Antimicrobial Activity of Garenoxacin (GRN), a Novel Des-F(6) Quinolone Tested Against 29,460 Community-Acquired Respiratory Pathogens (CA-RTI) from Europe, North America and Latin America

RN JONES, TA BLACK, HS SADER, TR FRITSCHE
JMI Laboratories, North Liberty, Iowa, USA; Schering-Plough, Kenilworth, New Jersey, USA

Schering-Plough

ICID 2006

JMI Laboratories
North Liberty, IA, USA
www.jmilabs.com
319.665.3370,
fax 319.665.3371
ronald-jones@jmilabs.com

Abstract

Background: Contemporary (1999-2005) activity of GRN and comparison fluoroquinolones (FQs) were tested against *S. pneumoniae* (SPN), *H. influenzae* (HI) and *M. catarrhalis* (MCAT) isolates from 3 continents. Trends in resistance and effects of co-R on GRN potency was assessed.

Methods: SPN (13,273), HI (11,695) and MCAT (4,492) were collected from more than 100 laboratories worldwide and tested by CLSI methods in a central reference laboratory, with QC results within published CLSI ranges. Subsets with penicillin-R (PEN; ≥2 mg/L, SPN), levofloxacin-R (LEVO; ≥4 mg/L, SPN) and ampicillin-R (AMP; ≥2 mg/L, HI) were analyzed separately.

Results: GRN exhibited excellent potency (MIC_{90}, ≤0.06 mg/L) against all 3 CA-RTI associated species, not differing by continent or Co-R patterns among PEN or AMP-R subsets. GRN MIC results by continent are shown in the table.

Organism	Continent (no)[a]	MIC_{90}	Highest MIC	% ≤1 mg/L
SPN	NA (6909)	0.06	>4	99.86
	LA (1554)	0.06	2	99.94
	EU (4810)	0.06	2	99.96
HI	NA(6069)	≤0.03	16	99.97
	LA (1178)	≤0.03	0.12	100.0
	EU (4448)	≤0.03	0.25	100.0
MCAT	NA (2685)	≤0.03	0.12	100.0
	LA (289)	≤0.03	0.12	100.0
	EU (1518)	≤0.03	0.12	100.0

a. NA, N. America; LA, Latin America; EU, Europe

All GRN MICs for MCAT were ≤0.03 mg/L and only one HI strain was FQ-R (GRN S rate at 99.99%). 13 SPN strains had GRN MICs at >1 mg/L (0.09%) with only 3 strains with a MIC of ≥4 mg/L (0.02% of SPN). No trend toward greater GRN-R was noted over time (1999-2005), and GRN was 4- and 16-fold more active than moxifloxacin and LEVO, respectively, versus SPN. LEVO-R rate among SPN was 0.95%, 10-fold greater than GRN. GRN MICs (MIC_{90}, 1 mg/L) for LEVO-R SPN were 16-fold higher, making the MIC_{90} for GRN against LEVO-R SPN the same as the MIC_{90} for LEVO against the wild-type (non-mutant) population of SPN.

Conclusions: GRN is a very potent investigational agent with an activity and spectrum at ≤1 mg/L, highest among tested FQ agents. Rare GRN-R SPN and HI strains (≤0.1%) were noted, each having ≥4 QRDR mutations (data not shown). GRN should be a welcome advance for CA-RTI therapy.

Introduction

Garenoxacin is a novel des-F(6)-quinolone that lacks the C6-position fluorine and has a unique difluoromethoxy substitution at position C8. These alterations resulted in a quinolone with improved potency against both DNA gyrase and topoisomerase IV. Garenoxacin has been described as highly active against important Gram-positive and –negative pathogens including: Enterobacteriaceae, staphylococci, streptococci (*S. pneumoniae*, viridans group species and ß-haemolytic streptococci), *Acinetobacter* spp. and some other Gram-negative non-fermentative bacilli, *Haemophilus influenzae*, *Moraxella catarrhalis*, atypical respiratory tract pathogens (mycoplasmas, *C. pneumoniae*, and *Legionella* spp.), many enterococci and anaerobes, especially Gram-positive species. These features are complemented by a favorable pharmacokinetic/pharmacodynamic profile, leading to a high AUC/MIC ratio. This high AUC/MIC ratio predicts a greater probability of favorable target attainment that has been associated with successful bacterial eradication and minimization of mutational events among indicated species (low MPC values). These elements of spectrum and potency favor garenoxacin applications for community-acquired respiratory tract infections (CA-RTI; hospitalized or ambulatory patients).

Since garenoxacin features high potency and breadth of spectrum against common CA-RTI pathogens (29,460 strains tested since 1999 on three continents [Europe, North and Latin America]), subsets of strains having elevated MIC values to currently utilized agents were selected for further studies and compared to all studied strains of *S. pneumoniae*, *H. influenzae*, and *M. catarrhalis*. Garenoxacin MICs versus those organisms and the determination of QRDR mutations for some isolates were used to establish the role of this investigational des-F(6)-quinolone against emerging resistant pathogen types.

Materials and Methods

Bacterial strains tested: Recent RTI strains were categorized into those treated in the ambulatory setting (CA-RTI). The organisms were cultured during 1999-2005 from Latin America (10 medical centers), North America (USA and Canada; 23) and Europe (20). The number of strains was: *S. pneumoniae* (13,273), *H. influenzae* (11,695) and *M. catarrhalis* (4,492). These CA-RTI isolates came from non-duplicated cultures in 23 nations.

Susceptibility testing methods: All MIC values were generated using broth microdilution methods (CLSI M7-A7) with panels produced by TREK Diagnostics (Cleveland, Ohio, USA). Cation-adjusted Mueller-Hinton broth was supplemented where indicated with 2-5% lysed horse blood (streptococci) and HTM components (*Haemophilus*). Concurrent quality assurance was maintained via use of CLSI-recommended strains including *Escherichia coli* ATCC 25922 and 35218, *Pseudomonas aeruginosa* ATCC 27853, *Enterococcus faecalis* ATCC 29212, *Staphylococcus aureus* ATCC 25923 and 29213, *H. influenzae* ATCC 49247 and 49766, and *S. pneumoniae* ATCC 49619. All quality control results were within published MIC ranges (M100-S16) for each agent tested. More than 30 different antimicrobial agents were processed each year with selected agents compared to garenoxacin in this presentation (see Tables). A breakpoint for garenoxacin susceptibility and resistance at ≤1/≥4 mg/L was used for comparison purposes only.

Molecular methods: The QRDR was assessed for mutations in *gyrA* or *gyrB* and, *parC* or *parE* by PCR amplication and sequence analyses. *S. pneumoniae* and *H. influenzae* isolates with elevated garenoxacin MIC results (2->4 mg/L) were processed to detect mutations as were selected isolates with fluoroquinolone MICs ≥ three log_2 dilutions above the wild-type distributions. A total of 124 isolates had QRDR sequences determined.

Results

- A large amount of quantitative (MIC values) activity information has been accumulated on garenoxacin against CA-RTI pathogens (29,460 strains; Tables 1-3) from three continents.
- No significant variations in garenoxacin potency were observed among *S. pneumoniae* ($MIC_{50\ and\ 90}$, 0.06 mg/L), *H. influenzae* (MIC_{90}, ≤0.03 mg/L) and *M. catarrhalis* (MIC_{90}, ≤0.03 mg/L) between strains originating from Europe, North America or Latin America.
- Only two *H. influenzae* strains (North America), and 12 *S. pneumoniae* strains (two in Europe, nine in North America, one in Latin America) were detected with a garenoxacin MIC at >1 mg/L. All potentially garenoxacin-non-susceptible strains had multiple [≥3] mutations in the QRDR (data not shown).

Table 1. Activity of garenoxacin tested against 29,460 CA-RTI pathogens and listed by continent or region of isolation (SENTRY Program, 1999-2005)[a].

Organism (No. tested/ Antimicrobial agent)	Europe MIC_{50}/MIC_{90}	Europe % susceptible	North America MIC_{50}/MIC_{90}	North America % susceptible	Latin America MIC_{50}/MIC_{90}	Latin America % susceptible
H. influenzae	(4,448)		(6,069)		(1,178)	
Garenoxacin	≤0.03/≤0.03	100.0	≤0.03/≤0.03	>99.9	≤0.03/≤0.03	100.0
Ciprofloxacin	≤0.03/≤0.03	100.0	≤0.03/≤0.03	>99.9	≤0.03/≤0.03	100.0
Levofloxacin	≤0.03/≤0.03	100.0	≤0.03/≤0.03	>99.9	≤0.03/≤0.03	100.0
Moxifloxacin	≤0.03/≤0.03	100.0	≤0.03/≤0.03	>99.9	≤0.03/≤0.03	100.0
Ampicillin	≤0.5/>4	84.0	≤0.5/>4	72.1	≤0.5/>4	84.5
Amox/clav[b]	0.5/1	99.8	0.5/1	>99.9	0.5/1	100.0
Azithromycin	1/2	99.8	1/2	99.6	1/2	99.8
Cefuroxime axetil	1/2	98.2	1/2	98.7	1/2	98.7
Tetracycline	≤2/≤2	97.3	≤2/≤2	99.4	≤2/≤2	96.5
Trim/sulfa[b]	≤0.5/>4	79.5	≤0.5/>4	79.3	≤0.5>4	61.1
M. catarrhalis	(1,518)		(2,685)		(289)	
Garenoxacin	≤0.03/≤0.03	-[c]	≤0.03/≤0.03	-	≤0.03/≤0.03	-
Ciprofloxacin	≤0.03/0.06	-	≤0.03/0.06	-	≤0.03/0.06	-
Levofloxacin	≤0.03/0.06	-	≤0.03/0.06	-	≤0.03/0.06	-
Moxifloxacin	0.06/0.06	-	0.06/0.06	-	0.06/0.06	-
Penicillin	4/>4	(4.9)[d]	>4/>4	(4.8)	>4/>4	(2.8)
Amox/clav[b]	≤0.25/≤0.25	-	≤0.25/≤0.25	-	≤0.25/≤0.25	-
Erythromycin	≤0.25/≤0.25	-	≤0.25/≤0.25	-	≤0.25/≤0.25	-
Cefuroxime axetil	1/2	-	1/2	-	1/2	-
Tetracycline	≤2/≤2	-	≤2/≤2	-	≤2/≤2	-
Trim/sulfa[b]	≤0.5/≤0.5	-	≤0.5/≤0.5	-	≤0.5/≤0.5	-
S. pneumoniae	(4,810)		(6,909)		(1,554)	
Garenoxacin	0.06/0.06	>99.9	0.06/0.06	>99.9	0.06/0.06	>99.9
Ciprofloxacin	1/2	(4.6)[e]	1/2	(3.2)	1/2	(3.3)
Levofloxacin	1/1	99.0	1/1	98.9	1/1	99.7
Moxifloxacin	0.12/0.25	99.2	0.12/0.25	99.1	0.12/0.25	99.7
Penicillin	≤0.03/2	69.3	≤0.03/2	65.4	≤0.03/2	70.9
Cefuroxime axetil	≤1/4	78.9	≤1/4	75.4	≤1/4	85.1
Ceftriaxone	≤0.25/1	97.6	≤0.25/1	96.2	≤0.25/1	98.8
Amox/clav[b]	≤1/2	97.0	≤1/2	92.9	≤1/2	97.4
Erythromycin	≤0.25/>2	70.7	≤0.25/>2	71.4	≤0.25/>2	85.8
Clindamycin	≤0.25/>2	79.9	≤0.25/≤0.25	90.1	≤0.25/≤0.25	94.6
Tetracycline	≤2/>8	72.9	≤2/>8	82.5	≤2/>8	81.6
Trim/sulfa[b]	≤0.5/>2	65.8	≤0.5/>2	67.2	≤0.5/>2	55.0

a. Susceptibility criteria of the CLSI (2006) was applied with a breakpoint of ≤1/≥4 mg/L used for garenoxacin.
b. Amox/clav = amoxicillin/clavulanate (2:1 ratio) and trim/sulfa = trimethoprim/sulfamethoxazole (1:19 ratio).
c. - = no criteria are published for *M. catarrhalis*.
d. ß-lactamase-positive test results were used to determine resistance.
e. Percentage in parentheses indicate the ciprofloxacin MICs at ≥4 mg/L, possible QRDR mutants.

Table 2. Impact of various resistances in CA-RTI pathogens on the activity of garenoxacin in three geographic regions.

Organism/resistant group (no. tested)	Garenoxacin MICs (mg/L) by region: Europe 50%	Europe 90%	Europe Range	North America 50%	North America 90%	North America Range	Latin America 50%	Latin America 90%	Latin America Range
H. influenzae	(3,728/710)			(4,368/1,690)			(995/183)		
Ampicillin-susceptible	≤0.03	≤0.03	≤0.03-0.25	≤0.03	≤0.03	≤0.03-16[a]	≤0.03	≤0.03	≤0.03-0.12
Ampicillin-resistant	≤0.03	≤0.03	≤0.03-0.12	≤0.03	≤0.03	≤0.03-0.12	≤0.03	≤0.03	≤0.03-0.12
M. catarrhalis	(69/1,449)			(125/2,560)			(7/282)		
ß-lactamase-negative	≤0.03	≤0.03	≤0.03-0.12	≤0.03	≤0.03	≤0.03	≤0.03	-	≤0.03
ß-lactamase-positive	≤0.03	≤0.03	≤0.03-0.06	≤0.03	≤0.03	≤0.03-0.12	≤0.03	≤0.03	≤0.03-0.12
S. pneumoniae	(339/727/750)			(4,237/1,163/1,225)			(1,004/258/191)		
Penicillin-susceptible	0.06	0.06	≤0.03-2[b]	0.06	0.06	≤0.03-4[c]	0.06	0.06	≤0.03-2[d]
Penicillin-intermediate	0.06	0.06	≤0.03-1	0.06	0.06	≤0.03-2[e]	0.06	0.06	≤0.03-0.12
Penicillin-resistant	0.06	0.06	≤0.03-1	0.06	0.06	≤0.03->4[f]	0.06	0.06	≤0.03-0.12

a. Two strains with garenoxacin MIC results at 2 and 16 mg/L, multiple QRDR mutations confirmed.
b. Two strains at 2 mg/L.
c. One strain each at 2 and 4 mg/L.
d. One strain at 2 mg/L.
e. Four strains at 2 mg/L.
f. One strain at 2 and two isolates at >4 mg/L.

- Among 126 levofloxacin-non-susceptible *S. pneumoniae* (MIC, ≥4 mg/L), garenoxacin MIC values were ≤1 mg/L for nearly 90% of strains (MIC_{50}, 0.5 mg/L).

Table 3. MIC distributions of five quinolones tested against 126 levofloxacin non-susceptible (MIC, ≥4 mg/L) *S. pneumoniae* strains from the SENTRY Program (1999-2005).

Fluoroquinolone	Cum. % inhibited at MIC (mg/L) ≤0.03	0.06	0.12	0.25	0.5	1	2	4	% Susceptible[a]
Garenoxacin	2.4	4.8	9.5	18.3	55.6	89.7	97.6	98.4	89.7[b]
Ciprofloxacin	0.0	0.0	0.0	0.0	0.0	0.0	0.8	-[c]	_[d]
Gatifloxacin	0.0	0.0	0.0	0.0	0.8	3.2	14.3	78.6	3.2
Levofloxacin	0.0	0.0	0.0	0.0	0.0	0.0	0.0	9.5	0.0
Moxifloxacin	0.0	0.0	0.0	2.7	4.5	14.4	67.6	94.6	14.4

a. CLSI (2006) criteria for susceptibility.
b. Susceptible percentage for a breakpoint of only ≤1 mg/L.
c. - = untested concentration.
d. - = no criteria for this species (CLSI, 2006).

Conclusions

- Garenoxacin in vitro susceptibility testing results for nearly 30,000 CA-RTI pathogens revealed a potential resistance rate of only 0.05% (MIC, > 1 mg/L).
- Garenoxacin should be a welcome addition for CA-RTI therapy, especially for use against emerging isolates that are resistant to ß-lactams, MLS_B class agents, trimethoprim/sulfamethoxazole, tetracyclines, and recently introduced fluoroquinolones, such as levofloxacin.

Selected References

Anderegg TR, Jones RN (2004). Bactericidal activity of garenoxacin tested by kill-curve methodology against wild type and QRDR mutant strains of *Streptococcus pneumoniae*. *Diagn Microbiol Infect Dis* 50: 213-217.

Andes D, Craig WA (2003). Pharmacodynamics of the new des-f(6)-quinolone garenoxacin in a murine thigh infection model. *Antimicrob Agents Chemother* 47: 3935-3941.

Biedenbach DJ, Jones RN, Pfaller MA (2001). Activity of BMS284756 against 2,681 recent clinical isolates of *Haemophilus influenzae* and *Moraxella catarrhalis*: Report from The SENTRY Antimicrobial Surveillance Program (2000) in Europe, Canada and the United States. *Diagn Microbiol Infect Dis* 39: 245-250.

Clinical and Laboratory Standards Institute. (2006). *Methods for dilution antimicrobial susceptibility tests for bacteria that grow aerobically, 7th ed. Approved Standard M7-A7*. Wayne, PA: CLSI, 2006.

Clinical and Laboratory Standards Institute. (2006). *Performance standards for antimicrobial susceptibility testing, 16th informational supplement M100-S16.* Wayne, PA: CLSI.

Ince D, Zhang X, Silver LC, Hooper DC (2002). Dual targeting of DNA gyrase and topoisomerase IV: Target interactions of garenoxacin (BMS-284756, T-3811ME), a new desfluoroquinolone. *Antimicrob Agents Chemother* 46: 3370-3380.

Jones RN, Biedenbach DJ (2003). Comparative activity of garenoxacin (BMS 284756), a novel desfluoroquinolone, tested against 8,331 isolates from community-acquired respiratory tract infections: North American results from the SENTRY Antimicrobial Surveillance Program (1999-2001). *Diagn Microbiol Infect Dis* 45: 273-278.

Saravolatz L, Manzor O, Pawlak J, Belian B (2001). Antimicrobial activity of BMS 284756, a novel des-fluoro (6) quinolone and seven fluoroquinolones against *Streptococcus pneumoniae*. *Clin Microbiol Infect* 7: 572-573.

Van Wart S, Phillips L, Ludwig EA, Russo R, Gajjar DA, Bello A, Ambrose PG, Costanzo C, Grasela TH, Echols R, Grasela DM (2004). Population pharmacokinetics and pharmacodynamics of garenoxacin in patients with community-acquired respiratory tract infections. *Antimicrob Agents Chemother* 48: 4766-4777.