

Delafloxacin maintains activity against strains showing resistance to other fluoroquinolones, as exemplified in studies comparing MIC distribution of delafloxacin and other fluoroquinolones against fluoroquinolone-resistant isolates of staphylococci, streptococci, enterococci, *H. influenzae, M. catarrhalis* and a series of enterobacteriaceae [18,23,25,37]. Although delafloxacin is affected by target modifications/mutations, its intrinsic activity is so much higher than that of other fluoroquinolones that the elevations in MICs are not as impactful. Delafloxacin is also claimed to be a poor substrate for efflux pumps from Gram-positive bacteria [37] and for the NorM efflux pump extruding fluoroquinolones out of *N. gonorrheae* [38].

The capacity of delafloxacin to select resistance has been studied in MRSA *in vitro* [37] and was found infrequent (10^{-9} to 10^{-11}). Moreover, concentrations preventing the selection of mutations (MPC [39]) are low, ranging from one to four times the initial MIC, and globally 8–32-fold lower than for other quinolones. Preliminary data also confirm a low capacity of selecting resistance in *N. gonorrheae*, with MIC of the resistant isolates only four times higher than initial values [40]. The equilibrated dual-targeting ability of delafloxacin may contribute to reduce resistance selection, as a mutation in each enzyme would be necessary to significantly affect susceptibility [37].

Antibacterial activity

MIC distributions of delafloxacin are illustrated in **Tables 1** and **2** for a series of human pathogens, in comparison with those of marketed fluoroquinolones. Delafloxacin shows very low MIC against Gram-positive pathogens **(Table 1)**, with MIC being typically two-to-fourfold lower than those of moxifloxacin, considered nowadays as the most potent anti-Gram-positive fluoroquinolone on the market [12]. At acidic pH, the difference in potency between the two antibiotics can even reach seven dilutions [25], as explained before. Noteworthy, delafloxacin MICs remain low against bacteria resistant to fluoroquinolones (levofloxacin, moxifloxacin), with maximal values of 2 mg/l, 1 mg/l and 0.5 mg/l observed, respectively, in *S. aureus*, coagulase-negative staphylococci and *S. pneumoniae,* including in recent surveys [41,42]. Delafloxacin proves also more potent than moxifloxacin or levofloxacin against other respiratory pathogens like *H. influenzae*, *M. catarrhalis*, *L. pneumophila*, or *M. pneumoniae.* Although more potent than other fluoroquinolones against fluoroquinolone-susceptible enterococci, MICs can reach values as high as 32 mg/l in resistant strains. While designed as an anti-Gram positive drug, delafloxacin shows also useful activity against Gram-negative bacteria. Its MICs are particularly low against *N. gonorrheae* or *H. pylori.* In these cases, activity is expected to be even higher *in vivo,* because of the acidic pH prevailing in the vagina [43] or in the stomach. This holds also true for *E. coli* or other enterobacteriaceae when involved in urine infection [27]. Globally, delafloxacin is as active (one dilution difference in MIC) as ciprofloxacin (considered as the most potent commercially available fluoroquinolone against Gram-negative bacteria [12]) against enterobacteriaceae and *P. aeruginosa*. Importantly, it remains more active than ciprofloxacin against ciprofloxacin-resistant enterobacteriaceae. Delafloxacin is also remarkably active against anaerobes, including *C. difficile.*

Pharmacokinetics & pharmacodynamics

Pharmacokinetics of delafloxacin were first studied in healthy volunteers (Phase I trials). After intravenous administration of 300, 450 mg, 600, or 900 mg, C_{max} values (reached after 1 h) were, respectively, 10, 16, 23 and 30 mg/l [44]. AUC were 24, 40, 59 and 92 mg.h/l, indicating that this parameter increased greater than proportionally in that range. More than 30% of the drug was excreted unchanged in urine within the first 12 h after a single dose administration but metabolites have been detected, among which was a glucorono-conjugate [44,45]. Protein binding was estimated at 16% [46]. Half-life ranged from 8 h after administration of 300 mg to 12 h after administration of higher doses. After oral administration of 450 mg [47] or 900 mg [48], C_{max} reached, respectively, 6 mg/l after 0.8 h and 10 mg/l after 1.38 h and AUC were 21 mg.h/l and 44 mg.h/l. In these conditions, an equivalence in total exposure is obtained when administering 300 mg intravenously or 450 mg orally [47]. C_{max} was reduced by 20.5% and T_{max} was delayed by 1.25 h when administered under fed conditions but AUC was not affected [49]. The main pharmacokinetic data are presented for dosages used in clinical studies in **Table 3**.

Table 2. Susceptibility of relevant Gram-negative pathogens to delafloxacin and other commercially available fluoroquinolones.

Species	Phenotype	Number of strains	Antibiotic	MIC_{50} (mg/l)	MIC_{90} (mg/l)	MIC range (mg/l)	Ref.[†]
H. influenzae	FQ-S	110	Levofloxacin	0.015	0.03	0.002–0.5	[23]
		110	Moxifloxacin	0.015	0.06	0.004–0.12	[23]
		110	Delafloxacin	0.005	0.002	≤0.00025–0.004	[23]
	FQ-R	6	Levofloxacin			0.06–8	[23]
		6	Moxifloxacin			0.06–8	[23]
		6	Delafloxacin			0.004–0.5	[23]
M. catarrhalis	FQ-S	50	Levofloxacin	0.03	0.06	0.15–0.25	[23]
		50	Moxifloxacin	0.06	0.06	0.03–0.12	[23]
		50	Delafloxacin	0.002	0.004	0.0005–0.03	[23]
L. pneumophila	All	5	Levofloxacin	0.5	0.5	0.5	[18]
		5	Delafloxacin	0.12	0.12	0.12	[18]
M. pneumoniae	All	18	Levofloxacin	1	2	1–2	[18]
		18	Delafloxacin	0.5	0.5	0.25–0.5	[18]
H. pylori	All	45	Levofloxacin	0.5	0.5	0.12–1	[18]
		45	Delafloxacin	0.03	0.12	0.015–0.12	[18]
N. gonorrheae	All	100	Ciprofloxacin	≤0.015	8	≤0.015–16	[71]
		100	Delafloxacin	0.001	0.06	≤0.0005–0.06	[71]
E. coli	FQ-S	45	Ciprofloxacin	0.015	0.06	0.004–0.25	[23]
		10				0.008–0.25	[26]
		45	Delafloxacin	0.03	0.06	0.04–0.25	[23]
		10				0.016–0.25	[26]
	FQ-R	27	Ciprofloxacin	128	>128	4–>128	[23]
		21				64–>128	[26]
		27	Delafloxacin	4	8	1–16	[23]
		21				2–128	[26]
Enterobacter spp.	FQ-S	20	Ciprofloxacin	0.015	0.03	0.008–0.12	[23]
		20	Delafloxacin	0.06	0.25	0.03–0.25	[23]
	FQ-R	4	Ciprofloxacin			8–16	[23]
		4	Delafloxacin			32–128	[23]
K. pneumoniae	FQ-S	32	Ciprofloxacin	0.06	1	0.015–2	[23]
		32	Levofloxacin	0.12	2	0.03–2	[23]
		32	Delafloxacin	0.12	0.5	0.015–0.5	[23]
	FQ-R	22	Ciprofloxacin	32	64	4–64	[23]
		22	Levofloxacin	16	16	4–32	[23]
		22	Delafloxacin	2	4	1–4	[23]
Acinetobacter spp.	FQ-S	14	Ciprofloxacin	0.25	1	0.015–1	[23]
		14	Delafloxacin	0.12	0.25	≤0.001–0.5	[23]
	FQ-R	14	Ciprofloxacin	128	>128	32–>128	[23]
		14	Delafloxacin	2	16	1–32	[23]
P. aeruginosa	FQ-S	19	Ciprofloxacin	0.12	0.5	0.06–2	[23]
		19				0.25–2	[26]
		19	Delafloxacin	0.25	0.5	0.06–2	[23]
		19				0.016–1	[26]
	FQ-R	21	Ciprofloxacin	32	128	2–128	[23]
		12				64–>128	[26]
		21	Delafloxacin	32	128	1–128	[23]
		12				4–32	[26]

[†]Comparison of MIC distributions among antibiotics should be performed using data from a same bibliographic reference.
FQ-R: Fluoroquinolone resistant (based in most cases on CLSI susceptibility breakpoints for marketed comparators); FQ-S: Fluoroquinolone susceptible.

Table 2. Susceptibility of relevant Gram-negative pathogens to delafloxacin and other commercially available fluoroquinolones (cont.).

Species	Phenotype	Number of strains	Antibiotic	MIC_{50} (mg/l)	MIC_{90} (mg/l)	MIC range (mg/l)	Ref.[†]
B. fragilis	All	16	Levofloxacin	2	2	1–2	[18]
		16	Delafloxacin	0.06	0.12	0.03–0.12	[18]

[†]Comparison of MIC distributions among antibiotics should be performed using data from a same bibliographic reference.
FQ-R: Fluoroquinolone resistant (based in most cases on CLSI susceptibility breakpoints for marketed comparators); FQ-S: Fluoroquinolone susceptible.

Additional pharmacokinetic studies were performed during Phase II trials. In Phase II patients treated for acute bacterial skin and skin structure infections by a daily dose of 300 mg intravenous b.i.d. (*bis in die*), C_{max} were slightly lower (~7 mg/l) but AUC remained similar to that determined in healthy volunteers (21 mg.h/l) [50]. The dose should be reduced from 300 mg to 200 mg IV in case of severe renal insufficiency (eGFR <30ml/min/1.73 m^2) [51].

As other fluoroquinolones, delafloxacin is bactericidal *in vitro* [23,25]. Its efficacy has been documented in a series of animal models treated by delafloxacin or another fluoroquinolone administered at the same dose. In renal abscesses by *S. aureus* it was more effective than moxifloxacin to reduce bacterial burden at doses of 10 or 30 mg/kg [52]. In granuloma pouch infection by Gram-negative bacteria, it was as effective as ciprofloxacin at doses of 25 or 50 mg/kg and reached higher concentration in the pouch despite lower plasma levels [53].

Pharmacodynamic studies claim that the *f*AUC/MIC ratio (where *f*AUC is the AUC for the free drug) is the main driver of efficacy for fluoroquinolones [54]. This also holds true for delafloxacin based on recent animal data obtained in a model of *S. aureus* murine thigh infection, which showed that a 1 log decrease in inoculum can be reached for fAUC/MIC ratios ≤14.3 h [55].

A target value for *f*AUC/MIC of 25–40 h is usually considered as acceptable for Gram-positive infections [54]. Considering that a free AUC of 18 mg.h/l can be reached after administration of 300 mg intravenously [50], a pharmacodynamic susceptibility breakpoint of 0.5 mg/l could be proposed, which would cover most of the strains, including most fluoroquinolone-resistant Gram-positive bacteria except enterococci (see MIC_{90} values in **Table 1**) and anaerobes. This is in accordance with Monte Carlo simulations, which concluded to a >90% target attainment rate for MICs ≤0.5 mg/l upon administration of both 300 and 450 mg b.i.d. [46], rationalizing the doses that have been used in Phase III trials. At this stage, however, PK/PD breakpoints for Gram-negative bacteria have not been determined.

Early studies using *in vitro* pharmacodynamic models also examined delafloxacin properties but neglected to consider the possible role of protein binding. They predicted that delafloxacin could achieve higher bacterial killing against *S. aureus* and *S. pneumoniae* and have a lower propensity to select for resistance than levofloxacin at simulated regimens mimicking AUC achievable in humans [56,57] essentially because higher AUC/MIC ratios could be achieved with delafloxacin (12-times higher against *S. aureus* and 21–63-times higher for *S. pneumoniae* considering the specific strains used in these studies). Delafloxacin was also as effective as ciprofloxacin against *E. coli* at clinically-relevant simulated doses (400 mg delafloxacin vs 500 mg twice daily ciprofloxacin, generating AUC/MIC ratios of 2200 and 1740 h, respectively) but required higher simulated doses (400 mg twice daily) to be at least as effective as ciprofloxacin against *P. aeruginosa* (AUC/MIC ratios of 280 and 120 h for delafloxacin and ciprofloxacin, respectively) [58].

Activity of delafloxacin against persistent forms of infections

For specific bacterial species like staphylococci, the persistent character of infection has been attributed to their capacity to survive intracellularly [59] or to form biofilms [60]. Acting on these specific forms seems thus important in order to avoid recurrence of these infections. In *in vitro* models of intracellular infection by *S. aureus* [25], delafloxacin proved as effective as moxifloxacin despite a lower cell accumulation level (2.5-fold vs sevenfold for moxifloxacin). Yet, its activity was improved when cells are incubated at acidic pH (5.5), related to a tenfold increase in its cellular accumulation level. Cell fractionation studies demonstrated that the bulk of the drug was located in the cytosol [25], as observed for other fluoroquinolones [61,62], which suggests that the drug is able to freely cross biological

Table 3. Pharmacokinetic properties of delafloxacin.[†]

Population	IV 300 mg				Oral 450 mg (volunteers) or 400 mg (patients)			
	C_{max} (mg/l)	AUC_{inf} (mg.h/l)	Half-life (h)	Total clearance (l/h)	C_{max} (mg/l)	AUC_{inf} (mg.h/l)	Half-life (h)	Total clearance (l/h)
Healthy volunteers	10.5 (9.87–11.2)	26.2 (24.7– 27.8)	10.1		5.80 (5.44–6.17)	23.0 (21.6– 24.4)	14.9	
Normal renal function (eGFR 91.9 ± 11.4 ml/min/1.73 m^2)	9.28 ± 2.35	22.6 ± 4.5	9.28 ± 4.33	13.7 ± 2.62	7.16 ± 2.50	25.4 ± 8.01	15.4 ± 6.66	17.6 ± 7.08
Mild renal impairment (eGFR 62.8 ± 7.8 ml/min/1.73 m^2)	9.80 ± 1.09	31.3 ± 6.0	10.7 ± 2.45	9.92 ± 2.01	5.67 ± 1.94	28.3 ± 8.18	12.5 ± 2.73	13.8 ± 9.70
Moderate renal impairment (eGFR 38.6 ± 4.9 ml/min/1.73 m^2)	9.86 ± 2.53	38.4 ± 10.7	8.90 ± 2.98	8.25 ± 1.89	6.00 ± 1.78	37.3 ± 7.03	10.5 ± 4.24	11.0 ± 2.03
Severe renal impairment (eGFR 22.3 ± 6.2 ml/min/1.73 m^2)	19.5 ± 27.3	51.1 ± 20.9	14.2 ± 6.02	6.59 ± 2.07	5.35 ± 1.33	39.5 ± 11.0	15.5 ± 5.23	10.8 ± 2.77
End-stage renal disease (predialysis)	54.4 ± 124.3	84.3 ± 100.5	10.6 ± 5.01	6.58 ± 4.35				

[†]Mean values with confidence interval (healthy volunteers) or standard deviation (others) based on [47,51].

membranes within the cells to gain access to the infected compartments. In *in vitro* models of biofilms, delafloxacin is also one of the most effective drugs among representatives of the main antistaphylococcal classes [63], but its effect on biofilms formed by different clinical isolates depends on the local pH within the biofilm and on its capacity to diffuse within the structure [64]. Additional studies are needed, however, to determine whether these concepts also apply *in vivo*.

Clinical efficacy of delafloxacin

At this stage, two intravenous Phase II studies have been completed and two Phase III trials are ongoing [10]. A first Phase II trial completed in 2008 (ClinicalTrials.gov identifier: NCT00719810) compared delafloxacin (300 mg or 450 mg intravenously b.i.d.) to tigecycline (100 mg on day 1 then 50 mg intravenously b.i.d.) for the treatment of complicated acute bacterial skin and skin structure infections [65]. Target infections were (a) wound infection having developed within 30 days of surgery, trauma, or an animal/insect bite injury, in patients having required purulent drainage from the wound or presenting at least three of the following symptoms: fever, swelling, erythema of ≥10 mm, pain, or tenderness; (b) abscess, without an open wound, having developed during the 7 days before enrollment, with purulent drainage or aspirate in patients showing evidence of a loculated fluid collection that required intervention within 48 h of enrollment and erythema and/or induration of ≥20 mm in diameter or tenderness; (c) cellulitis having developed during the 7 days before enrollment, with advancing edema, erythema, or induration; in patients showing documented fever or reported fever during the 3 days before enrollment, a white blood cell count of 10×10^9/l or ≥10% band forms, or lymphangitis and adenopathy. Treatment duration was 5–14 days, based on the investigator's judgment. About 50 patients were enrolled in each arm, among whom more than 90% completed the study (48, 51, 50 in the 300 and 450 mg delafloxacin and tigecycline arms, respectively, for the mITT population); more than 70% were clinically evaluable and more than 50%, microbiologically evaluable. The mean duration of the treatment in the intent-to-treat population was 7.9 days, 7.5 days and 6.8 days in the delafloxacin 300-mg, delafloxacin 450-mg and tigecycline arms, respectively. *S. aureus* was isolated in 88% of patients

having positive culture at baseline, among which two thirds were MRSA. Delafloxacin MIC in these strains ranged from ≤0.004 to 0.12 mg/l for both MSSA and MRSA. Cure rates at the test-of-cure visit in the clinically evaluable population were higher than 90% in all arms with no statistical differences among them or between infection types. Cure rates were slightly higher with delafloxacin treatment for MRSA infections, but the difference did not reach significance. Based on this Phase II study (see 'Safety profile' section), a dose of 300 mg IV was considered for further clinical trials.

A second Phase II trial completed in 2011 (ClinicalTrials.gov identifier: NCT01283581) compared 5–14 days treatments of acute bacterial skin and skin structure infections with a minimum of 75 cm^2 erythema with delafloxacin (81 patients; 300 mg intravenously b.i.d.), linezolid (77 patients; 600 mg intravenously b.i.d.) or vancomycin (98 patients; 15 mg/kg, up to 1250 mg, intravenously b.i.d.) [66,67]. This study also concluded the equivalence of these treatments (72–78% success rate) when considering as endpoint the cessation of lesion spread and the absence of fever in the 48–72 h timeframe but better efficacy of delafloxacin when considering as endpoint a reduction >20% or >30% in lesion size after 48–72 h.

Two Phase III trials are ongoing or completed with delafloxacin. They focus again on the treatment of acute bacterial skin and skin structure infections. The first one compares delafloxacin (300 mg intravenously) b.i.d. for up 5–14 days (ClinicalTrials.gov identifier: NCT01811732) and has been recently completed. The second one compares delafloxacin 300 mg intravenously b.i.d. for 3 days followed by 450 mg oral b.i.d. for up 5–14 days total (ClinicalTrials.gov identifier: NCT01984684) to vancomycin (15 mg/kg intravenously) + aztreonam (2 g intravenously) b.i.d. In addition, the US FDA has granted delafloxacin as a Qualified Infectious Disease Product for the indications of acute bacterial skin and skin structure infections (ABSSSI) and community-acquired bacterial pneumonia (CABP) in October 2012. One may expect from these studies to highlight the possible advantages of delafloxacin in terms of spectrum, allowing its use as a monotherapy and of oral bioavailability.

Safety profile

Fluoroquinolones present a large panel of adverse effects, some of which have been related to specific structural characteristics [12]. Yet, except for the halogen in position 8, which has been related to a risk of phototoxicity, the substituents present in delafloxacin are not found in other fluoroquinolones. No report has been published regarding the safety of the molecule in animal models and human data are limited to those collected in Phase II and ongoing Phase III studies. In the first Phase II study [65], delafloxacin was well tolerated at a dose of 300 mg intravenously every 12 h. Adverse events were more frequent in patients receiving 450 mg b.i.d., including gastrointestinal disorders and infusion-site pain. The treatment was discontinued in five patients because of adverse events (two in the delafloxacin 450-mg arm and three in the tigecycline arm) and eight serious adverse events were recorded (one in the delafloxacin 300-mg arm, three in the delafloxacin 450-mg arm and four in the tigecycline arm) among which only a generalized seizure in a 53-year-old male in the delafloxacin 450-mg arm was considered as possibly related to the treatment. Globally, however, safety data available so far from Phase I and other Phase II studies did not reveal any specific adverse event [44,68], including on cardiac function (no prolongation of QTc interval) [69].

Conclusion

With its impressively low MICs against Gram-positive (especially MRSA) and anaerobic bacteria and reasonable activity against Gram-negative bacteria, delafloxacin may represent a breakthrough for the treatment of Gram-positive infections, especially when co-infections by other germs could be suspected. This is clearly the case for skin and soft tissues, where its clinical efficacy has already been demonstrated. Its other advantages in this context are its bactericidal character (as for other fluoroquinolones), its increased potency at the acidic pH of the skin and abscesses [43], its low propensity of selecting resistance (with MPC close to the MIC), its availability as intravenous or oral formulations, its long half-life allowing twice- or once-a-day administration, and its activity on intracellular bacteria and on biofilms, which may play a role in these types of infections. The most promising other indication for this drug would be community-acquired bacterial pneumonia (taking advantage of its very high potency on pneumococci and on atypical pathogens) for which it was already granted as a Qualified Infectious Disease Product by the FDA. Its improved potency at acidic pH needs to be further exploited by exploring other

specific indications for infections developing in acidic environments [43] and caused by susceptible bacteria like gastritis caused by *H. pylori* [18]. Its high activity on *C. difficile* infections may prevent the development of pseudomembranous colitis, as observed for other fluoroquinolones less active on anaerobes [70]. Further studies are needed to document its usefulness in these or other indications. Safety data are reassuring but still very preliminary, with Phase 3 studies ongoing. Thorough follow-up is needed as the occurrence of rare but serious side effects has been a reason for the abandon or withdrawal of many molecules within this class [10,12].

Authors' note

Note that the posters cited in this reference list are available on the website of Melinta Therapeutics at: www.melinta.com/data_publications.php

Disclosure

In addition to the peer-review process, with the author(s) consent, the manufacturer of the product(s) discussed in this article was given the opportunity to review the manuscript for factual accuracy. Changes were made at the discretion of the author(s) and based on scientific or editorial merit only.

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EXECUTIVE SUMMARY

- Delafloxacin is a fluoroquinolone in Phase III of clinical development showing the unusual property of lacking a basic substituent in position 7. This markedly improves its potency at acidic pH.
- Delafloxacin is highly potent on Gram-positive bacteria including MRSA and fluoroquinolone-resistant strains (except for enterococci), anaerobes, and, to a lesser extent, Gram-negative bacteria. Monte-Carlo simulations suggest that 0.5 mg/l could be a reasonable PK/PD breakpoint, which is well above the MIC of most target Gram-positive organisms.
- Delafloxacin is administered twice (or once daily in specific situations) with equivalence in exposure obtained for 300 mg intravenously and 450 mg orally.
- Delafloxacin was as effective as tigecycline, vancomycin, or linezolid in Phase II trials of complicated skin and skin structures infections and is now studied in Phase III trials for the same indication (intravenously only or intravenously with shift to oral route after 3 days.

References

Papers of special note have been highlighted as:
• of interest; •• of considerable interest

1 Pendleton JN, Gorman SP, Gilmore BF. Clinical relevance of the ESKAPE pathogens. *Expert Rev. Anti Infect. Ther.* 11(3), 297–308 (2013).

2 Kang CI, Song JH. Antimicrobial resistance in Asia: current epidemiology and clinical implications. *Infect. Chemother.* 45(1), 22–31 (2013).

3 Molton JS, Tambyah PA, Ang BSP, Ling ML, Fisher DA. The global spread of healthcare-associated multidrug-resistant bacteria: a perspective from Asia. *Clin. Infect. Dis.* 56(9), 1310–1318 (2013).

4 Cassir N, Rolain JM, Brouqui P. A new strategy to fight antimicrobial resistance: the revival of old antibiotics. *Front Microbiol.* 5, 551 (2014).

5 Nation RL, Li J. Colistin in the 21st century. *Curr. Opin. Infect. Dis.* 22(6), 535–543 (2009).

6 Mouton JW, Ambrose PG, Canton R *et al.* Conserving antibiotics for the future: new ways to use old and new drugs from a pharmacokinetic and pharmacodynamic perspective. *Drug Resist. Updat.* 14(2), 107–117 (2011).

7 Butler MS, Blaskovich MA, Cooper MA. Antibiotics in the clinical pipeline in 2013. *J. Antibiot. (Tokyo)* 66(10), 571–591 (2013).

8 Anstead GM, Cadena J, Javeri H. Treatment of infections due to resistant *Staphylococcus aureus*. *Methods Mol. Biol.* 1085, 259–309 (2014).

9 Gould IM, Bal AM. New antibiotic agents in the pipeline and how they can help overcome microbial resistance. *Virulence* 4(2), 185–191 (2013).

10 Van Bambeke F. Renaissance of antibiotics against difficult infections: focus on oritavancin and new ketolides and quinolones. *Ann. Med.* 46(7), 512–529 (2014).

•• **Recent review paper comparing delafloxacin with other quinolones in clinical development.**

11 Emami S, Shafiee A, Forounmadi A. Quinolones: recent structural and clinical developments. *Ir. J. Pharm. Res.* 4(3), 123–136 (2005).

12 Van Bambeke F, Michot JM, Van Eldere J, Tulkens PM. Quinolones in 2005: an update. *Clin. Microbiol. Infect.* 11(4), 256–280 (2005).

13 Yazaki A, Niino Y, Ohshita Y *et al.* WO1996JP02710 19960920(US5998436) (1999).

14 Goldstein EJC, Citron DM, Merriam CV *et al. In vitro* activities of ABT-492, a new

fluoroquinolone, against 155 aerobic and 171 anaerobic pathogens isolated from antral sinus puncture specimens from patients with sinusitis. *Antimicrob. Agents Chemother.* 47(9), 3008–3011 (2003).

15 Zhanel GG, Palatnick L, Nichol KA *et al.* Antimicrobial resistance in respiratory tract *Streptococcus pneumoniae* isolates: results of the Canadian Respiratory Organism Susceptibility Study, 1997–2002. *Antimicrob. Agents Chemother.* 47(6), 1867–1874 (2003).

16 Harnett SJ, Fraise AP, Andrews JM *et al.* Comparative study of the *in vitro* activity of a new fluoroquinolone, ABT-492. *J. Antimicrob. Chemother.* 53(5), 783–792 (2004).

17 Zhanel GG, Palatnick L, Nichol KA, Low DE, Hoban DJ. Antimicrobial resistance in *Haemophilus influenzae* and *Moraxella catarrhalis* respiratory tract isolates: results of the Canadian Respiratory Organism Susceptibility Study, 1997 to 2002. *Antimicrob. Agents Chemother.* 47(6), 1875–1881 (2003).

18 Nilius AM, Shen LL, Hensey-Rudloff D *et al.* *In vitro* antibacterial potency and spectrum of ABT-492, a new fluoroquinolone. *Antimicrob. Agents Chemother.* 47(10), 3260–3269 (2003).

• **Pioneer work examining the *in vitro* activity of delafloxacin.**

19 Hammerschlag MR, Roblin PM. The *in vitro* activity of a new fluoroquinolone, ABT-492, against recent clinical isolates of Chlamydia pneumoniae. *J. Antimicrob. Chemother.* 54(1), 281–282 (2004).

20 Waites KB, Crabb DM, Duffy LB. Comparative *in vitro* susceptibilities and bactericidal activities of investigational fluoroquinolone ABT-492 and other antimicrobial agents against human mycoplasmas and ureaplasmas. *Antimicrob. Agents Chemother.* 47(12), 3973–3975 (2003).

21 Frean J, Klugman KP, Arntzen L, Bukofzer S. Susceptibility of *Yersinia pestis* to novel and conventional antimicrobial agents. *J. Antimicrob. Chemother.* 52(2), 294–296 (2003).

22 Tomioka H, Sato K, Kajitani H, Akaki T, Shishido S. Comparative antimicrobial activities of the newly synthesized quinolone WQ-3034, levofloxacin, sparfloxacin, and ciprofloxacin against *Mycobacterium tuberculosis* and *Mycobacterium avium* complex. *Antimicrob. Agents Chemother.* 44(2), 283–286 (2000).

23 Almer LS, Hoffrage JB, Keller EL, Flamm RK, Shortridge VD. *In vitro* and bactericidal activities of ABT-492, a novel fluoroquinolone, against Gram-positive and Gram-negative organisms. *Antimicrob. Agents Chemother.* 48(7), 2771–2777 (2004).

• **Pioneer work examining the *in vitro* activity of delafloxacin.**

24 Duffy E, Devito JA, Remy J, Burak E. Delafloxacin chemical properties lead to increased potency against Gram-positive pathogens, including quinolone-resistant pathogens II. Preseneted at: *50th Interscience Conference on Antimicrobial Agents and Chemotherapy (ICAAC)*. MA, USA, 12–15 September 2010 (Abstract E183).

25 Lemaire S, Tulkens PM, Van Bambeke F. Contrasting effects of acidic pH on the extracellular and intracellular activities of the anti-Gram-positive fluoroquinolones moxifloxacin and delafloxacin against *Staphylococcus aureus*. *Antimicrob. Agents Chemother.* 55(2), 649–658 (2011).

•• **Pharmacological study examining the influence of pH on the activity of delafloxacin against *S. aureus* (extracellular and intracellular forms) as well as on its accumulation in bacterial and eucaryotic cells.**

26 Burak E, DeVito JA, Remy J, Duffy E. Delafloxacin chemical properties lead to increased potency against Gram-positive pathogens, including quinolone-resistant pathogens I. Presented at: *50th Interscience Conference on Antimicrobial Agents and Chemotherapy (ICAAC)*. MA, USA, 12–15 September 2010 (Abstract E182).

27 So W, Crandon JL, Nicolau DP. Effects of urine matrix and pH on the potency of delafloxacin and ciprofloxacin against urogenic *E. coli* and *K. pneumonia*. *J. Urol.* doi: 10.1016/j.juro.2015.01.094 (2015) (Epub ahead of print).

28 Drlica K, Zhao X. DNA gyrase, topoisomerase IV, and the 4-quinolones. *Microbiol. Mol. Biol. Rev.* 61(3), 377–392 (1997).

29 Alam MM, Kobayashi N, Uehara N, Watanabe N. Analysis on distribution and genomic diversity of high-level antiseptic resistance genes qacA and qacB in human clinical isolates of *Staphylococcus aureus*. *Microb. Drug Resist.* 9(2), 109–121 (2003).

30 Costa SS, Falcao C, Viveiros M *et al.* Exploring the contribution of efflux on the resistance to fluoroquinolones in clinical isolates of *Staphylococcus aureus*. *BMC Microbiol.* 11, 241 (2011).

31 Munoz-Bellido JL, Alonzo MM, Martinez Andres JA *et al.* Efflux pump-mediated quinolone resistance in *Staphylococcus aureus* strains wild type for gyrA, gyrB, grlA, and norA. *Antimicrob. Agents Chemother.* 43(2), 354–356 (1999).

32 Garvey MI, Baylay AJ, Wong RL, Piddock LJ. Overexpression of patA and patB, which encode ABC transporters, is associated with fluoroquinolone resistance in clinical isolates of *Streptococcus pneumoniae*. *Antimicrob. Agents Chemother.* 55(1), 190–196 (2011).

33 Gill MJ, Brenwald NP, Wise R. Identification of an efflux pump gene, *pmrA*, associated with fluoroquinolone resistance in *Streptococcus pneumoniae*. *Antimicrob. Agents Chemother.* 43(1), 187–189 (1999).

34 Van Bambeke F, Glupczynski Y, Plesiat P, Pechere JC, Tulkens PM. Antibiotic efflux pumps in prokaryotic cells: occurrence, impact on resistance and strategies for the future of antimicrobial therapy. *J. Antimicrob. Chemother.* 51(5), 1055–1065 (2003).

35 Tran JH, Jacoby GA. Mechanism of plasmid-mediated quinolone resistance. *Proc. Natl Acad. Sci. USA* 99(8), 5638–5642 (2002).

36 Robicsek A, Strahilevitz J, Jacoby GA *et al.* Fluoroquinolone-modifying enzyme: a new adaptation of a common aminoglycoside acetyltransferase. *Nat. Med.* 12(1), 83–88 (2006).

37 Remy JM, Tow-Keogh CA, McConnell TS, Dalton JM, Devito JA. Activity of delafloxacin against methicillin-resistant *Staphylococcus aureus*: resistance selection and characterization. *J. Antimicrob. Chemother.* 67(12), 2814–2820 (2012).

•• **Study of the activity of delafloxacin against MRSA and of its ability to select resistance.**

38 Kurz SG, Duffy E, Balthazar J, Bomono RA, Shafer WM. Delafloxacin exerts potent anti-gonococcal activity despite mutations that decrease antibiotic susceptibility due to target modification or drug efflux. Presented at: *54th Interscience Conference on Antimicrobial Agents and Chemotherapy*. DC, USA, C-1399 (2014).

39 Drlica K, Zhao X. Mutant selection window hypothesis updated. *Clin. Infect. Dis.* 44(5), 681–688 (2007).

40 Remy J, Marra A, Duffy E. Evaluation of the potential for delafloxacin resistance development in *Neisseria gonorrhoeae*. Presented at: *54th Interscience Conference on Antimicrobial Agents and Chemotherapy*. DC, USA, 6–9 September 2014 (Abstract C-1412a).

41 Lawrence L, Pillar C, Sahm D, Burak E. *In vitro* activity of delafloxacin against European isolates of *Staphylococcus aureus*

from skin and soft tissue, respiratory, urine and blood specimens. Presented at: *21st European Congress of Clinical Microbiology and Infectious Diseases & 27th International Congress of Chemotherapy*. Milan, Italy, 7–10 May 2011 (Abstract P-1185).

42 Lawrence L, Hopkins S, Sahm D *et al.* Characterization and *In vitro* Activity of delafloxacin (DLX) against isolates from a phase 2 study of acute bacterial skin and skin structure infections (ABSSSI). Presented at: *52nd Interscience Conference on Antimicrobial Agents and Chemotherapy (ICAAC)*. CA, USA, 9–12 September 2012 (Abstract E-208).

43 Weinrick B, Dunman PM, McAleese F *et al.* Effect of mild acid on gene expression in *Staphylococcus aureus*. *J. Bacteriol.* 186(24), 8407–8423 (2004).

44 Lawrence L, Benedict M, Hart J *et al.* Pharmacokinetics (PK) and safety of single doses of delafloxacin administered intravenously in healthy human subjets. Presented at: *51st Interscience Conference on Antimicrobial Agents and Chemotherapy (ICAAC)*. IL, USA, 17–20 September 2011 (A2–045a).

45 Lawrence L, Longcor J, Li D *et al.* Metabolism and mass balance of [^{14}C]-Delafloxacin in healthy human volunteers following intravenous administration. *52nd Interscience Conference on Antimicrobial Agents and Chemotherapy (ICAAC)*.CA, USA, 9–12 September 2012 (Abstract A1956).

46 Rubino CM, Bhavnani SM, Burak E, Ambrose PG. Pharmacokinetic-pharmacodynamic target attainment analyses supporting delafloxacin phase 3 dose regimen decisions. Poster presented at: *50th Interscience Conference on Antimicrobial Agents and Chemotherapy*. MA, USA, 12–15 September 2010 (Poster A1–681).

47 Hoover R, Lawrence L, Benedict M *et al.* Pharmacokinetics and relative bioavailability of intravenous and oral formulations of delafloxacin (DLX) in healthy subjects. Presented at: *54th Interscience Conference on Antimicrobial Agents and Chemotherapy*. DC, USA, 5–9 September 2014 (Abstract A-682).

48 Hoover R, Lawrence L, Benedict M *et al.* Pharmacokinetics of a 900 mg oral formulation of delafloxacin (DLX) healthy subjects supporting a gonorrhea phase 3 study. Presented at: *54th Interscience Conference on Antimicrobial Agents and Chemotherapy*. DC, USA, 5–9 September 2014 (Abstract A-056a).

49 Hoover R, Lawrence L, Benedict M *et al.* A Phase 1, open-label, crossover study to determine the effect of food on the pharmacokinetics of a single dose of oral delafloxacin (DLX) in healthy subjects. Presented at: *54th Interscience Conference on Antimicrobial Agents and Chemotherapy*. DC, USA, 5–9 September 2014 (Abstract A-683).

50 Hoover R, Lawrence L, Longcor J, Greenfield J. Pharmacokinetics (PK) of pelafloxacin (DLX), vancomycin (VAN), and linezolid (LNZ) in a phase 2 exploratory study in subjects with acute bacterial skin and skin structure infections (ABSSSI). Presented at: *52nd Interscience Conference on Antimicrobial Agents and Chemotherapy (ICAAC)*. CA, USA, 9–12 September 2012 (Abstract A1957).

51 Hoover R, Lawrence L, Smith C, Longcor J. Pharmacokinetics (PK) of delafloxacin (DLX) in patients with varying degrees of renal impairment. Presented at: *53rd Interscience Conference on Antimicrobial Agents and Chemotherapy (ICAAC)*. CO, USA, 10–13 September 2013 (Abstract A-017E).

52 Ding Y, Villet MA, Lee JC, Hooper DC. Treatment of renal abscesses caused by *Staphylococcus aureus* MW2, using delafloxacin and moxifloxacin. Presented at: *21st European Congress of Clinical Microbiology and Infectious Diseases & 27th International Congress of Chemotherapy*. Milan, Italy, 10 May 2011 (Poster P1506).

53 Marra A, Bortolon E, Molstad D *et al.* Evaluation of delafloxacin in rat granuloma pouch infections caused by Gram-negative pathogens. Presented at; *50th Interscience Conference on Antimicrobial Agents and Chemotherapy (ICAAC)*. MA, USA, 12–15 September 2010 (Abstract A1–680).

54 Wright DH, Brown GH, Peterson ML, Rotschafer JC. Application of fluoroquinolone pharmacodynamics. *J. Antimicrob. Chemother.* 46(5), 669–683 (2000).

55 Burak E, Bortolon E, Molstad D *et al.* Pharmacokinetics and pharmacodynamics of delafloxacin in *S. aureus* murine thigh infection models. *49th Interscience Conference on Antimicrobial Agents and Chemotherapy.* CA, USA, 12–15 September 2009 (Abstract A1–1941).

56 Firsov AA, Vostrov SN, Lubenko IY *et al.* ABT492 and levofloxacin: comparison of their pharmacodynamics and their abilities to prevent the selection of resistant *Staphylococcus aureus* in an *in vitro* dynamic model. *J. Antimicrob. Chemother.* 54(1), 178–186 (2004).

57 Firsov AA, Alferova IV, Smirnova MV *et al.* Comparative pharmacodynamics of the new fluoroquinolone ABT492 and levofloxacin with *Streptococcus pneumoniae* in an *in vitro* dynamic model. *Int. J. Antimicrob. Agents* 25(5), 409–413 (2005).

58 Zinner SH, Vostrov SN, Alferova IV *et al.* Comparative pharmacodynamics of the new fluoroquinolone ABT492 and ciprofloxacin with *Escherichia coli* and *Pseudomonas aeruginosa* in an *in vitro* dynamic model. *Int. J. Antimicrob. Agents* 24(2), 173–177 (2004).

59 Garzoni C, Kelley WL. *Staphylococcus aureus*: new evidence for intracellular persistence. *Trends Microbiol.* 17(2), 59–65 (2009).

60 Archer NK, Mazaitis MJ, Costerton JW *et al.* *Staphylococcus aureus* biofilms: properties, regulation, and roles in human disease. *Virulence* 2(5), 445–459 (2011).

61 Seral C, Carryn S, Tulkens PM, Van Bambeke F. Influence of P-glycoprotein and MRP efflux pump inhibitors on the intracellular activity of azithromycin and ciprofloxacin in macrophages infected by Listeria monocytogenes or *Staphylococcus aureus*. *J. Antimicrob. Chemother.* 51(5), 1167–1173 (2003).

62 Vallet CM, Marquez B, Ngabirano E *et al.* Cellular accumulation of fluoroquinolones is not predictive of their intracellular activity: studies with gemifloxacin, moxifloxacin and ciprofloxacin in a pharmacokinetic/pharmacodynamic model of uninfected and infected macrophages. *Int. J. Antimicrob. Agents* 38(3), 249–256 (2011).

63 Bauer J, Siala W, Tulkens PM, Van Bambeke F. A combined pharmacodynamic quantitative and qualitative model reveals the potent activity of daptomycin and delafloxacin against *Staphylococcus aureus* biofilms. *Antimicrob. Agents Chemother.* 57(6), 2726–2737 (2013).

64 Siala W, Mingeot-Leclercq MP, Tulkens PM *et al.* Comparison of the antibiotic activities of daptomycin, vancomycin, and the investigational fluoroquinolone delafloxacin against biofilms from *Staphylococcus aureus* clinical isolates. *Antimicrob. Agents Chemother.* 58(11), 6385–6397 (2014).

•• **Study examining the activity of delafloxacin in biofilms of *S. aureus* and the factors that can modulate it.**

65 O'Riordan W, Mehra P, Manos P *et al.* A randomized Phase 2 study comparing two doses of delafloxacin with tigecycline in adults with complicated skin and skin-structure infections. *Int. J. Infect. Dis.* 30C, 67–73 (2014).

66 Longcor J, Lawrence L, Duffy E, Hopkins S. Objective measures of clinical efficacy in a Phase 2B Exploratory Study of delafloxacin (DLX) compared to vancomycin (VAN) and

linezolid (LNZ) in adults with acute bacterial skin and skin structure infections (ABSSSI). Presented at: *52nd Interscience Conference on Antimicrobial Agents and Chemotherapy (ICAAC).* CA, USA, 9–12 September 2012 (Abstract L1–1667c).

67 Longcor J, Hopkins S, Lawrence L *et al.* Results of a Phase 2 study of delafloxacin (DLX) compared to vancomycin (VAN) and linezolid (LNZ) in acute bacterial skin and skin structure infections (ABSSSI). Presented at: *52nd Interscience Conference on Antimicrobial Agents and Chemotherapy (ICAAC).* CA, USA, September 2012 (Abstract L1–1663 9–12).

68 Longcor J, Hopkins S, Lawrence L *et al.* Results of a phase 2 study of delafloxacin (DLX) compared with vancomycin (VAN) and linezolid (LNZ) in acute bacterial skin and skin structure infections (ABSSSI). Presented at: *52nd Interscience Conference on Antimicrobial Agents and Chemotherapy (ICAAC)*. CA, USA, 9–12 September 2012 (Abstract L1–1663).

69 Lawrence L, Benedict M, Litwin J *et al.* A thorough Phase 1 QTc study of delafloxacin (DLX) compared with placebo and moxifloxacin (MXF). Presented at: *52nd Interscience Conference on Antimicrobial Agents and Chemotherapy (ICAAC)*. CA, USA, 9–12 September 2012 (Abstract A1958)

70 Deshpande A, Pant C, Jain A, Fraser TG, Rolston DDK. Do fluoroquinolones predispose patients to *Clostridium difficile* associated disease? A review of the evidence. *Curr. Med. Res. Opin.* 24(2), 329–333 (2008).

71 Soge OO, Roberts MC, Lenderman C *et al.* Evaluation of *In vitro* activity of delafloxacin against contemporary neisseria gonorrhoeae isolates. Presented at: *54th Interscience Conference on Antimicrobial Agents and Chemotherapy*. DC, USA, 5–9 September 2014 (Abstract C-1401).