Comparative In Vitro Activity of Zabofloxacin (DW-224a) Tested Against Multidrug-Resistant *Neisseria Gonorrhoeae*

RN JONES, DJ BIEDENBACH, HS SADER, TR FRITSCHE, P AMBROSE, M WIKLER
JMI Laboratories, North Liberty, IA, USA;
Institute for Clinical Pharmacodynamics at Ordway Research Institute, Albany, NY;
Pacific Beach BioSciences, San Diego, CA, USA

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

ABSTRACT

Background: Zabofloxacin, a novel and potent fluoroquinolone active against respiratory pathogens (*Streptococcus pneumoniae*, *Haemophilus influenzae* and *Moraxella catarrhalis*) is currently undergoing early clinical development. Activity of zabofloxacin tested against *N. gonorrhoeae* (GC) having multidrug-resistant (MDR) patterns could position this new oral agent for wide applications in STD therapy.

Methods: A global collection of 35 highly selected GC isolates including strains with various penicillin, tetracycline and ciprofloxacin susceptibility patterns were tested by CLSI agar dilution methods using GC agar base with defined growth supplement. Concurrent quality controls (QC) used GC ATCC 49226 and commonly used facultative controls *E. faecalis* ATCC 29212 and *S. aureus* ATCC 29213. All QC results were within defined limits. Interpretive criteria were those of the CLSI (M100-S18, 2008), where available.

Results: The challenge GC were generally non-susceptible to penicillin (71%; 23% at ≥2 mg/L), tetracycline (77%; 31% at ≥2 mg/L) and fluoroquinolones (ciprofloxacin 40%; 17% at ≥1 mg/L); only one GC isolate had a zabofloxacin MIC at >0.5 mg/L. Zabofloxacin was four- to -eight-fold more active than ciprofloxacin (MIC_{50}, 0.06 mg/L). Ceftriaxone remained active (MIC_{50}, 0.06 mg/L; 100.0% susceptible) against all GC tested. GC strains with elevated ciprofloxacin MICs (multiple QRDR mutations) also had higher zabofloxacin MIC results with ciprofloxacin-susceptible, -intermediate and -resistant strains having zabofloxacin MIC_{50} results at 0.008, 0.03 and 0.5 mg/L, respectively. GC agar base supplements did not adversely effect zabofloxacin activity.

See Table 1.

Conclusions: Zabofloxacin was significantly more potent (eight-fold) than ciprofloxacin against all MDR-GC. 97% of strains had a zabofloxacin MIC at ≤0.5 mg/L indicating activity against many strains currently defined as resistant to marketed fluoroquinolones; clinical trials should be considered.

INTRODUCTION

Zabofloxacin (formerly DW-224a), a 6-F naphthyridine (Figure 1), has been studied in experiments reported at international meetings in 2003-2008 as well as in several publications. Generally, this so-called "fluoroquinolone" has been described as two- to 32-fold more potent than marketed agents in the same class against Gram-positive and fastidious Gram-negative pathogens, and toxicity analysis showed limited adverse events (genetic toxicity, phototoxicity, convulsions). Zabofloxacin has intracellular activity against *L. pneumophilia* and other studies showed superior potency against *M. hominis*, *C. pneumoniae* and *L. pneumophilia* when compared to moxifloxacin, gatifloxacin and levofloxacin. Appelbaum and colleagues established the $MIC_{50/90}$ of zabofloxacin against *S. pneumoniae* at 0.016/0.03 mg/L for levofloxacin-susceptible wildtype (WT) strains and for ciprofloxacin-resistant (MIC, ≥4 mg/L) strains, the zabofloxacin $MIC_{50/90}$ was only 0.12/0.25 mg/L. Subsequent ICAAC abstracts showed zabofloxacin was bactericidal and did not alter blood glucose levels in diabetic rodent models. In 2006, Kwak et al. and Park et al., respectively, demonstrated a dual targeting (DNA gyrase and DNA topoisomerase IV) feature against *S. pneumoniae* and an activity against *N. gonorrhoeae* equal to gemifloxacin.

Continued emergence of antimicrobial resistance in *N. gonorrhoeae* has required modifications in treatment guidelines (Centers for Disease Control and Prevention [CDC]), including the rapid appearance of fluoroquinolone-resistant (QRNG) strains. This problem has been compounded by limited availability of key broad-spectrum ß-lactams (cefixime) that could have sustained the use of orally administered agents against uncomplicated gonorrhea. Currently few agents remain active against >95% of contemporary *N. gonorrhoeae* isolates in the United States (USA), necessitating greater use of the systemic antimicrobials such as ceftriaxone and cefotaxime, or applying other drug classes (macrolides,carbapenems, penems). The rates of QRNG in the CDC GISP surveillance was 9.4 and 13.3% nationwide in 2005 and 2006, respectively, rates requiring alternative agents to the fluoroquinolones or local surveillance susceptibility data to direct therapy. This study evaluated the activity of zabofloxacin and other agents against a selected collection of *N. gonorrhoeae* isolates, including numerous resistant phenotypes and genotypes.

MATERIALS AND METHODS

Susceptibility testing: All MIC values were generated using the reference agar dilution method as described by the Clinical and Laboratory Standards Institute (CLSI) document M7-A7 (2006). Interpretive criteria for categorical interpretations were those from CLSI M100-S18 (2008) and quality control (QC) guidelines were derived from the same document. All QC results were within published ranges. The tested antimicrobials were provided by Pacific Beach BioSciences (zabofloxacin) and their domestic manufacturers or Sigma Chemical Co. (St. Louis, MO, USA). Two quinolones were tested including zabofloxacin and ciprofloxacin.

Organisms tested: Recent clinical isolates of *N. gonorrhoeae* were collected from numerous sites in the United States (USA) where resistance to commonly prescribed anti-gonococcal agents (fluoroquinolones) was known to be endemic. These sites were located in New York (three isolates), California (four isolates), Washington (five isolates), Oregon (three isolates), Hawaii (four isolates) and Ohio (four isolates). The remaining 12 strains were from older stock cultures collected from medical centers located outside the USA, including Japan and the Netherlands. The overall collection consisted of 71.0% penicillin non-susceptible strains (23.0% ß-lactamase positive) and 40.0% ciprofloxacin non-susceptible gonococci. Several of the ciprofloxacin-resistant isolates had multiple documented amino acid mutations within the quinolone resistance determining region (QRDR).

RESULTS

- Zabofloxacin MIC_{90} results indicated an eight-fold greater potency when compared to ciprofloxacin (0.5 versus 4 mg/L).
- Overall ceftriaxone was the most active (MIC_{90}, 0.06 mg/L; 100.0% susceptible) agent tested.
- Other tested antimicrobials were only active against 23 to 60% of gonococcal isolates or did not have interpretive criteria (azithromycin).

Table 1. In vitro activity of zabofloxacin and five comparison agents tested against *N. gonorrhoeae* (35 strains) with various resistance mechanisms.

Antimicrobial agent	MIC (mg/L) 50%	90%	Range	% Susceptible/resistant[a]
Zabofloxacin	0.015	0.5	0.002-1	NA/NA
Ciprofloxacin	0.06	4	≤0.008->4	60/17
Penicillin G	0.25	>4	≤0.008->4	29/23
Ceftriaxone	≤0.008	0.06	≤0.008-0.12	100/-
Tetracycline	1	>4	0.12->4	23/34
Azithromycin	0.25	0.5	0.06-2	NA/NA

a. NA = breakpoint criteria not available.

Figure 1. DW-244a chemical structure.

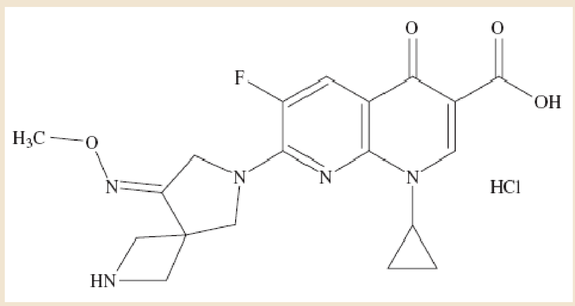
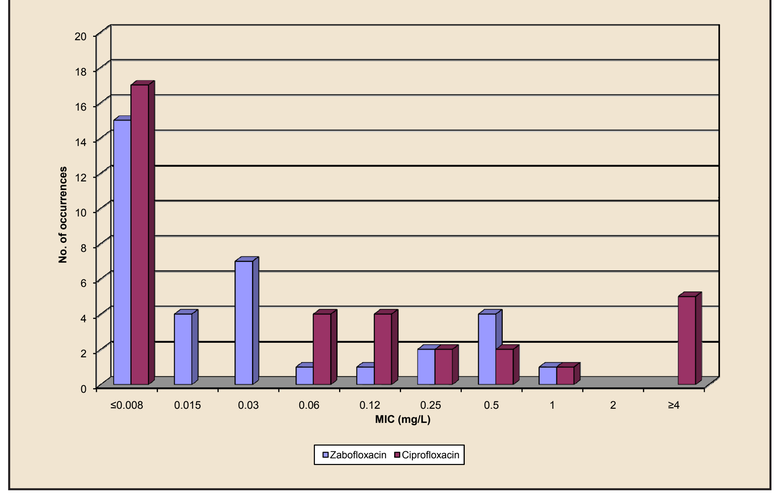


Figure 2. MIC distributions for zabofloxacin and ciprofloxacin.



CONCLUSIONS

- Against epidemic, emerging fluoroquinolone-non-susceptible *N. gonorrhoeae*, zabofloxacin was among the most active agents ($MIC_{50/90}$, 0.015/0.5 mg/L) tested.
- High single-dose therapies with zabofloxacin should be considered in geographic regions where ciprofloxacin resistances have become common.

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