



In Vitro Activity of Nafithromycin (WCK 4873) against *Chlamydia pneumoniae*

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Nafithromycin (WCK 4873) is a novel lactone ketolide under clinical development as an orally administered antibiotic for treatment of community-acquired pneumonia (CAP) caused by *Streptococcus pneumoniae*, *Haemophilus influenzae*, *Moraxella catarrhalis*, and methicillin-susceptible *Staphylococcus aureus*. A recent worldwide antibiotic resistance surveillance study found that nafithromycin had an MIC₉₀ of 0.12 mg/liter against macrolide-resistant and telithromycin-nonsusceptible *S. pneumoniae* isolates (1). However, there are no published data on the activity of nafithromycin against atypical respiratory pathogens, including *Chlamydia pneumoniae* and *Mycoplasma pneumoniae*, which are important causative agents of CAP (2). We tested the in vitro activity of nafithromycin compared to azithromycin, doxycycline, and levofloxacin against 10 clinical isolates of *C. pneumoniae* from patients with community-acquired pneumonia.

The antibiotics were provided as powders and solubilized according to the manufacturers' instructions. Susceptibility testing of *C. pneumoniae* was performed in cell culture using HEp-2 cells grown in 96-well microtiter plates as previously described (3). Each well was inoculated with 0.2 ml of the *C. pneumoniae* isolate diluted to yield 10⁴ inclusion-forming units per ml; the plates were centrifuged at 1,700 × *g* for 1 h and incubated at 35°C for 1 h. Wells were then aspirated and overlaid with medium containing 1 μg/ml of cycloheximide and serial 2-fold dilutions of the test drugs. After incubation at 35°C for 72 h, cultures were fixed and stained for inclusions with fluorescein-conjugated antibody to the chlamydial lipopolysaccharide genus-specific antigen (Pathfinder *Chlamydia* Culture Confirmation System). The MIC was the lowest antibiotic concentration at which no inclusions were seen. The activity of nafithromycin and comparators against individual isolates of *C. pneumoniae* is shown in Table 1. The MICs of nafithromycin ranged from 0.03 to 1 mg/liter, the MIC₅₀ was 0.03 mg/liter, and the MIC₉₀ was 0.25 mg/liter. The MIC ranges of azithromycin were from 0.03 to 0.125 mg/liter with an MIC₅₀ of 0.03 mg/liter and MIC₉₀ of 0.06 mg/liter. The MIC range for doxycycline was 0.03 to 0.25 mg/liter, with MIC₅₀ and MIC₉₀ of 0.125 mg/liter. The MIC range of levofloxacin was 0.25 to 0.5 mg/liter; MIC₅₀ and MIC₉₀ were 0.5 mg/liter.

The in vitro activity of nafithromycin against *C. pneumoniae* was comparable to the other antibiotics tested. Recent pharmacokinetic studies have demonstrated good absorption after oral dosing with nafithromycin maximum concentration of drug in serum (C_{max}) ranging from 1.34 to 2.987 mg/liter after 7 days, which is significantly above the MIC₉₀ for *C. pneumoniae* (0.25 mg/liter) reported here (4). Nafithromycin has also demonstrated sustained levels in epithelial lining fluid (ELF) and alveolar macrophages (AM) with ratios of ELF to plasma concentrations based on the median area under the concentration-time curve from 0 to 24 h (AUC_{0–24}) values of >10 and >350, respectively (5). A phase II, randomized placebo-controlled study has compared oral nafithromycin to moxifloxacin for treatment of CAP in adults; preliminary results showed clinical equivalence, but no microbiologic data were reported (<https://clinicaltrials.gov/ct2/show/results/NCT02903836>). The in vitro susceptibility methods used to perform this study have been shown to correlate with microbiological eradication of *C. pneumoniae* from the respiratory tract by other antibiotics, including azithromycin,

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TABLE 1 MICs of individual *C. pneumoniae* isolates to nafithromycin and comparators

<i>C. pneumoniae</i> isolate	MIC (mg/liter) of:			
	Nafithromycin	Azithromycin	Doxycycline	Levofloxacin
BAL13	1	0.03	0.25	0.5
BAL15	0.03	0.03	0.125	0.5
BAL16	0.25	0.06	0.125	0.5
BAL19	0.03	0.06	0.06	0.5
BAL37	0.03	0.06	0.125	0.25
BAL62	0.03	0.06	0.06	0.25
TW2040	0.25	0.03	0.03	0.5
TW183	0.03	0.03	0.125	0.5
CM-1	0.25	0.03	0.125	0.25
AR39	0.25	0.125	0.125	0.25
MIC ₅₀	0.03	0.03	0.125	0.5
MIC ₉₀	0.25	0.06	0.125	0.5

clarithromycin, levofloxacin, and moxifloxacin (2, 3, 6). The role of nafithromycin in the treatment of *C. pneumoniae* infection will depend on the results of clinical studies that assess microbiological efficacy.

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