

Comparative *In Vitro* Activity of Prulifloxacin against Bacteria Isolated from Hospitalized Patients at Siriraj Hospital

Visanu Thamlikitkul, M.D.,
Surapee Tiengrim, M.Sc.

ABSTRACT

In vitro activity of prulifloxacin against 257 clinical isolates of β -hemolytic streptococcus group A, *Streptococcus pneumoniae*, methicillin-susceptible *S. aureus*, ESBL-non-producing *E. coli*, ESBL-producing *E. coli*, ESBL-non-producing *Klebsiella pneumoniae*, ESBL-producing *K. pneumoniae*, *Pseudomonas aeruginosa* and *Salmonella* spp. was conducted by Kirby-Bauer disk diffusion and agar dilution. The study results of Kirby-Bauer disk diffusion revealed that prulifloxacin was as active as ciprofloxacin, levofloxacin and moxifloxacin against the aforementioned organisms. All tested gram-positive bacteria had prulifloxacin MIC₅₀ \leq 1 μ g/ml and MIC₉₀ \leq 2 μ g/ml. All tested gram-negative bacteria had prulifloxacin MIC₅₀ \leq 1 μ g/ml and MIC₉₀ \geq 2 μ g/ml. *In vitro* susceptibility tests of prulifloxacin determined by Kirby-Bauer disk diffusion and agar dilution were well correlated. (*J Infect Dis Antimicrob Agents* 2010;27:61-8.)

INTRODUCTION

Prulifloxacin is a new oral fluoroquinolone with broad spectrum of *in vitro* activity against various gram-positive and gram-negative bacteria.¹⁻² Prulifloxacin is a lipophilic prodrug of ulifloxacin. After absorption, prulifloxacin is metabolized by esterases to ulifloxacin which is generally more active than other fluoroquinolones against a variety of bacterial clinical isolates and also has the lowest potential of inducing

emergence of resistance. Prulifloxacin exhibited good penetration in target tissues and fluids including prostate gland, lung and gynaecological tissues.⁵⁻⁷ Prulifloxacin has been available in some countries including Japan and Italy for many years. Prulifloxacin was approved by Thai Food and Drug Administration in 2009.

The objective of the study was to determine *in vitro* activity of prulifloxacin against common bacteria

Division of Infectious Diseases and Tropical Medicine, Department of Medicine, Faculty of Medicine Siriraj Hospital, Mahidol University, Bangkok 10700, Thailand.

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Reprint request: Visanu Thamlikitkul, M.D., Division of Infectious Diseases and Tropical Medicine, Department of Medicine, Faculty of Medicine Siriraj Hospital, Mahidol University, Bangkok 10700, Thailand.

E-mail: sivth@mahidol.ac.th

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isolated from the patients at Siriraj Hospital.

MATERIALS AND METHODS

Study Organisms

Clinical isolates of β -hemolytic streptococcus group A (N=18), *S. pneumoniae* (N=46), methicillin-susceptible *S. aureus* (N=20), ESBL-non-producing *E. coli* (N=46), ESBL-producing *E. coli* (N=30), ESBL-non-producing *K. pneumoniae* (N=30), ESBL-producing *K. pneumoniae* (N=30), *P. aeruginosa* (N=30) and *Salmonella* spp. (N=7) were included. The studied organisms were isolated from blood, urine, sputum or pus of the hospitalized patients at Siriraj Hospital who had infections.

Susceptibility Test

The activity of prulifloxacin, levofloxacin, ciprofloxacin and moxifloxacin against the studied organisms was determined by Kirby-Bauer disk diffusion. The prulifloxacin disk (5 μ g) [Eiken Chemical, Japan], levofloxacin disk (5 μ g), ciprofloxacin disk (5 μ g) and moxifloxacin disk (5 μ g) [Oxoid, UK] were used. Determination of minimum inhibitory concentration (MIC) of prulifloxacin was also done by agar dilution. The prulifloxacin disk and standard powder were provided by Meiji Pharmaceuticals (Thailand). Quality control was performed by testing the susceptibility of *S. aureus* ATCC 25923, *S. pneumoniae* ATCC 49619, *E. coli* ATCC 25922 and *P. aeruginosa* ATCC 27853. The methodology for susceptibility testing was by direct colony suspension or growth method as recommended by the CLSI.⁵ Gram-positive isolate was grown overnight on blood agar at 35°C and its colonies were picked up and suspended in Mueller-Hinton broth equivalent to 0.5 McFarland standard. Gram-negative isolate was grown overnight on blood agar at 35°C and its colonies were picked up and suspended in Mueller-Hinton broth that

was incubated for several hours at 35°C in order to achieve a turbidity of 0.5 McFarland standard. The suspension was inoculated on Mueller-Hinton agar for *S. aureus* and gram-negative bacilli and Mueller-Hinton agar supplemented with 5 percent sheep blood for *S. pneumoniae* and β -hemolytic *Streptococcus* group A. The susceptibility disks were placed on the inoculated agar plate according to the manufacturer's recommendations. The agar plates were incubated at 35°C (5% CO₂ for *S. pneumoniae* and β -hemolytic *Streptococcus* group A) for 16-18 hours before the inhibition zone diameters were measured. The interpretative criteria for susceptibility of the aforementioned bacteria to prulifloxacin, levofloxacin, ciprofloxacin and moxifloxacin are shown in Table 1.

RESULTS

The inhibition zone diameters and MICs of the quality control strains are shown in Table 2 and Table 3. The susceptibility profiles of β -hemolytic streptococci group A, *S. pneumoniae*, *S. aureus*, ESBL-non-producing *K. pneumoniae*, ESBL-non-producing *E. coli*, ESBL-producing *K. pneumoniae*, ESBL-producing *E. coli*, *P. aeruginosa* and *Salmonella* spp. are shown in Table 4 and 5. Prulifloxacin seems to have in vitro activity against bacteria isolated from Thai patients similar to that of other fluoroquinolones. MIC data of prulifloxacin against β -hemolytic streptococci group A, *S. pneumoniae*, *S. aureus*, ESBL-non-producing *K. pneumoniae*, ESBL-non-producing *E. coli*, *P. aeruginosa* and *Salmonella* spp. are shown in Table 6. *In vitro* susceptibility test of prulifloxacin determined by Kirby-Bauer disk diffusion and agar dilution against β -hemolytic streptococci group A, *S. pneumoniae*, *S. aureus*, ESBL-non-producing *K. pneumoniae*, ESBL-non-producing *E. coli*, *P. aeruginosa* and *Salmonella* spp. are well correlated as shown in Table 4, 5 and 6.

Table 1. Zone diameter interpretative criteria for disk diffusion susceptibility of prulifloxacin, levofloxacin, ciprofloxacin and moxifloxacin.

Antibiotic (disc content)	Zone diameter breakpoints (mm.)*		
	Susceptible	Intermediate	Resistant
Prulifloxacin (5 µg/disc)	≥ 16	13 - 15	≤ 12
Levofloxacin (5 µg/disc)			
- Gram-positive cocci	≥ 19	16 - 18	≤ 15
- Gram-negative bacilli	≥ 17	14 - 16	≤ 13
Ciprofloxacin (5 µg/disc)			
- Gram-negative bacilli and <i>Staphylococcus</i> spp.	≥ 21	16 - 20	≤ 15
Moxifloxacin (5 µg/disc)			
- Gram-positive cocci	≥ 24	21 - 23	≤ 20

*Zone diameter breakpoints of prulifloxacin from Eiken Chemical, Japan,

Zone diameter breakpoints of levofloxacin, ciprofloxacin and moxifloxacin from CLSI 2010

DISCUSSION

Our findings indicated that the activity of prulifloxacin against gram-positive cocci commonly caused community-acquired infections was similar to that of levofloxacin and moxifloxacin and it seemed to be better than ciprofloxacin. Prulifloxacin was as active as levofloxacin and ciprofloxacin against *E. coli*, *K. pneumoniae* and *P. aeruginosa*. However, only 65 percent to 77 percent of ESBL-non-producing *E. coli*, ESBL-non-producing *K. pneumoniae* and *P. aeruginosa* were susceptible to prulifloxacin because all isolates were collected from the hospitalized patients and nosocomial isolates of gram-negative are usually more resistant to antibiotics when compared with community-acquired isolates. Therefore, prulifloxacin should be more active against community-acquired strains of *E. coli* and *K. pneumoniae*. Prulifloxacin,

like other fluoroquinolones, was not active against most of ESBL-producing gram-negative bacilli. The aforementioned observations were similar to the previous reports from other countries.^{1,2} Prulifloxacin was found to be very active against a worldwide collection of gastroenteritis-producing pathogens, including those causing traveler's diarrhea, such as *E. coli*, *Salmonella* spp., *Shigella* spp., *Yersinia* spp., *Vibrio* spp., *Aeromonas* spp., *Plesiomonas* spp. and *Campylobacter* spp.⁷ Prulifloxacin was reported to be efficacious and safe for therapy of patients with acute exacerbations of chronic bronchitis, uncomplicated and complicated urinary tract infections, chronic prostatitis due to bacteria and *Chlamydia trachomatis* and prophylaxis of infection after transrectal prostate biopsy.⁸⁻¹³ Prulifloxacin has an acceptable toxicity profile, comparable to that of other fluoroquinolones.

Table 2. Inhibition zone diameters of prulifloxacin, ciprofloxacin, levofloxacin and moxifloxacin against the quality control organisms.

Control Strain	Antibiotic	Inhibition Zone Diameter (mm)	
		CLSI 2010	Tested Result
<i>S. aureus</i> ATCC 25923	Prulifloxacin	20 - 26	26
	Ciprofloxacin	22 - 30	26
	Levofloxacin	25 - 30	28
	Moxifloxacin	28 - 35	31
<i>S. pneumoniae</i> ATCC 49619	Levofloxacin	20 - 25	23
	Moxifloxacin	25 - 31	29
<i>E. coli</i> ATCC 25922	Prulifloxacin	32 - 38	34, 34
	Ciprofloxacin	30 - 40	33, 34
	Levofloxacin	29 - 37	31, 31
	Moxifloxacin	28 - 35	29, 29
<i>P. aeruginosa</i> ATCC 27853	Prulifloxacin	27 - 33	33
	Ciprofloxacin	25 - 33	33
	Levofloxacin	19 - 26	26
	Moxifloxacin	17 - 25	22

Table 3. MICs of prulifloxacin against the quality control organisms.

Organisms	MIC range ($\mu\text{g/ml}$)	Tested MIC ($\mu\text{g/ml}$)
<i>E. coli</i> ATCC 25922	0.008 - 0.06	0.03
<i>P. aeruginosa</i> ATCC 27853	0.125 - 1.0	0.5
<i>S. aureus</i> ATCC 29213	0.125 - 0.5	0.25

Table 4. Susceptibility of gram-positive cocci to prulifloxacin, ciprofloxacin, levofloxacin and moxifloxacin by Kirby-Bauer disk diffusion.

Organism	Antibiotic	Total	Susceptible	Intermediate	Resistant				
<i>S. aureus</i> (MSSA)	Prulifloxacin	20	20	100	0	0	0	0	0
	Ciprofloxacin	20	19	95	1	5	0	0	0
	Levofloxacin	20	20	100	0	0	0	0	0
	Moxifloxacin	20	20	100	0	0	0	0	0
<i>S. pneumoniae</i> (PSSP)	Prulifloxacin	15	15	100	0	0	0	0	0
	Ciprofloxacin*	15	14	93.3	1	6.7	0	0	0
	Levofloxacin	15	15	100	0	0	0	0	0
	Moxifloxacin	15	15	100	0	0	0	0	0
<i>S. pneumoniae</i> (PISP)	Prulifloxacin	17	17	100	0	0	0	0	0
	Ciprofloxacin*	17	16	94.1	1	5.9	0	0	0
	Levofloxacin	17	17	100	0	0	0	0	0
	Moxifloxacin	17	17	100	0	0	0	0	0
<i>S. pneumoniae</i> (PRSP)	Prulifloxacin	14	14	100	0	0	0	0	0
	Ciprofloxacin*	14	14	100	0	0	0	0	0
	Levofloxacin	14	14	100	0	0	0	0	0
	Moxifloxacin	14	14	100	0	0	0	0	0
β -hemolytic streptococcus group A	Prulifloxacin	18	18	100	0	0	0	0	0
	Ciprofloxacin*	18	18	100	0	0	0	0	0
	Levofloxacin	18	18	100	0	0	0	0	0
	Moxifloxacin	18	18	100	0	0	0	0	0

PSSP=Penicillin susceptible *S. pneumoniae*PISP=Penicillin intermediately-susceptible *S. pneumoniae*PRSP=Penicillin resistant *S. pneumoniae** Using the same breakpoint as *S. aureus*

Table 5. Susceptibility of gram-negative bacilli to prulifloxacin, ciprofloxacin and levofloxacin by Kirby-Bauer disk diffusion.

Organism	Antibiotic	Total	Susceptible		Intermediate		Resistant	
			N	%	N	%	N	%
<i>P. aeruginosa</i>	Prulifloxacin	30	23	76.7	0	0	7	23.3
	Ciprofloxacin	30	23	76.7	0	0	7	23.3
	Levofloxacin	30	21	70.0	1	3.3	8	26.7
<i>K. pneumoniae</i> ESBL +ve	Prulifloxacin	30	8	26.7	5	16.6	17	56.7
	Ciprofloxacin	30	7	23.3	1	3.3	22	73.4
	Levofloxacin	30	7	23.3	1	3.3	22	73.4
<i>K. pneumoniae</i> ESBL-ve	Prulifloxacin	30	21	70.0	2	6.7	7	23.3
	Ciprofloxacin	30	18	60.0	4	13.3	8	26.7
	Levofloxacin	30	23	76.7	0	0	7	23.3
<i>E. coli</i> ESBL+ve	Prulifloxacin	30	7	23.3	0	0	23	76.7
	Ciprofloxacin	30	4	13.3	2	6.7	24	80.0
	Levofloxacin	30	6	20.0	0	0	24	80.0
<i>E. coli</i> ESBL-ve	Prulifloxacin	46	30	65.2	1	2.2	15	32.6
	Ciprofloxacin	46	29	63.0	0	0	17	37.0
	Levofloxacin	46	29	63.0	0	0	17	37.0
<i>Salmonella</i> spp.	Prulifloxacin	7	7	100	0	0	0	0
	Ciprofloxacin	7	6	85.7	1	14.3	0	0
	Levofloxacin	7	7	100	0	0	0	0

Table 6. MICs of prulifloxacin against tested organisms.

Organisms (N)	MIC ($\mu\text{g/ml}$)			% Susceptible*
	Range	MIC ₅₀	MIC ₉₀	
Gram-negative				
<i>P. aeruginosa</i> (30)	0.5 - >16	0.5	>16	76.7
<i>E. coli</i> ESBL -ve (46)	0.06 - >16	0.125	>16	65.2
<i>K. pneumoniae</i> ESBL -ve (30)	0.06 - >16	0.25	>16	70
<i>Salmonella</i> spp. (7)	0.125 - 2	1	2	100
Gram-positive				
<i>S. aureus</i> MSSA (20)	0.125 - 2	1	2	100
<i>S. pneumoniae</i> : PSSP (15)	0.5 - 2	1	2	100
<i>S. pneumoniae</i> : PISP (17)	0.5 - 2	1	2	100
<i>S. pneumoniae</i> : PRSP (14)	1 - 2	1	2	100
β -hemolytic streptococci group A (18)	0.25 - 0.5	0.25	0.25	100

*MIC breakpoint (Japanese Society of Chemotherapy): ≤ 2 ($\mu\text{g/ml}$) = Susceptible

The reported side effects include gastric disturbances, diarrhea, nausea and skin rash of mild-to-moderate severity.^{1,2} Prulifloxacin at steady state after therapeutic doses had no significant effects on the QTc interval.¹⁴ Prulifloxacin could preserve the normal vaginal lactobacillus microflorae in healthy women after they received repeated administration of prulifloxacin 600 mg tablets.¹⁵ Prulifloxacin has a long elimination half-life, allowing once-daily administration. The recommended dosage of prulifloxacin is 200 to 300 mg twice a day or 600 mg once a day. Prulifloxacin could be an oral antimicrobial therapy for mild to moderate infections and maintenance therapy after parenteral antimicrobial

treatment of severe infections in Thai patients.

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