

Comparative In Vitro Activities of Sparfloxacin

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Abstract

Sparfloxacin is a third generation quinolone (second generation fluoroquinolone) with enhanced activity against gram-positive cocci. We determined the antibacterial activity of sparfloxacin in comparison to relevant agents against 608 clinical bacterial isolates obtained from patients hospitalized in Siriraj Hospital, a tertiary care, 2,000-bed university hospital in Bangkok, Thailand. Sparfloxacin was active against methicillin-sensitive *S. aureus* (MSSA), penicillin-sensitive *S. pneumoniae* (PSSP), penicillin-resistant *S. pneumoniae* (PRSP) and enterococci, but not methicillin-resistant *S. aureus* (MRSA). The overall activity against gram-positive cocci and coverage of sparfloxacin were similar to standard agents of each organism but were superior to the activities and coverage of first generation fluoroquinolones when such organisms were resistant to their first-line agents. The activity of sparfloxacin against gram-negative bacteria, on the other hand, were similar to, or slightly less active than ciprofloxacin, its congener. The anti-pseudomonal activity of sparfloxacin and its coverage were not remarkable. The activity of sparfloxacin against non-fermenters, however, was impressive, with lower MIC₉₀ than ciprofloxacin and better coverage than ciprofloxacin, ceftazidime and amikacin. The antibacterial action and the potential clinical use of sparfloxacin are discussed. (*J Infect Dis Antimicrob Agents* 1999;16:7-12.)

Fluoroquinolones are potent antibacterial agents. The agents in older generation (ciprofloxacin, ofloxacin and levofloxacin) are active against gram-negative bacteria. The newer generation, although retaining gram-negative activities, have an extended activity range that covers many problematic gram-positive bacteria.¹ Sparfloxacin is one of the newer fluoroquinolones which covers a wide range of gram-positive bacteria as well as other important respiratory pathogens.^{2,3} There have been numerous clinical trials which show that this particular agent may have a key role in treating respiratory tract infections.^{2,4} We determined the local susceptibility pattern of bacteria

isolated from patients hospitalized at Siriraj Hospital, Bangkok, Thailand, to sparfloxacin, and compared this to other relevant antibacterial agents. It was anticipated that the results would reveal the possible usefulness of sparfloxacin in this area of Thailand.

MATERIALS AND METHODS

Bacteria

A total of 608 recent clinical bacterial isolates were obtained from patients hospitalized at Siriraj Hospital, a 2,000-bed tertiary care university hospital in Bangkok, Thailand. Each represented a single isolate

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from patients, acquired either in the community or in the hospital.

Antimicrobial agents

Standard powders of sparfloxacin, ofloxacin, ciprofloxacin, trovafloxacin, ceftazidime, imipenem, cefoperazone-sulbactam, and amikacin, were kindly provided by the representative manufacturers. Penicillin, oxacillin, ampicillin, and vancomycin were E-test strips (AB Biodisk®, Swedens). Trimethoprim-sulfamethoxazole (TMP-SMX) was standard powder purchased from Sigma Chemical Co.

Susceptibility tests

Quantitative methods were performed either by agar dilution or using E-test strips.

For agar dilution, the method described by the National Committee for Clinical Laboratory Standard (NCCLS) was used.⁵ Briefly, the final inocula of approximately 10^5 cfu/ml were applied by a replicator onto antibiotic-containing media. Mueller-Hinton agar was used. Plates were incubated at 35°C for 18-24 hours.

E-test, a gradient diffusion method was used as described previously.⁶ Briefly, inocula were prepared

by emulsification of colonies from an overnight agar plate in a Mueller-Hinton broth to achieve a 0.5 McFarland turbidity standard. These were inoculated onto Mueller-Hinton agar. Strips were applied on a dry surface. Plates were incubated at 35°C for 18-24 hours.

For *S. pneumoniae*, the test media (both in E-test and agar dilution) were supplemented with 5 percent defibrinated sheep blood and the atmosphere was supplemented with 5 percent CO₂ during incubation.

Reference strains

S. aureus ATCC 29213, *S. pneumoniae* ATCC 49619, *E. coli* ATCC 25922 and *P. aeruginosa* ATCC 27853, were used as quality controls.

The MICs were the lowest concentrations of antibiotic which completely inhibited bacterial growth. These were reported as MIC₅₀, MIC₉₀ and range of MICs. Percentage susceptibility was obtained by using standard susceptibility breakpoints. Mostly, the NCCLS breakpoints were used when available.

RESULTS

Activities of sparfloxacin and other antibiotics against gram-positive bacteria are shown in Table 1.

Table 1. *In vitro* activity of sparfloxacin against gram-positive bacteria, compared to other agents.

Organisms (no. of strains)	Antimicrobial agents	MIC (µg/ml)			% susceptibility
		Range	MIC ₅₀	MIC ₉₀	
<i>S. aureus</i> (MRSA) (62)	Sparfloxacin	0.025 - 0.25	0.05	0.1	100
	Ofloxacin	0.25 - 0.5	0.5	0.5	100
	Ciprofloxacin	0.25 - 1	0.5	0.5	100
	Oxacillin	0.2 - 0.4	0.4	0.4	100
<i>S. aureus</i> (MRSA) (31)	Sparfloxacin	0.05 - 16	16	16	6.4
	Ciprofloxacin	0.25 - >16	>16	>16	6.4
	Vancomycin	0.75 - 2	1.5	2	100
<i>S. pneumoniae</i> (penicillin-sensitive, 30)	Sparfloxacin	0.023 - 0.38	0.19	0.25	100
	Ciprofloxacin	0.5 - 4	2	4	33.3
	Trovafloxacin	0.05 - 0.2	0.1	0.2	100
	Penicillin	0.008 - 0.064	0.016	0.047	100
<i>S. pneumoniae</i> (penicillin-resistant, 30)	Sparfloxacin	0.025 - 24	0.125	0.25	95.9
	Ciprofloxacin	0.5 - >16	2	>16	21.4
	Trovafloxacin	0.05 - 16	0.25	16	82.3
	Penicillin	0.125 - 4	0.5	2	0
<i>Enterococcus</i> spp. (31)	Sparfloxacin	0.25 - >16	1	8	67.7
	Ciprofloxacin	0.5 - >16	2	16	35.5
	Trovafloxacin	0.1 - 16	0.25	2	90.3
	Ampicillin	0.5 - 64	1	4	93.4
	Vancomycin	0.75 - 4	1.5	3	100

Sparfloxacin inhibited methicillin-sensitive *S. aureus* (MSSA) and penicillin-susceptible *S. pneumoniae* (PSSP) at an MIC₉₀ of ≤ 0.2 $\mu\text{g/ml}$. Sparfloxacin was the most active drug for MSSA based on the MIC value when compared to first generation fluoroquinolone (second generation quinolones) or even to oxacillin. For PSSP, sparfloxacin was the second active agent (from penicillin G), and was more active than first generation fluoroquinolones. All isolates (100%) of MSSA were susceptible to all quinolones tested and to oxacillin. All isolates of PSSP tested were susceptible to sparfloxacin, trovafloxacin and penicillin, while only 30 percent were susceptible to ciprofloxacin. Methicillin-resistant *S. aureus* (MRSA) was not susceptible to sparfloxacin and older fluoroquinolones, but it was susceptible to vancomycin. Penicillin-resistant *S. pneumoniae* (PRSP) was highly susceptible to sparfloxacin, with the lowest MIC₉₀ (0.25 $\mu\text{g/ml}$), when compared to other fluoroquinolones including trovafloxacin. Ninety-six percent of PRSP were susceptible to sparfloxacin. For enterococci, sparfloxacin was not as active as trovafloxacin, another second generation fluoroquinolone, and vancomycin. The coverage of enterococci by sparfloxacin was also not as good as trovafloxacin, vancomycin and ampicillin.

The activities of sparfloxacin against Enterobacteriaceae were similar to or slightly less active than ciprofloxacin, the first generation fluoroquinolone with the most potent activity against gram-negative bacteria. Sparfloxacin also exerted equivalent activity against gram-negative bacteria as amikacin, based on the MIC₉₀ basis. Its activities against *Enterobacter* spp., however, was significantly higher than amikacin. The percentage of coverage of these agents for common gram-negative bacilli was also similar, and was within 80-100 percent.

Antipseudomonal activities of sparfloxacin and ciprofloxacin were similar. The coverage for this organism by these two quinolones was not as good as ceftazidime nor imipenem.

The activities and the coverage of sparfloxacin were not good for *Acinetobacter* spp. and *Burkholderia pseudomallei*, and were also inferior to their relevant comparators. The activities of sparfloxacin against non-fermenters, on the other hand, were impressive, with the MIC₅₀ and MIC₉₀ of 0.5 and 4 $\mu\text{g/ml}$. This was also true for its coverage. Both the activities and coverage were superior to compara-

tors.

Table 2 shows the *in vitro* activities of sparfloxacin and its comparative agents against gram-negative bacilli commonly encountered in clinical practice.

DISCUSSION

The results of this study show that sparfloxacin had enhanced activity against gram-positive bacteria over the older generation fluoroquinolones. Its activity against gram-negative bacteria, however, was similar or slightly less than its congeners. The results of this study are similar to those reported elsewhere.^{2,3,7-9} The increased potency against gram-positive bacteria of sparfloxacin was ascribed to the NH₂ group substitution at position 5 of the quinolone ring structure.¹⁰ Recently, much evidence has accumulated indicating that topoisomerase IV is the primary target of fluoroquinolones in *S. aureus*.¹¹⁻¹³ Moreover, it was found that the first generation fluoroquinolones (e.g. ciprofloxacin, ofloxacin) were approximately two fold more effective in inhibiting gyrase enzymes than in inhibiting topoisomerase IV.^{14,15} This explains why gram-positive bacteria are less sensitive to older fluoroquinolones. It was shown that *S. aureus* strain with quinolone-resistant gyrase and quinolone-susceptible topoisomerase IV was entirely susceptible to sparfloxacin.¹⁶ This result suggests that sparfloxacin targets topoisomerase IV in *S. aureus*, and should be placed among the second generation fluoroquinolones or third generation quinolones.¹⁷

In this study, as well as in other reports,¹⁸⁻²⁰ sparfloxacin was not active against MRSA. Some studies, however, have found that MRSA are susceptible to sparfloxacin.^{21,22} The reason could be variation in the strains used for susceptibility testing. 'True' MRSA tend to be multidrug-resistant (MDR). Fey and colleagues found that in MRSA (and in methicillin-resistant *S. epidermidis*, MRSE as well), the genes *gyrA* and *mecA* were in the same relative orientation, with a distance between them of 38 and 42 kb in all strains tested.²³ In our study, our MRSA strains were 'true' MRSA which were MDR and might have acquired the *gyrA* gene in *mec*-associated DNA.

In this study, sparfloxacin exerted high potency (MIC₉₀ of 0.25 $\mu\text{g/ml}$) against penicillin-resistant *S. pneumoniae*, and also provided wide coverage of this organism (96% susceptible). This is similar to

Table 2. *In vitro* activity of sparfloxacin against gram-negative bacteria, compared to other agents.

Organisms (no. of strains)	Antimicrobial agents	MIC ($\mu\text{g/ml}$)			% susceptibility	
		Range	MIC ₅₀	MIC ₉₀		
<i>E. coli</i> (62)	Sparfloxacin	0.0025 - >16	0.005	8	85.4	
	Ofloxacin	0.025 - >16	0.05	16	85.4	
	Ciprofloxacin	0.0025 - >16	0.025	16	80.6	
	Amikacin	0.05 - 64	2	4	98.4	
<i>K. pneumoniae</i> (62)	Sparfloxacin	0.005 - >16	0.05	8	82.3	
	Ofloxacin	0.05 - >16	0.1	4	83.8	
	Ciprofloxacin	0.025 - >16	0.05	2	82.0	
	Amikacin	1 - 64	2	16	90.3	
<i>Proteus</i> spp. (21)	Sparfloxacin	0.05 - >16	0.25	16	76.2	
	Ofloxacin	0.1 - 16	0.25	2	90.5	
	Ciprofloxacin	0.005 - 8	0.025	0.5	90.3	
	Amikacin	1 - 4	4	4	100	
<i>Enterobacter</i> spp. (62)	Sparfloxacin	0.01 - 16	0.05	1	93.6	
	Ofloxacin	0.01 - >16	1	2	91.8	
	Ciprofloxacin	0.005 - >16	0.05	1	93.6	
	Amikacin	1 - 64	2	64	82.2	
<i>P. aeruginosa</i> (93)	Sparfloxacin	0.1 - >16	1	>16	59.2	
	Ciprofloxacin	≤ 0.1 - >16	0.5	>16	62.3	
	Amikacin	1 - >128	4	>128	72.0	
	Ceftazidime	0.25 - >128	2	>128	72.1	
	Imipenem	0.5 - >128	2	8	86.0	
	Sparfloxacin	0.005 - 8	0.025	8	75.3	
<i>Acinetobacter</i> spp. (62)	Ciprofloxacin	0.05 - >16	0.5	>16	61.2	
	Cefoperazone- sulbactam	0.25 - 32	2	8	98.4	
	Imipenem	0.05 - 4	0.25	1	100	
	<i>Burkholderia pseudomallei</i> (31)	Sparfloxacin	0.5 - 4	2	4	22.6
		Ciprofloxacin	4 - 16	8	16	0
Ceftazidime		1 - 4	2	2	100	
TMP-SMX		0.05 - 4	1	4	64.6	
Non-fermenters (31)	Sparfloxacin	0.01 - 8	0.5	4	70.8	
	Ciprofloxacin	0.1 - >16	2	16	29.0	
	Ceftazidime	0.25 - >128	16	128	41.9	
	Amikacin	0.25 - >128	64	>128	26.8	
	TMP-SMX	0.01 - 64	1	8	77.4	

previous reports.^{8,24} Interestingly, Pan and Fisher found that sparfloxacin targets primarily DNA gyrase of *S. pneumoniae* and not topoisomerase IV as in *S. aureus*.²⁵ This suggests that sparfloxacin has at least two different targets in gram-positive species. It remains to be elucidated why sparfloxacin is more active than ciprofloxacin against *S. pneumoniae*, when both quinolones act at the same target.

Sparfloxacin also had good activities against other major respiratory pathogens not included in this study.^{2,3} These pathogens are *Chlamydia pneumo-*

niae, *Legionella pneumophila*, *Mycoplasma pneumoniae* and *Mycobacterium tuberculosis*. Based on *in vitro* studies, sparfloxacin may be one of the drugs of first choice for empiric treatment of respiratory tract infections, provided that the safety and tolerability are in an acceptable range.^{3,26-28} In Thailand, sparfloxacin is used infrequently in this situation. This may be due to the high cost and a concern about adverse effects. More importantly, use of fluoroquinolones in empiric treatment of respiratory tract infections should be reserved for physicians specia-

lized in pulmonology, infectious diseases and ear-nose-throat, in response to the global warnings of the coming of a post-antibiotic era and the threat of MDR infections spreading worldwide.²⁹⁻³²

The activity against gram-negative bacteria of sparfloxacin were similar to or slightly less than its congener, ciprofloxacin, making the use of sparfloxacin in treating infections caused by these organisms less interesting. Besides, there was cross-resistant between ciprofloxacin and related quinolones possessing similar chemical structures.^{10,33} The search for antimicrobials active against gram-negative bacteria with different chemical structures will be worthwhile.

In summary, sparfloxacin proved to be active against a wide range of common bacterial isolates obtained locally in a university hospital in Bangkok, Thailand. The judicious use of this agent, however, should be emphasized. The cost, concern about adverse events, and the need to slow the emergence of drug resistance, are the main reasons that it should be used prudently.

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