Aminoglycoside Breakpoints for Enterobacterales 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 QC ranges.

The details of the necessary and recommended data for selecting appropriate breakpoints and QC ranges, as well as how the data are presented for evaluation, are described in CLSI M23.[1] CLSI antibacterial breakpoints are provided in CLSI M100[2] and CLSI 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 Aminoglycoside Ad Hoc Working Group.

2 Introduction

Gentamicin, tobramycin, and amikacin are members of the aminoglycoside group of antimicrobial agents. The aminoglycosides demonstrate bactericidal activity through inhibition of protein synthesis, binding with high affinity to the A-site on the 16S ribosomal RNA of the 30S ribosome. The first aminoglycoside, streptomycin, was isolated from *Streptomyces griseus* and introduced into clinical use in 1944. Gentamicin and tobramycin are natural antibiotics derived from *Micromonospora purpurea* and *Streptoalloteichus tenebrarius*, respectively, whereas amikacin is derived from kanamycin.[4] Aminoglycosides have *in vitro* activity against gram-negative and gram-positive bacteria, including the Enterobacterales, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, many *Mycobacterium* spp., and to a lesser extent, *Acinetobacter baumannii*. Active electron transport is required for aminoglycoside uptake into cells, resulting in intrinsic resistance among anaerobic bacteria. The aminoglycosides also lack activity against *Burkholderia* spp., *Stenotrophomonas maltophilia*, *Streptococcus* spp., and *Enterococcus* spp. (excluding as part of synergy). Aminoglycoside resistance occurs through several pathways, including:

- Enzymatic modification
- Target site modification (16S methylation)
- Efflux

Enzymatic modification of the aminoglycosides occurs through aminoglycoside acetyltransferases, which compose the largest group of aminoglycoside-modifying enzymes, as well as through aminoglycoside phosphotransferases and aminoglycoside nucleotransferases.

US Food and Drug Administration (FDA)-approved indications for aminoglycoside use are broad. Gentamicin is approved for the treatment of serious infections caused by susceptible isolates of *P. aeruginosa*, *Proteus* spp., *Escherichia coli*, *Klebsiella* spp., *Enterobacter* spp., *Serratia* spp., *Citrobacter* spp., and *Staphylococcus* spp., including bacterial neonatal sepsis; bacterial septicemia; and infections of the nervous system (meningitis), urinary tract, respiratory tract, gastrointestinal tract (including peritonitis), and skin, bone, and soft tissue (including burns).

Tobramycin is approved by the FDA for the treatment of septicemia caused by *P. aeruginosa*, *E. coli*, and *Klebsiella* spp.; lower respiratory tract infections caused by *P. aeruginosa*, *Klebsiella* spp., *Enterobacter* spp., *Serratia* spp., *E. coli*, and *S. aureus*; serious central nervous system infections caused by susceptible organisms; intraabdominal infections, including peritonitis caused by *E. coli*, *Klebsiella* spp., and *Enterobacter* spp.; skin, bone, and skin structure infections caused by *P. aeruginosa*, *Proteus* spp., *E. coli*, *Klebsiella* spp., *Enterobacter* spp., and *S. aureus*; and complicated urinary tract infections caused by *P. aeruginosa*, *Proteus* spp., *E. coli*, *Klebsiella* spp., *Enterobacter* spp., *Serratia* spp., *S. aureus*, *Providencia* spp., and *Citrobacter* spp.

Finally, amikacin is FDA approved for the treatment of serious infections caused by susceptible strains of gram-negative bacteria, including *Pseudomonas* spp., *E. coli*, *Proteus* spp., *Providencia* spp., *Klebsiella* spp., *Enterobacter* spp., *Serratia* spp., and *Acinetobacter* spp. Clinical studies have also shown amikacin to be effective in serious, complicated, and recurrent urinary tract infections caused by these organisms. It is also effective in bacterial septicemia (including neonatal sepsis); serious infections of the respiratory tract, bones and joints, central nervous system (including meningitis), and skin and soft tissue; intraabdominal infections (including peritonitis); burns; and postoperative infections (including postvascular surgery).

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

Table 1. Current CLSI Aminoglycoside MIC Breakpoints[a]

Organism Group	Antimicrobial Agent	Interpretive Categories and MIC Breakpoints, µg/mL			
		S	SDD	I	R
Enterobacterales	Gentamicin	≤ 2	–	4^	≥ 8
	Tobramycin	≤ 2	–	4^	≥ 8
	Amikacin	≤ 4	-	8^	≥ 16
P. aeruginosa	Gentamicin	-	-	-	-
	Tobramycin	≤ 1	-	2^	≥ 4
	Amikacin (U)[b]	≤ 16	-	32^	≥ 64

Abbreviations: I, intermediate; MIC, minimal inhibitory concentration; R, resistant; S, susceptible; SDD, susceptible-dose dependent; U, urine.
Symbol: ^, designation for agents that have the potential to concentrate in the urine.
[a] Last reviewed June 2022; first published in CLSI M100-Ed33.[2]
[b] Report only on organisms isolated from the urinary tract.

Table 2. Historical CLSI Aminoglycoside MIC Breakpoints Replaced by Current Aminoglycoside Breakpoints[a]

Organism Group	Antimicrobial Agent	Interpretive Categories and MIC Breakpoints, µg/mL			
		S	SDD	I	R
Enterobacterales	Gentamicin	≤ 4	–	8^	≥ 16
	Tobramycin	≤ 4	–	8^	≥ 16
	Amikacin	≤ 16	-	32^	≥ 64
P. aeruginosa	Gentamicin	≤ 4	–	8^	≥ 16
	Tobramycin	≤ 4	–	8^	≥ 16
	Amikacin	≤ 16	-	32^	≥ 64

Abbreviations: I, intermediate; MIC, minimal inhibitory concentration; R, resistant; S, susceptible; SDD, susceptible-dose dependent.
Symbol: ^, designation for agents that have the potential to concentrate in the urine.
[a] First published in NCCLS document M2-A2-S2 in 1979.

3 Standard Dosages and Pharmacokinetic Data

Extended-interval aminoglycoside doses used to establish breakpoints are shown in Table 3. Clinical studies have reported a lower incidence of nephrotoxicity with a once-daily aminoglycoside dosage regimen, even though some doses are higher than those described in prescribing information.[5-7] Because of its advantages, a once-daily dosage regimen is widely accepted and has become the standard of care.[8] Routine practice is to administer gentamicin and tobramycin as a 5-to-7-mg/kg once-daily intravenous infusion and amikacin as a 15-to-20-mg/kg once-daily intravenous infusion. International treatment guidelines recommend an extended-interval dosage regimen for gentamicin,[9-17] tobramycin,[10,12,14-16,18] and amikacin.[10-12,14,16]

Table 3. Dosage Regimen Used for Breakpoint Determination[a]

Antimicrobial Agent	Dosage Regimen
Gentamicin	7 mg/kg IV every 24 h
Tobramycin	7 mg/kg IV every 24 h
Amikacin	15 mg/kg IV every 24 h

Abbreviations: h, hour(s); IV, intravenous.
[a] As published in CLSI M100-Ed33.[2]