Fluoroquinolone Breakpoints for *Enterobacteriaceae* and *Pseudomonas aeruginosa*

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1 Foreword

The Clinical and Laboratory Standards Institute (CLSI) is a not-for-profit membership organization that brings together the varied perspectives and expertise of the worldwide laboratory community for the advancement of a common cause: to foster excellence in laboratory medicine by developing and implementing medical laboratory standards and guidelines that help laboratories fulfill their responsibilities with efficiency, effectiveness, and global applicability.

Using the CLSI voluntary consensus process, the Subcommittee on Antimicrobial Susceptibility Testing develops standards that promote accurate antimicrobial susceptibility testing and appropriate reporting. The subcommittee reviews data from various sources and studies (eg, *in vitro*, pharmacokinetic-pharmacodynamic [PK-PD], and clinical studies) to establish antimicrobial susceptibility test methods, breakpoints, and quality control (QC) ranges.

The details of the necessary and recommended data for selecting appropriate breakpoints and QC ranges, and how the data are presented for evaluation, are described in CLSI document M23.[1] CLSI antibacterial breakpoints are provided in CLSI documents M100[2] and M45.[3]

Over time, a microorganism's susceptibility to an antimicrobial agent may decrease, resulting in a lack of clinical efficacy and/or safety. In addition, microbiological methods, QC parameters, and the manner in which breakpoints are established may be refined to ensure more accurate results. Because of these types of changes, CLSI continually monitors and updates information in its documents. Although CLSI standards and guidelines are developed using the most current information available at the time, the field of science and medicine is always changing; therefore, standards and guidelines should always be used in conjunction with clinical judgment, current knowledge, and clinically relevant laboratory test results to guide patient treatment. For more information, visit www.clsi.org.

This CLSI rationale document is based on CLSI agenda items submitted by the CLSI-EUCAST Joint Fluoroquinolone Ad Hoc Working Group.

2 Introduction

Ciprofloxacin and levofloxacin are members of the fluoroquinolone group of antimicrobial agents. The fluoroquinolones possess a fluorinated 4-quinolone nucleus.[4] The bactericidal action of fluoroquinolones results from inhibition of the enzymes topoisomerase II (DNA gyrase) and topoisomerase IV, which are necessary for bacterial DNA replication, transcription, repair, and recombination.[5] The fluoroquinolones, including ciprofloxacin and levofloxacin, have *in vitro* activity against gram-negative and gram-positive bacteria, including the *Enterobacteriaceae, Pseudomonas aeruginosa, Acinetobacter baumannii, Yersinia pestis,* methicillin-susceptible *Staphylococcus aureus, Streptococcus pneumoniae* (including multidrug-resistant organisms), and *Bacillus anthracis*.[5,6]

Fluoroquinolone resistance can arise through mutations in defined regions of DNA gyrase or topoisomerase IV (termed the quinolone resistance–determining regions), decreased outer membrane permeability, or altered efflux.[6] Plasmid-mediated resistance mechanisms, including the *qnr* genes, have increased in frequency among some *Enterobacteriaceae*. Resistance to ciprofloxacin due to spontaneous mutations occurs at a general frequency of between $< 10^{-9}$ to 10^{-6}.[5] Resistance to levofloxacin due to spontaneous mutation *in vitro* is a rare occurrence (ie, range 10^{-10} to 10^{-9}).[6]

Ciprofloxacin and levofloxacin are approved by the US Food and Drug Administration for the treatment of acute or chronic infections due to gram-positive and gram-negative bacteria, including nosocomial and community-acquired pneumonia, skin and skin structure infections, urinary tract infections, chronic bacterial prostatitis, inhalation anthrax, and plague. Fluoroquinolones have been associated with disabling and potentially irreversible serious adverse reactions, including tendinitis and tendon rupture, peripheral neuropathy, and central nervous system effects. Therefore, for uncomplicated urinary tract infection, acute bacterial exacerbation of chronic bronchitis, and acute bacterial sinusitis, use should be reserved for patients with no alternative treatment options.[5,6]

For current and past fluoroquinolone breakpoints for *Enterobacteriaceae* and *P. aeruginosa*, see Tables 1 and 2, respectively.

Table 1. Current CLSI Fluoroquinolone Breakpoints*

Organism Group	Antimicrobial Agent	S	SDD	I	R
Enterobacteriaceae	Ciprofloxacin	≤ 0.25	-	0.5	≥ 1
	Levofloxacin	≤ 0.5	-	1	≥ 2
P. aeruginosa	Ciprofloxacin	≤ 0.5	N/A	1	≥ 2
	Levofloxacin	≤ 1	N/A	2	≥ 4

* Last reviewed January 2018; first published in CLSI document M100, 29th ed.[2]
Abbreviations: I, intermediate; N/A, not applicable; R, resistant; S, susceptible; SDD, susceptible-dose dependent.

Table 2. Historical CLSI Fluoroquinolone Breakpoints Replaced by Current Fluoroquinolone Breakpoints*

Organism Group	Antimicrobial Agent	S	SDD	I	R
Enterobacteriaceae	Ciprofloxacin	≤ 1	-	2	≥ 4
	Levofloxacin	≤ 2	-	4	≥ 8
P. aeruginosa	Ciprofloxacin	≤ 1	N/A	2	≥ 4
	Levofloxacin	≤ 2	N/A	4	≥ 8

* Last published in CLSI document M100, 28th ed.
Abbreviations: I, intermediate; N/A, not applicable; R, resistant; S, susceptible; SDD, susceptible-dose dependent.

3 Standard Dosages and Pharmacokinetic Data

Ciprofloxacin and levofloxacin dosages used to determine breakpoints are shown in Table 3.

Table 3. Dosages Used for Breakpoint Determination*

Organism Group	Drug	Dose
Enterobacteriaceae	Ciprofloxacin	400 mg IV or 500 mg PO administered every 12 hours
	Levofloxacin	750 mg IV or PO every administered every 24 hours
P. aeruginosa	Ciprofloxacin	400 mg IV administered every 8 hours
	Levofloxacin	750 mg IV or PO administered every 24 hours

* See CLSI document M100.[2]
Abbreviations: IV, intravenous; PO, oral.

Population pharmacokinetic (PK) parameters for ciprofloxacin and levofloxacin are shown in Tables 4, 5, and 6. For levofloxacin, Table 5 parameters are based on data derived from patients treated for community-acquired infections, while Table 6 parameters are derived from patients with life-threatening infections treated in the hospital setting.

Table 4. Population PK Parameters for Ciprofloxacin by Iterative Two-Stage Analysis[7] (Reprinted from Forrest A, Ballow CH, Nix DE, Birmingham MC, Schentag JJ, Development of a population pharmacokinetic model and optimal sampling strategies for intravenous ciprofloxacin, *Antimicrob Agents Chemother*, 1993, Vol 37/No 5, pp. 1065-1072, doi: 10.1128/AAC.37.5.1065. Reproduced with permission from American Society for Microbiology.)

Parameter	Mean	Interpatient % CV	Range
V_C (L/kg)	0.69	26	0.2–1.2
V_P (L/kg)	0.51	33	0.2–2.0
V_β (L/kg)	2.0	31	0.96–5.0
CL_D (L/h/1.73 m^2)	38	24	16–64
CL_T (L/h/1.73 m^2)	17	44	4.4–37
$T_{1/2\beta}$ (h)	6.5	50	1.6–22

Abbreviations: % CV, coefficient of variation expressed as a percentage; CL_D, distributional clearance of the central compartment; CL_T, total plasma clearance; PK, pharmacokinetic; $T_{1/2\beta}$, terminal half-life; V_β, volume of distribution that when associated with terminal rate constant for elimination will provide the correct clearance; V_C, central volume of distribution; V_P, distributional clearance of the peripheral compartment.

Table 5. Population PK Parameters for Levofloxacin ($N = 272$)[8] (Reprinted from Preston SL, Drusano GL, Berman AL, et al., Levofloxacin population pharmacokinetics and creation of a demographic model for prediction of individual drug clearance in patients with serious community-acquired infection, *Antimicrob Agents Chemother*, 1998, Vol 42/No 5, pp. 1098-1104, doi: 10.1128/AAC.42.5.1098. Reproduced with permission from American Society for Microbiology.)

Unit	K_{CP}, h^{-1}	K_{PC}, h^{-1}	VS, L/kg	CL_T, L/h
Mean	0.487	0.647	0.836	9.27
Median	0.384	0.596	0.795	9.01
SD	0.378	0.391	0.429	4.31

Abbreviations: CL_T, total plasma clearance; K_{CP}, transfer rate between the central compartment and the peripheral compartment; K_{PC}, transfer rate between the peripheral compartment and the central compartment; PK, pharmacokinetic; SD, standard deviation; VS, slope of the mean volume of distribution of the central compartment to body weight.

Table 6. Population PK Parameter Values Derived From 58 Patients With Nosocomial Pneumonia Receiving Levofloxacin (750 mg IV) as a 1.5-Hour Constant-Rate Infusion[9]
(Reprinted from Drusano GL, Preston SL, Fowler C, Corrado M, Weisinger B, Kahn J, Relationship between fluoroquinolone area under the curve: minimum inhibitory concentration ratio and the probability of eradication of the infecting pathogen, in patients with nosocomial pneumonia, *J Infect Dis*, 2004, Vol 189/No 9, pp. 1590-1597, by permission of Oxford University Press and the Infectious Diseases Society of America.)

Unit	V_C, L	K_{CP}, h^{-1}	K_{PC}, h^{-1}	CL_T, L/h
Mean	34.4	7.65	6.07	7.24
Median	23.3	2.66	0.924	6.24
SD	33.5	9.59	12.0	4.36

Abbreviations: CL_T, total plasma clearance; K_{CP}, transfer rate between the central compartment and the peripheral compartment; IV, intravenous; K_{PC}, transfer rate between the peripheral compartment and the central compartment; PK, pharmacokinetic; SD, standard deviation; V_C, central volume of distribution.

Mean estimates of area under the curve (AUC) or CLT and associated % CV for IV and oral ciprofloxacin and levofloxacin dosing regimens are shown in Tables 7 and 8. These estimates are based on data from infected patients, healthy subjects, and healthy subjects with inflated variance.

Table 7. Summary of Ciprofloxacin AUC or CL_T Estimates by IV or PO Dosing Regimen[10]
(Reprinted with permission from USCAST, The National Antimicrobial Susceptibility Testing Committee for the United States. Quinolone In Vitro Susceptibility Test Interpretive Criteria Evaluations. Version 1.3, 2018. http://www.uscast.org.)

Ciprofloxacin Dosing Regimen	Route of Administration	Population	Mean (% CV)		Reference
			AUC, mg • hr/L	CL_T, L/h/1.73 m^2	
400 mg every 12 hours	IV	Healthy subjects	25.0 (16.3*)		5
400 mg every 12 hours	IV	Infected patients		17.0 (44.0)	11
400 mg every 8 hours	IV	Healthy subjects	32.9 (16.3*)		5
400 mg every 8 hours	IV	Infected patients		17.0 (44.0)	11
500 mg every 12 hours	PO	Healthy subjects	27.4 (16.3*)		5
500 mg every 12 hours	PO	Healthy subjects with inflated variance	27.4 (44.0†)		
750 mg every 12 hours	PO	Healthy subjects	31.6 (16.3*)		
750 mg every 12 hours	PO	Healthy subjects with inflated variance	31.6 (44.0†)		

* Because the % CV was not available, a median value of 16.3, based on data from healthy volunteers for other IV or PO doses for each of the quinolones evaluated, was used.
† In the absence of mean AUC estimates for infected patients, estimates for healthy subjects and a % CV of 44.0 based on PK data from infected patients were used.[11]
Abbreviations: % CV, coefficient of variation expressed as a percentage; AUC, area under the curve; CL_T, total plasma clearance; IV, intravenous; PK, pharmacokinetic; PO, oral.

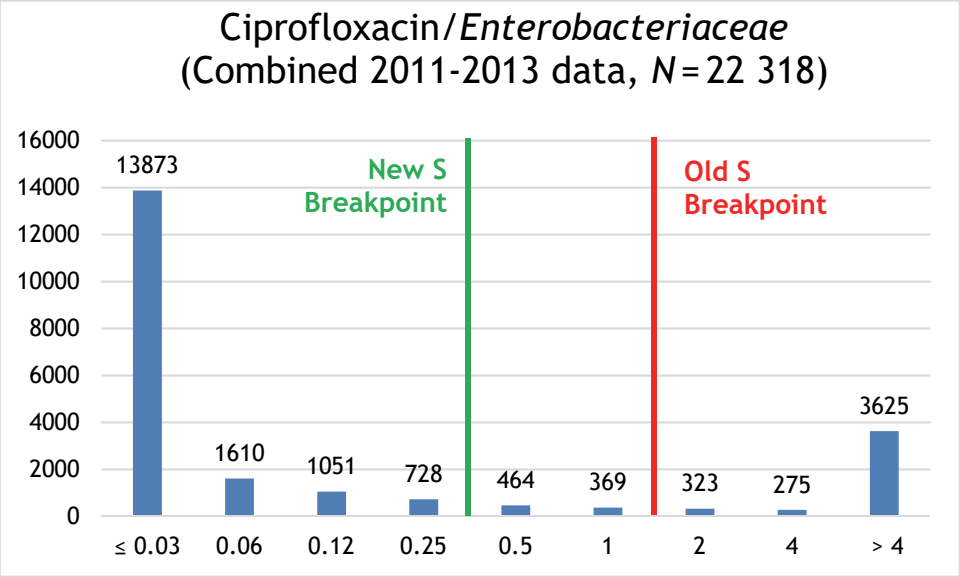
Table 8. Summary of Levofloxacin CL_T Estimates by IV or PO Dosing Regimen[10] (Reprinted with permission from USCAST, The National Antimicrobial Susceptibility Testing Committee for the United States. Quinolone In Vitro Susceptibility Test Interpretive Criteria Evaluations. Version 1.3, 2018. http://www.uscast.org.)

Levofloxacin Dosing Regimen	Route of Administration	Population	Mean (% CV) CL_T, L/h/1.73 m^2	Reference
500 mg every 24 hours	IV	Healthy subjects	9.48 (18.4)	6
500 mg every 24 hours	IV	Infected patients	9.27 (46.5)	8
750 mg every 24 hours	IV	Healthy subjects	7.56 (29.4)	6
750 mg every 24 hours	IV	Infected patients	7.24 (60.2)	9
500 mg every 24 hours	PO	Healthy subjects	10.5 (14.3)	6
750 mg every 24 hours	PO	Healthy subjects	8.58 (20.3)	
500 mg every 24 hours	PO	Healthy subjects with inflated variance	10.5 (44.0*)	
750 mg every 24 hours	PO	Healthy subjects with inflated variance	8.58 (44.0*)	

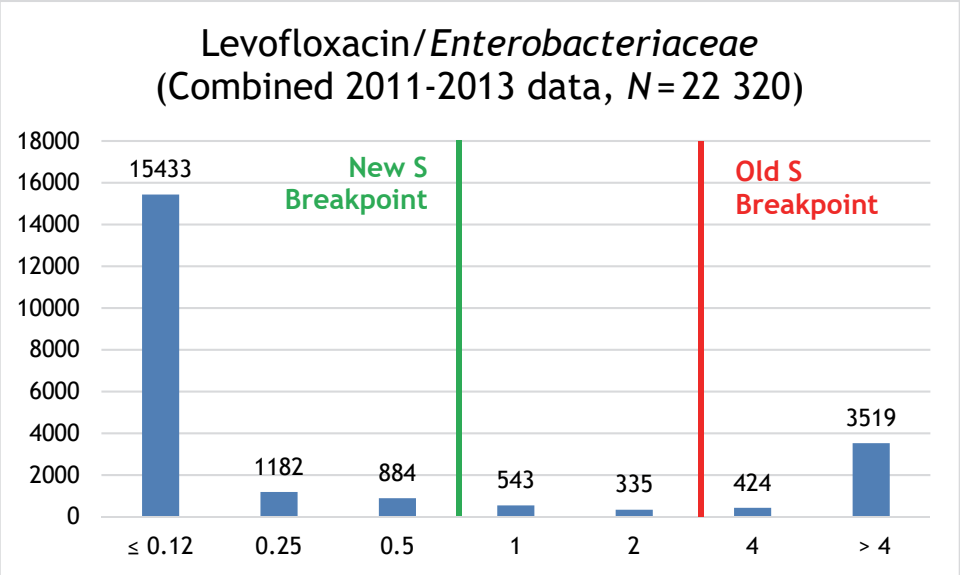
* In the absence of mean AUC estimates for infected patients, estimates for healthy subjects and a % CV of 44.0 based on PK data from infected patients were used.[11]
Abbreviations: % CV, coefficient of variation expressed as a percentage; AUC, area under the curve; CL_T, total plasma clearance; IV, intravenous; PK, pharmacokinetic; PO, oral.

4 Minimal Inhibitory Concentration Distribution Data

Minimal inhibitory concentration (MIC) distribution data from the United States from 2011 to 2013 were reviewed and are presented in Figures 1 to 4.[10]

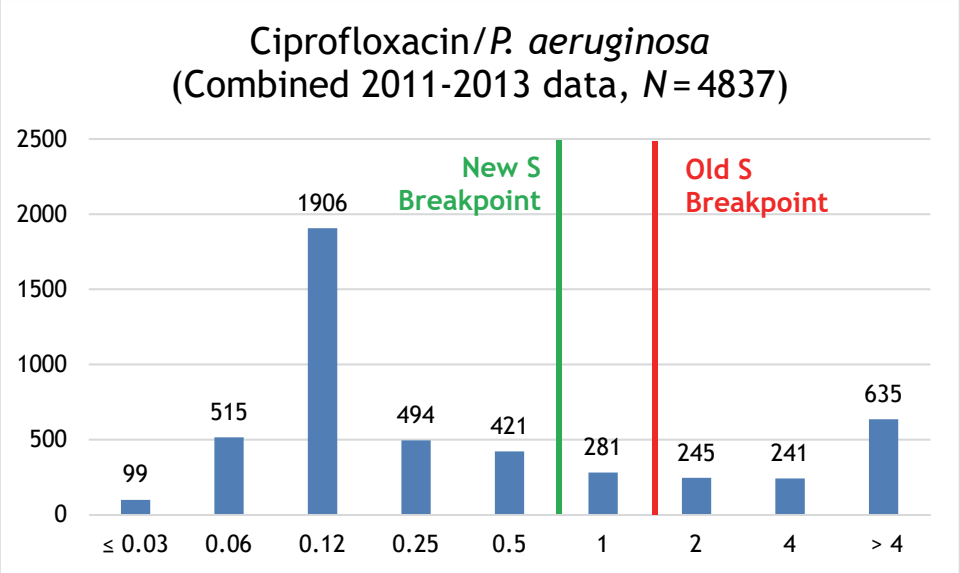


Abbreviations: MIC, minimal inhibitory concentration; S, susceptible.
Figure 1. MIC Distribution for *Enterobacteriaceae* and Ciprofloxacin[10]



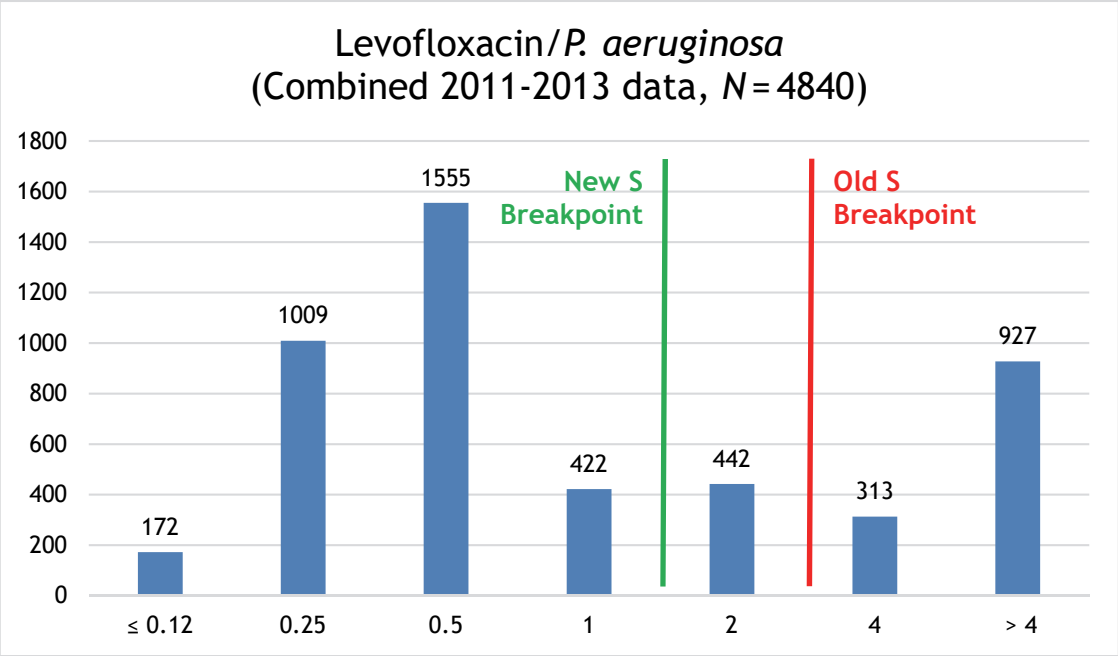
Abbreviations: MIC, minimal inhibitory concentration; S, susceptible.

Figure 2. MIC Distribution for *Enterobacteriaceae* and Levofloxacin[10]



Abbreviations: MIC, minimal inhibitory concentration; S, susceptible.

Figure 3. MIC Distribution for *P. aeruginosa* and Ciprofloxacin[10]

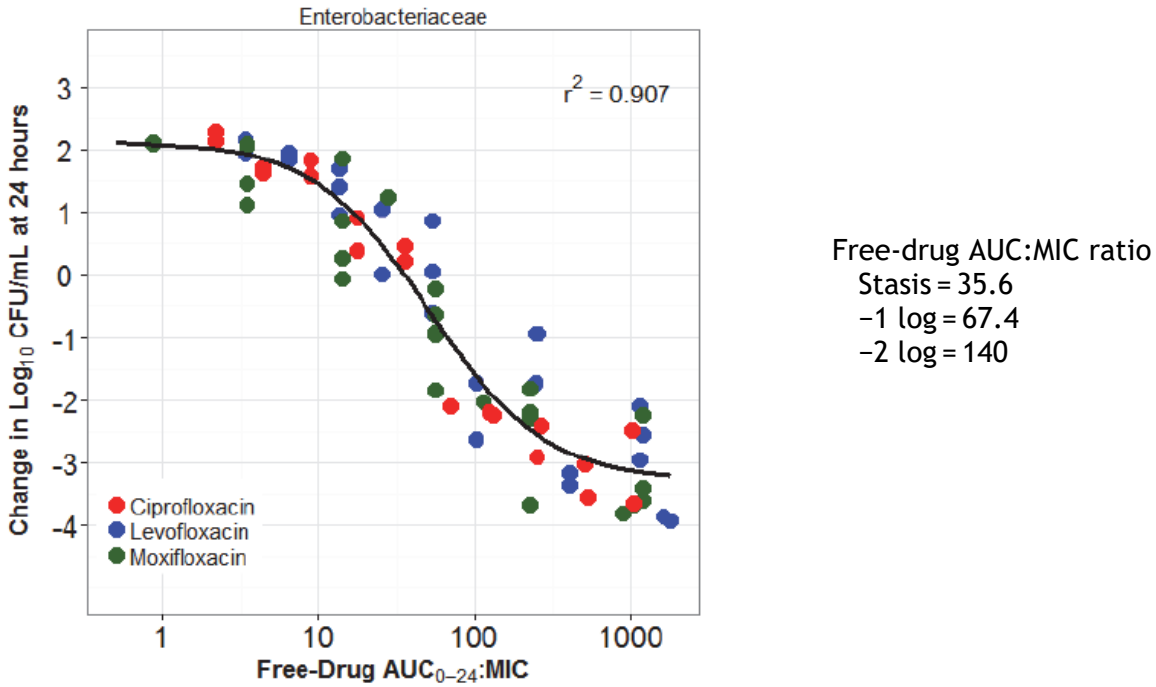


Abbreviations: MIC, minimal inhibitory concentration; S, susceptible.

Figure 4. MIC Distribution for *P. aeruginosa* and Levofloxacin[10]

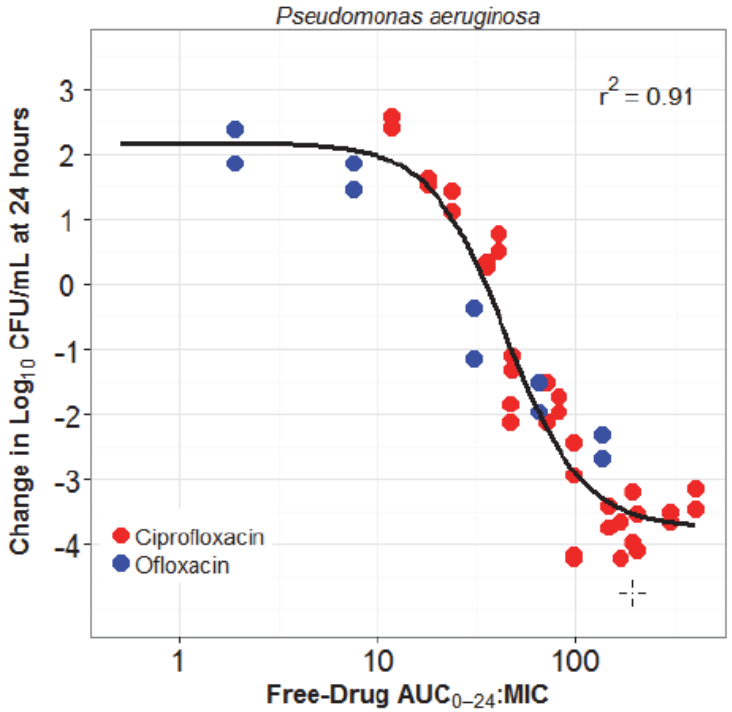
5 Pharmacodynamic Data

Nonclinical free-drug AUC:MIC ratio targets were obtained from neutropenic mouse thigh model data as shown in Figures 5 and 6.



Abbreviations: AUC, area under the curve; CFU, colony-forming unit; MIC, minimal inhibitory concentration.

Figure 5. Free-Drug AUC_{0-24}:MIC Ratio and Change in *Enterobacteriaceae* Bacterial Density - Neutropenic Mouse Thigh Model After 24 Hours[10] (Reprinted with permission from USCAST, The National Antimicrobial Susceptibility Testing Committee for the United States. Quinolone In Vitro Susceptibility Test Interpretive Criteria Evaluations. Version 1.3, 2018. http://www.uscast.org.)



Free-drug AUC:MIC ratio
Stasis = 34.8
−1 log = 47.3
−2 log = 65.4

Abbreviations: AUC, area under the curve; CFU, colony-forming unit; MIC, minimal inhibitory concentration.

Figure 6. Free-Drug AUC_{0-24}:MIC Ratio and Change in *P. aeruginosa* Bacterial Density - Neutropenic Mouse Thigh Model After 24 Hours[10] (Reprinted with permission from USCAST, The National Antimicrobial Susceptibility Testing Committee for the United States. Quinolone In Vitro Susceptibility Test Interpretive Criteria Evaluations. Version 1.3, 2018. http://www.uscast.org.)

Clinical AUC:MIC ratio targets are based on two published PK-PD efficacy analyses evaluating ciprofloxacin or levofloxacin for the treatment of patients with hospital-acquired pneumonia primarily due to gram-negative bacilli.[9,11] After adjusting for protein binding,[5,6] the total-drug AUC:MIC ratio targets for efficacy of 125 and 87 for ciprofloxacin and levofloxacin, respectively, obtained from the published literature, translate to free-drug AUC:MIC ratio targets of 87.5 and 61, respectively. From this, the clinical free-drug AUC:MIC ratio target of 72 was chosen to determine efficacy for *Enterobacteriaceae* and *P. aeruginosa*.[10]

Nonclinical and clinical free-drug AUC:MIC ratio targets are summarized in Table 9.

Table 9. Summary of Nonclinical and Clinical Free-Drug AUC:MIC Ratio Targets for Efficacy[10]
(Reprinted with permission from USCAST, The National Antimicrobial Susceptibility Testing Committee for the United States. Quinolone In Vitro Susceptibility Test Interpretive Criteria Evaluations. Version 1.3, 2018. http://www.uscast.org.)

Organism	Nonclinical Free-Drug AUC:MIC Ratio Targets			Clinical Free-Drug AUC:MIC Ratio Targets
	Net Bacterial Stasis	1-$\log_{10}$ CFU Reduction From Baseline	2-$\log_{10}$ CFU Reduction From Baseline	
Enterobacteriaceae	35.6	67.4	140.0	72.0
P. aeruginosa	34.8	47.3	65.4	72.0

Abbreviations: AUC, area under the curve; CFU, colony-forming unit; MIC, minimal inhibitory concentration.

Monte Carlo simulation data showing the percent probabilities of PK-PD target attainment for ciprofloxacin and levofloxacin are shown for *Enterobacteriaceae* in Table 10, Figure 7, and Figure 8 and for *P. aeruginosa* in Table 11, Figure 9, and Figure 10.

Table 10. Percent Probabilities of PK-PD Target Attainment by MIC Based on PK-PD Targets for *Enterobacteriaceae*[10] (Reprinted with permission from USCAST, The National Antimicrobial Susceptibility Testing Committee for the United States. Quinolone In Vitro Susceptibility Test Interpretive Criteria Evaluations. Version 1.3, 2018. http://www.uscast.org.)

Antimicrobial Agent	Route of Administration	Dosing Regimen	Population	MIC, µg/mL	End Points for Nonclinical Free-Drug AUC:MIC Ratio Targets (Magnitude of Target)			Clinical Free-Drug AUC:MIC Ratio Target (72)
					Net Bacterial Stasis (35.6)	1-$\log_{10}$ CFU Reduction From Baseline (67.4)	2-$\log_{10}$ CFU Reduction From Baseline (140)	
Ciprofloxacin	PO	500 mg every 12 hours	Healthy subjects with inflated variance	0.03	100	100	100	100
				0.06	100	100	95.8	100
				0.12	100	96.7	53.6	95.0
				0.25	94.4	53.3	4.16	47.2
				0.5	48.1	5.28	0.02	3.76
				1	3.88	0.04	0	0
				2	0	0	0	0
				4	0	0	0	0
				8	0	0	0	0
Ciprofloxacin	PO	750 mg every 12 hours	Healthy subjects with inflated variance	0.03	100	100	100	100
				0.06	100	100	98.0	100
				0.12	100	98.2	67.3	97.7
				0.25	97.2	67.1	9.08	61.0
				0.5	62.3	10.7	0.20	7.98
				1	8.52	0.30	0	0.140
				2	0.20	0	0	0
				4	0	0	0	0
				8	0	0	0	0

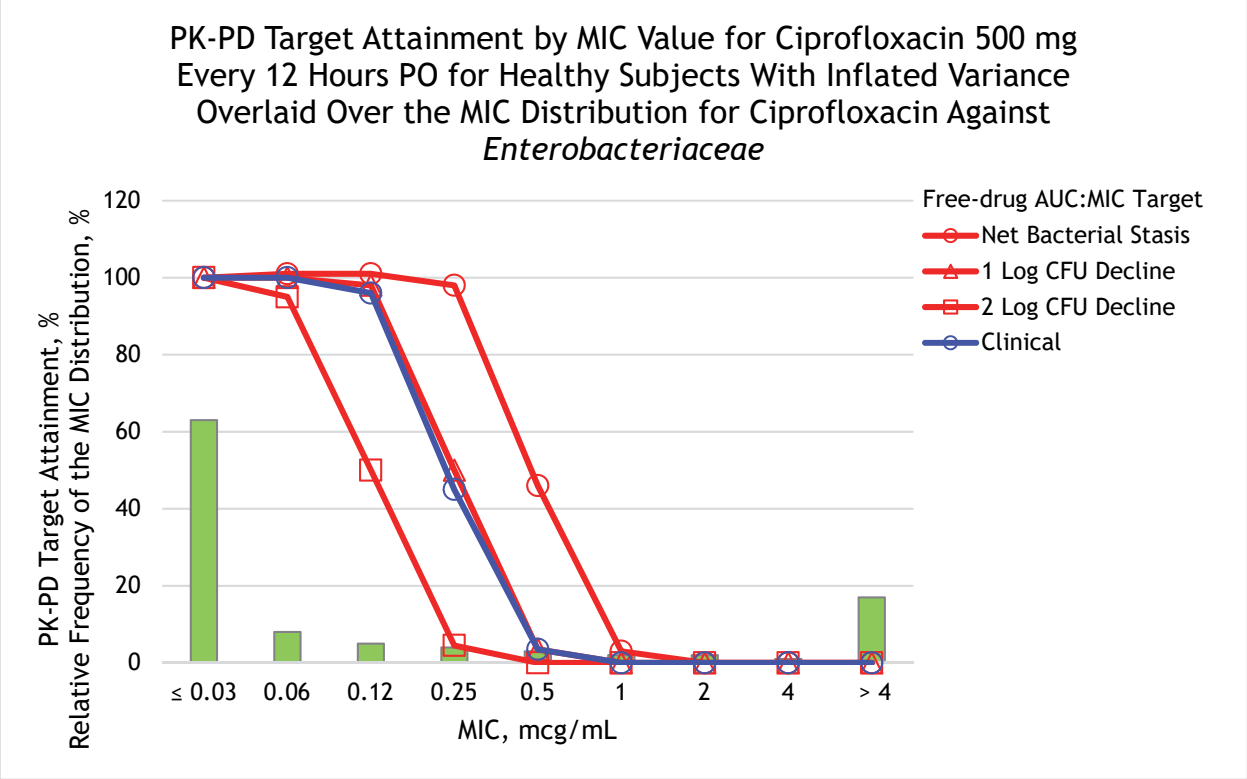
Table 10. (Continued)

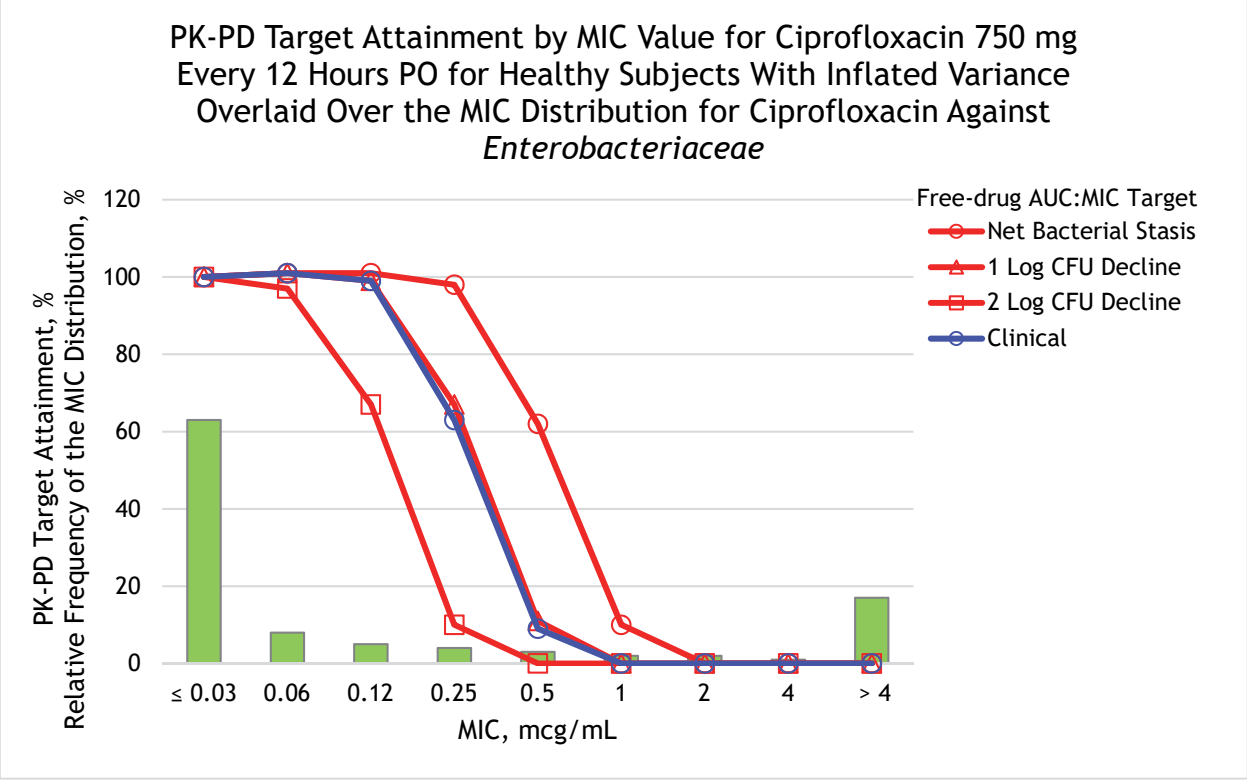
Antimicrobial Agent	Route of Administration	Dosing Regimen	Population	MIC, µg/mL	End Points for Nonclinical Free-Drug AUC:MIC Ratio Targets (Magnitude of Target)			Clinical Free-Drug AUC:MIC Ratio Target (72)
					Net Bacterial Stasis (35.6)	1-$\log_{10}$ CFU Reduction From Baseline (67.4)	2-$\log_{10}$ CFU Reduction From Baseline (140)	
Ciprofloxacin	IV	400 mg every 8 hours	Infected patients	0.03	100	100	100	100
				0.06	100	100	100	100
				0.12	100	100	99.6	100
				0.25	100	99.6	83.9	99.4
				0.5	99.4	86.0	26.6	82.3
				1	82.9	29.8	1.10	24.5
				2	25.5	1.42	0	0.94
				4	0.98	0	0	0
				8	0	0	0	0
Levofloxacin	PO	750 mg every 24 hours	Healthy subjects with inflated variance	0.03	100	100	100	100
				0.06	100	100	100	100
				0.12	100	100	100	100
				0.25	100	100	93.9	99.9
				0.5	99.9	94.9	44.9	93.0
				1	93.3	48.5	3.70	42.7
				2	43.6	4.46	0.06	3.16
				4	3.34	0.08	0	0.04
				8	0.04	0	0	0

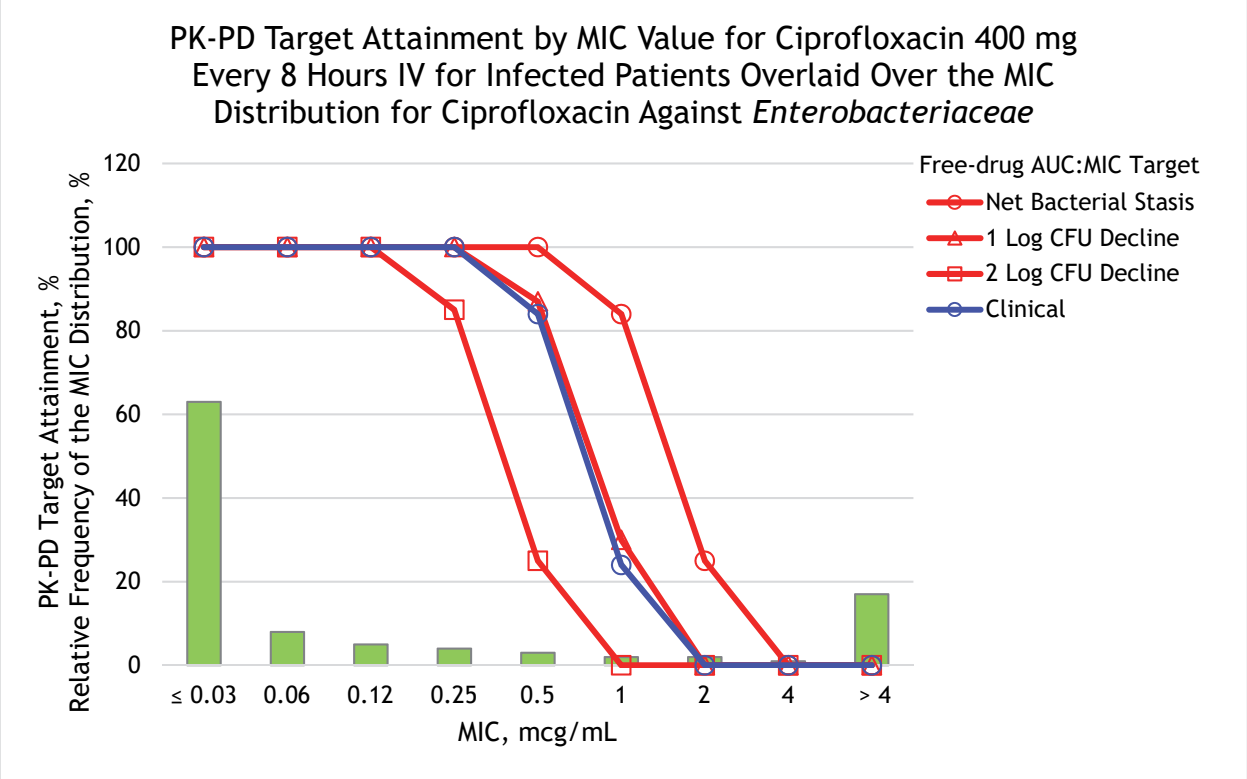
Table 10. (Continued)

Antimicrobial Agent	Route of Administration	Dosing Regimen	Population	MIC, µg/mL	End Points for Nonclinical Free-Drug AUC:MIC Ratio Targets (Magnitude of Target)			Clinical Free-Drug AUC:MIC Ratio Target (72)
					Net Bacterial Stasis (35.6)	1-$\log_{10}$ CFU Reduction From Baseline (67.4)	2-$\log_{10}$ CFU Reduction From Baseline (140)	
Levofloxacin	IV	750 mg every 24 hours	Infected patients	0.03	100	100	100	100
				0.06	100	100	100	100
				0.12	100	100	99.8	100
				0.25	100	99.8	94.4	99.7
				0.5	99.7	95.1	63.7	93.7
				1	94.0	66.0	18.5	61.5
				2	62.3	20.0	1.82	17.2
				4	17.5	2.08	0.06	1.68
				8	1.72	0.06	0	0.06

Abbreviations: AUC, area under the curve; CFU, colony-forming unit; IV, intravenous; MIC, minimal inhibitory concentration; PK-PD, pharmacokinetic-pharmacodynamic; PO, oral.

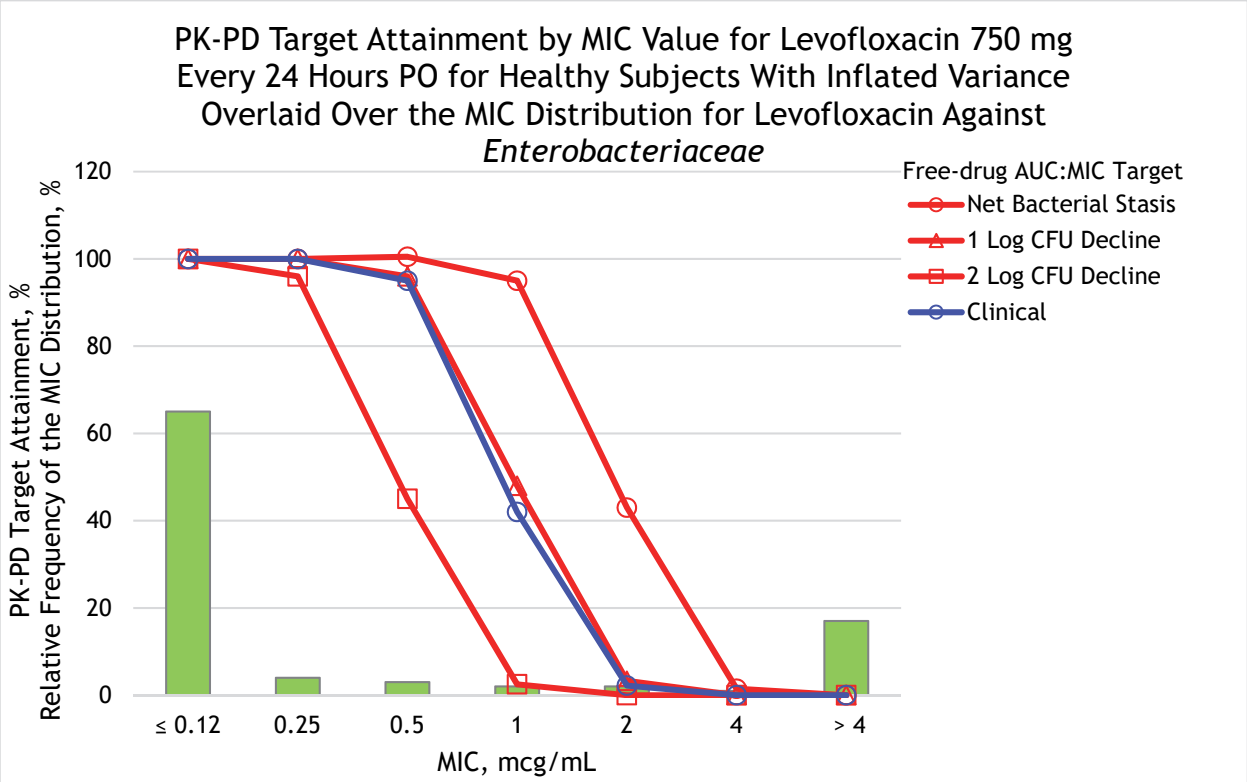


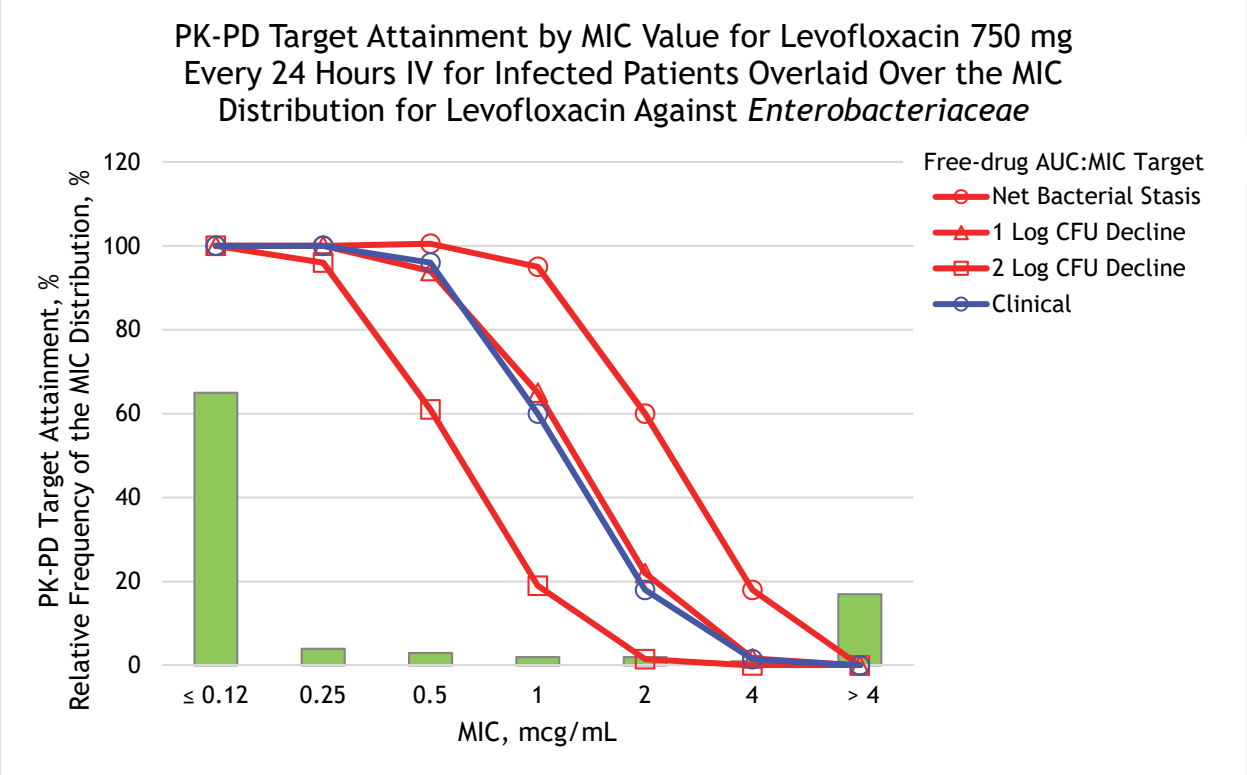




Abbreviations: AUC, area under the curve; CFU, colony-forming unit; IV, intravenous; MIC, minimal inhibitory concentration; PK-PD, pharmacokinetic-pharmacodynamic; PO, oral.

Figure 7. Percent Probabilities of Ciprofloxacin PK-PD Target Attainment Based on Free-Drug AUC:MIC Ratio Targets Relative to the MIC Distribution for *Enterobacteriaceae*[10]
(Reprinted with permission from USCAST, The National Antimicrobial Susceptibility Testing Committee for the United States. Quinolone In Vitro Susceptibility Test Interpretive Criteria Evaluations. Version 1.3, 2018. http://www.uscast.org.)





Abbreviations: AUC, area under the curve; CFU, colony-forming unit; IV, intravenous; MIC, minimal inhibitory concentration; PK-PD, pharmacokinetic-pharmacodynamic; PO, oral.

Figure 8. Percent Probabilities of Levofloxacin PK-PD Target Attainment Based on Free-Drug AUC:MIC Ratio Targets Relative to the MIC Distribution for *Enterobacteriaceae*[10]
(Reprinted with permission from USCAST, The National Antimicrobial Susceptibility Testing Committee for the United States. Quinolone In Vitro Susceptibility Test Interpretive Criteria Evaluations. Version 1.3, 2018. http://www.uscast.org.)

Table 11. Percent Probabilities of PK-PD Target Attainment by MIC Based on PK-PD Targets for *P. aeruginosa*[10] (Reprinted with permission from USCAST, The National Antimicrobial Susceptibility Testing Committee for the United States. Quinolone In Vitro Susceptibility Test Interpretive Criteria Evaluations. Version 1.3, 2018. http://www.uscast.org.)

Antimicrobial Agent	Route of Administration	Dosing Regimen	Population	MIC, µg/mL	End Points for Nonclinical AUC:MIC Ratio Targets (Magnitude of Target)			Clinical AUC:MIC Ratio Target (72)
					Net Bacterial Stasis (34.8)	1-$\log_{10}$ CFU Reduction From Baseline (47.3)	2-$\log_{10}$ CFU Reduction From Baseline (65.4)	
Ciprofloxacin	PO	500 mg every 12 hours	Healthy subjects with inflated variance	0.03	100	100	100	100
				0.06	100	100	100	100
				0.12	100	99.7	97.1	95.0
				0.25	94.9	82.6	56.0	47.2
				0.5	50.3	23.7	6.32	3.76
				1	4.34	0.76	0.06	0
				2	0.02	0	0	0
				4	0	0	0	0
				8	0	0	0	0
Ciprofloxacin	PO	750 mg every 12 hours	Healthy subjects with inflated variance	0.03	100	100	100	100
				0.06	100	100	100	100
				0.12	100	99.9	98.5	97.7
				0.25	97.5	89.5	69.6	61.0
				0.5	64.3	35.2	12.0	7.98
				1	9.30	2.04	0.34	0.14
				2	0.22	0	0	0
				4	0	0	0	0
				8	0	0	0	0

Table 11. (Continued)

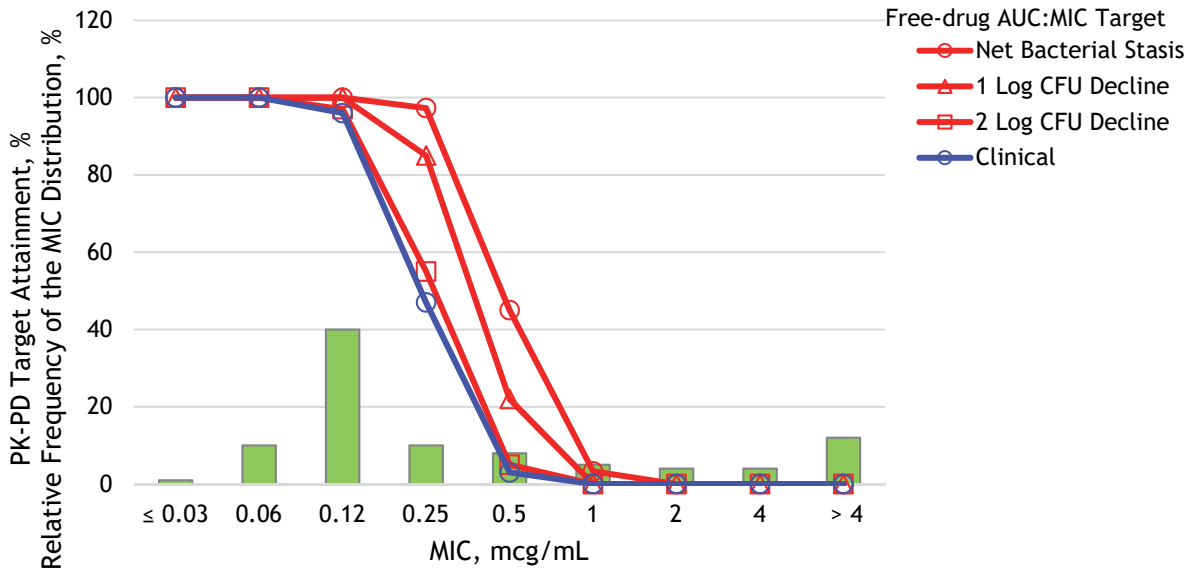
Antimicrobial Agent	Route of Administration	Dosing Regimen	Population	MIC, µg/mL	End Points for Nonclinical AUC:MIC Ratio Targets (Magnitude of Target)			Clinical AUC:MIC Ratio Target (72)
					Net Bacterial Stasis (34.8)	1-$\log_{10}$ CFU Reduction From Baseline (47.3)	2-$\log_{10}$ CFU Reduction From Baseline (65.4)	
Ciprofloxacin	IV	400 mg every 8 hours	Infected patients	0.03	100	100	100	100
				0.06	100	100	100	100
				0.12	100	100	100	100
				0.25	100	100	99.7	99.4
				0.5	99.6	97.2	87.4	82.3
				1	84.3	61.9	32.2	24.5
				2	27.1	9.52	1.78	0.94
				4	1.12	0.12	0	0
				8	0	0	0	0
Levofloxacin	PO	750 mg every 24 hours	Healthy subjects with inflated variance	0.03	100	100	100	100
				0.06	100	100	100	100
				0.12	100	100	100	100
				0.25	100	100	100	99.9
				0.5	99.9	99.4	95.6	93.0
				1	94.1	79.5	51.4	42.7
				2	45.4	20.4	5.28	3.16
				4	3.86	0.76	0.08	0.04
				8	0.06	0	0	0

Table 11. (Continued)

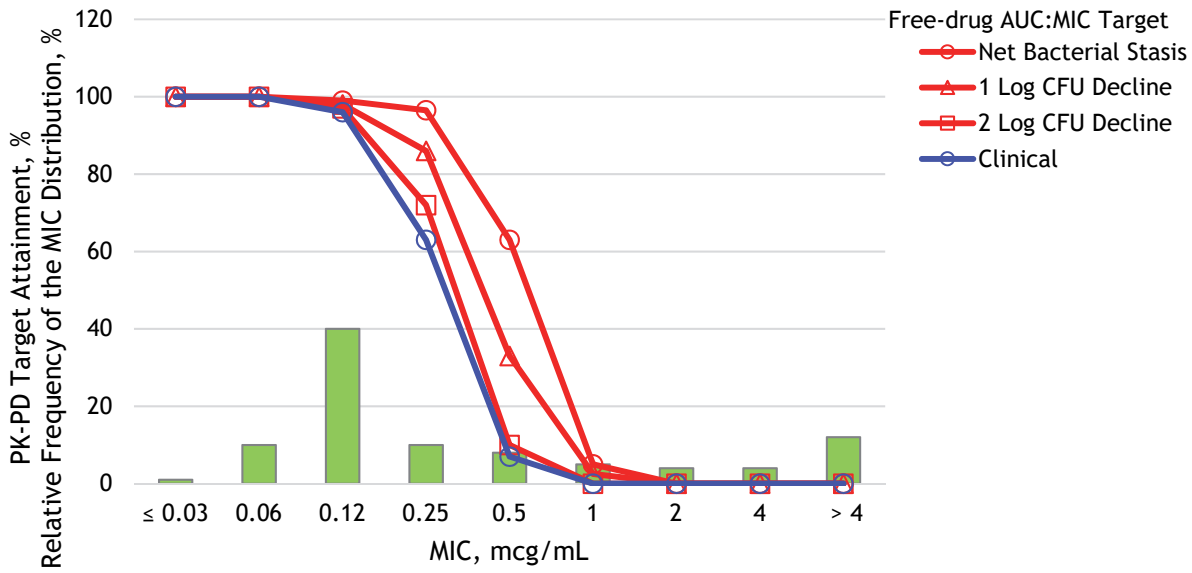
Antimicrobial Agent	Route of Administration	Dosing Regimen	Population	MIC, µg/mL	End Points for Nonclinical AUC:MIC Ratio Targets (Magnitude of Target)			Clinical AUC:MIC Ratio Target (72)
					Net Bacterial Stasis (34.8)	1-$\log_{10}$ CFU Reduction From Baseline (47.3)	2-$\log_{10}$ CFU Reduction From Baseline (65.4)	
Levofloxacin	IV	750 mg every 24 hours	Infected patients	0.03	100	100	100	100
				0.06	100	100	100	100
				0.12	100	100	100	100
				0.25	100	100	99.9	99.7
				0.5	99.8	98.8	95.5	93.7
				1	94.5	85.4	67.9	61.5
				2	64.1	41.8	21.4	17.2
				4	18.7	7.22	2.32	1.68
				8	1.84	0.42	0.06	0.06

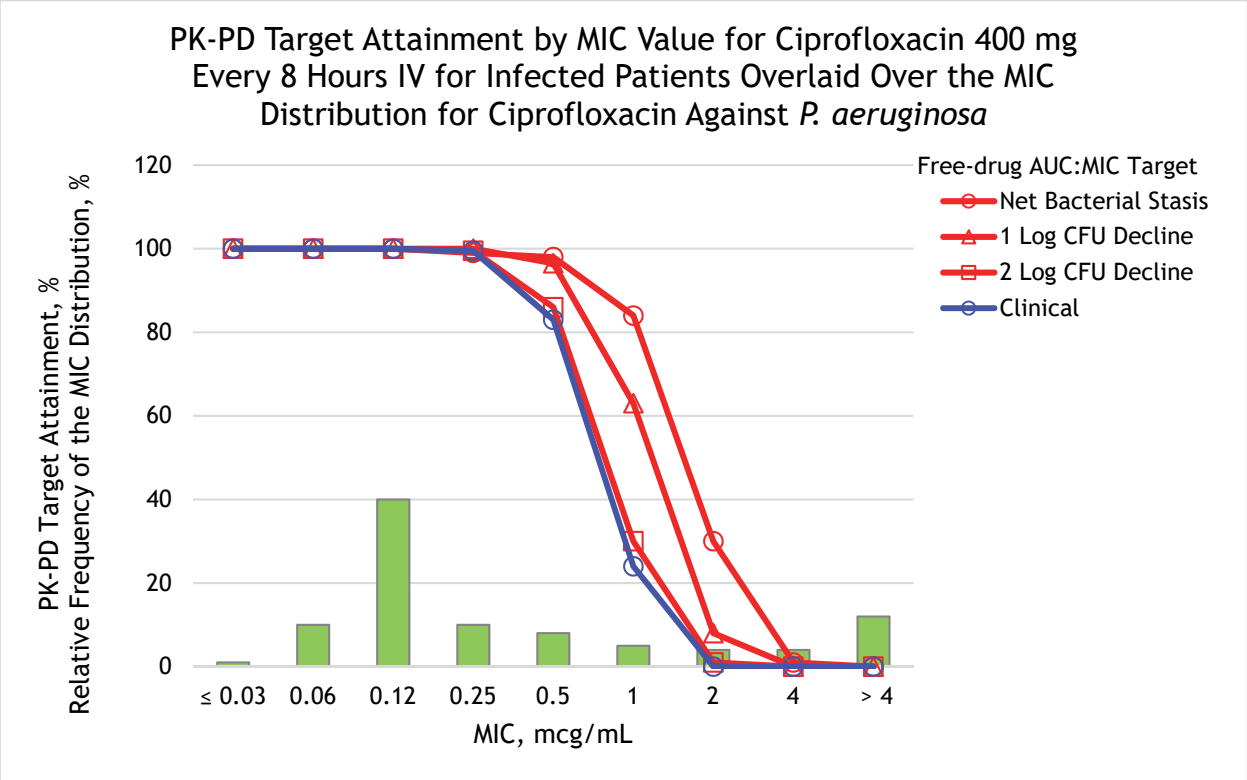
Abbreviations: AUC, area under the curve; CFU, colony-forming unit; IV, intravenous; MIC, minimal inhibitory concentration; PK-PD, pharmacokinetic-pharmacodynamic; PO, oral.

PK-PD Target Attainment by MIC Value for Ciprofloxacin 500 mg Every 12 Hours PO for Healthy Subjects With Inflated Variance Overlaid Over the MIC Distribution for Ciprofloxacin Against *P. aeruginosa*



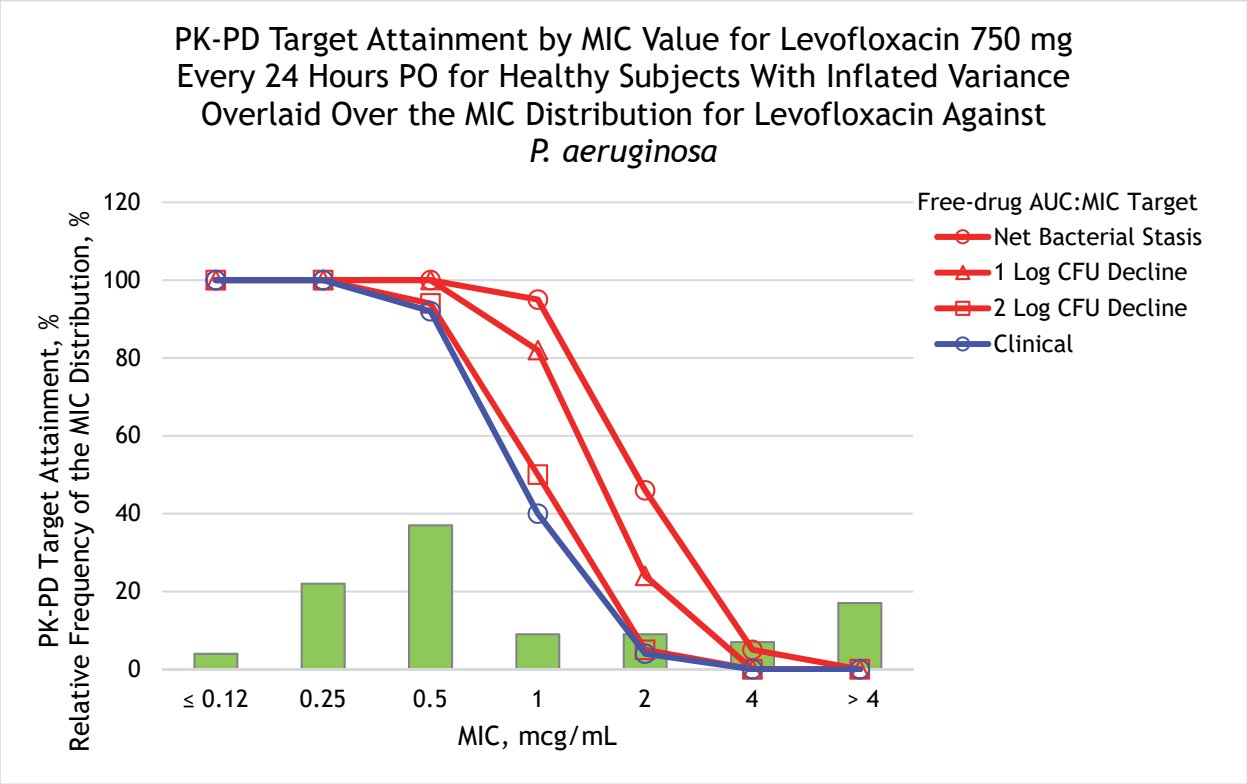
PK-PD Target Attainment by MIC Value for Ciprofloxacin 750 mg Every 12 Hours PO for Healthy Subjects With Inflated Variance Overlaid Over the MIC Distribution for Ciprofloxacin Against *P. aeruginosa*

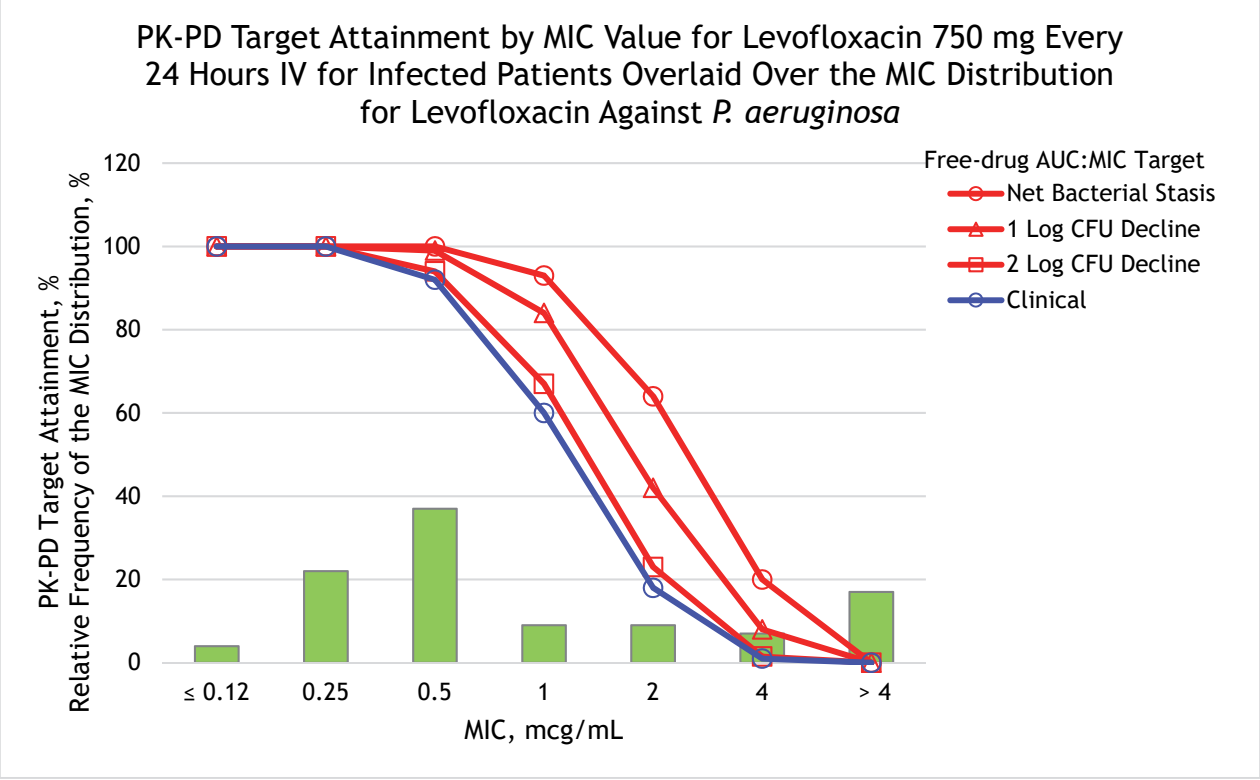




Abbreviations: AUC, area under the curve; CFU, colony-forming unit; IV, intravenous; MIC, minimal inhibitory concentration; PK-PD, pharmacokinetic-pharmacodynamic; PO, oral.

Figure 9. Percent Probabilities of Ciprofloxacin PK-PD Target Attainment Based on Free-Drug AUC:MIC Ratio Targets Relative to the MIC Distribution for *P. aeruginosa*[10] (Reprinted with permission from USCAST, The National Antimicrobial Susceptibility Testing Committee for the United States. Quinolone In Vitro Susceptibility Test Interpretive Criteria Evaluations. Version 1.3, 2018. http://www.uscast.org.)





Abbreviations: AUC, area under the curve; CFU, colony-forming unit; IV, intravenous; MIC, minimal inhibitory concentration; PK-PD, pharmacokinetic-pharmacodynamic; PO, oral.

Figure 10. Percent Probabilities of Levofloxacin PK-PD Target Attainment Based on Free-Drug AUC:MIC Ratio Targets Relative to the MIC Distribution for *P. aeruginosa*[10] (Reprinted with permission from USCAST, The National Antimicrobial Susceptibility Testing Committee for the United States. Quinolone In Vitro Susceptibility Test Interpretive Criteria Evaluations. Version 1.3, 2018. http://www.uscast.org.)

6 Clinical Efficacy

Limited clinical data from the ceftolozane-tazobactam/levofloxacin clinical trial showed both microbiological and clinical cures with levofloxacin (750 mg once a day) at 1 and 2 µg/mL.[12] No data were available for review for ciprofloxacin clinical efficacy.

7 Committee Rationale for the Breakpoint

For *Enterobacteriaceae*, more sophisticated PK-PD analyses provided the basis for lower breakpoints for ciprofloxacin and levofloxacin. The EUCAST 2017 breakpoints were defined by a fourfold lower MIC than the CLSI breakpoints. Among the *Enterobacteriaceae*, approximately 4% of CLSI-susceptible isolates would be considered resistant using the EUCAST breakpoints.

For *P. aeruginosa*, CLSI and EUCAST 2017 breakpoints for fluoroquinolones only differ by one dilution but encompass up to 10% of the population. A review of PK-PD targets showed that the clinical targets line up well with the animal targets for 1 or 2 log reductions. Monte Carlo simulations supported the revised breakpoints.

8 Final Table Entry

Tables 12 and 13 include the final table entries from CLSI document M100.[2]

Table 12. Excerpt From CLSI Document M100[2] Table 2A, Zone Diameter and MIC Breakpoints for *Enterobacteriaceae*

Test/Report Group	Antimicrobial Agent	Interpretive Categories and MIC breakpoints, µg/mL				Comments
		S	SDD	I	R	
B	Ciprofloxacin	≤0.25	–	0.5	≥1	**(39)** Breakpoints for ciprofloxacin are based on a dosage regimen of 400 mg IV or 500 mg orally administered every 12 h.
B	Levofloxacin	≤0.5	–	1	≥2	**(40)** Breakpoints for levofloxacin are based on a dosage regimen of 750 mg administered every 24 h.

Abbreviations: h, hours; I, intermediate; IV, intravenous; MIC, minimal inhibitory concentration; R, resistant; S, susceptible; SDD, susceptible-dose dependent.

Table 13. Excerpt From CLSI Document M100[2] Table 2B-1, Zone Diameter and MIC Breakpoints for *Pseudomonas aeruginosa*

Test/Report Group	Antimicrobial Agent	Interpretive Categories and MIC breakpoints, µg/mL			Comments
		S	I	R	
B	Ciprofloxacin	≤0.5	1	≥2	**(21)** Breakpoints are based on a dosage regimen of 400 mg IV administered every 8 h.
B	Levofloxacin	≤1	2	≥4	**(22)** Breakpoints are based on a dosage regimen of 750 mg administered every 24 h.

Abbreviations: h, hours; I, intermediate; IV, intravenous; MIC, minimal inhibitory concentration; R, resistant; S, susceptible.

9 Voting Record

In January 2017, a motion to accept the *Enterobacteriaceae* breakpoints for ciprofloxacin and levofloxacin, pending approval of the disk diffusion breakpoints in June 2017, including an intermediate category and dosing information, was made and seconded (9 approved, 3 opposed, 0 abstained). **NOTE:** The dissenting votes were related to the lack of evidence that treatment based on the 2017 CLSI breakpoints was not working clinically.

In January 2018, a motion to lower the ciprofloxacin breakpoints for *P. aeruginosa* to ≤ 0.5 µg/L (S), 1 µg/L (I), ≥ 2 µg/L (R) and the levofloxacin breakpoints to ≤ 1 µg/L (S), 2 µg/L (I), ≥ 4 µg/L (R) was made and seconded (11 approved, 0 opposed, 1 abstained, plus 1 was absent).

10 References

1 CLSI. *Development of* In Vitro *Susceptibility Testing Criteria and Quality Control Parameters*. 5th ed. CLSI guideline M23. Wayne, PA: Clinical and Laboratory Standards Institute; 2018.

2 CLSI. *Performance Standards for Antimicrobial Susceptibility Testing*. 29th ed. CLSI supplement M100. Wayne, PA: Clinical and Laboratory Standards Institute; 2019.

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12 Wagenlehner FM, Umeh O, Steenbergen J, Yuan G, Darouiche RO. Ceftolozane-tazobactam compared with levofloxacin in the treatment of complicated urinary-tract infections, including pyelonephritis: a randomized, double-blind, phase 3 trial (ASPECT-cUTI). *Lancet*. 2015;385(9981):1949-1956.

The findings, recommendations, and conclusions in this rationale document are those of the authors and have not been reviewed through the CLSI consensus process. They do not necessarily reflect the views of any single individual or organization.

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