CLINICAL AND LABORATORY STANDARDS INSTITUTE®

31st Edition

M100

Performance Standards for Antimicrobial Susceptibility Testing

This document includes updated tables for the Clinical and Laboratory Standards Institute antimicrobial susceptibility testing standards M02, M07, and M11.

A CLSI supplement for global application.

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Performance Standards for Antimicrobial Susceptibility Testing

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Abstract

The data in the tables are valid only if the methodologies in CLSI documents M02,[1] M07,[2] and M11[3] are followed. These standards contain information about disk diffusion (M02[1]) and dilution (M07[2] and M11[3]) test procedures for aerobic and anaerobic bacteria. Clinicians depend heavily on information from the microbiology laboratory for treating their seriously ill patients. The clinical importance of antimicrobial susceptibility test results demands that these tests be performed under optimal conditions and that laboratories have the capability to provide results for the newest antimicrobial agents. The tables presented in M100 represent the most current information for drug selection, interpretation, and quality control using the procedures standardized in M02,[1] M07,[2] and M11.[3] Users should replace previously published tables with these new tables. Changes in the tables since the previous edition appear in boldface type.

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Summary of CLSI Processes for Establishing Breakpoints and Quality Control Ranges

The Clinical and Laboratory Standards Institute (CLSI) is an international, voluntary, not-for-profit, interdisciplinary, standards-developing, and educational organization accredited by the American National Standards Institute that develops and promotes the use of consensus-developed standards and guidelines within the health care community. These consensus standards and guidelines are developed in an open and consensus-seeking forum to cover critical areas of diagnostic testing and patient health care. CLSI is open to anyone or any organization that has an interest in diagnostic testing and patient care. Information about CLSI can be found at www.clsi.org.

The CLSI Subcommittee on Antimicrobial Susceptibility Testing reviews data from a variety of sources and studies (eg, *in vitro*, pharmacokinetics-pharmacodynamics, and clinical studies) to establish antimicrobial susceptibility test methods, breakpoints, and QC parameters. The details of the data necessary to establish breakpoints, QC parameters, and how the data are presented for evaluation are described in CLSI document M23.[4]

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 and QC parameters may be refined to ensure more accurate and better performance of susceptibility test methods. 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 be used in conjunction with clinical judgment, current knowledge, and clinically relevant laboratory test results to guide patient treatment.

Additional information, updates, and changes in this document are found in the meeting summary minutes of the Subcommittee on Antimicrobial Susceptibility Testing at https://clsi.org/meetings/ast-file-resources/.

CLSI Reference Methods vs Commercial Methods and CLSI vs US Food and Drug Administration Breakpoints

It is important for users of M02,[1] M07,[2] and M100 to recognize that the standard methods described in CLSI documents are reference methods. These methods may be used for routine antimicrobial susceptibility testing of patient isolates, for evaluating commercial devices that will be used in medical laboratories, or by drug or device manufacturers for testing new agents or systems. Results generated by reference methods, such as those included in CLSI documents, may be used by regulatory authorities to evaluate the performance of commercial susceptibility testing devices as part of the approval process. Clearance by a regulatory authority indicates the commercial susceptibility testing device provides susceptibility results that are substantially equivalent to results generated using reference methods for the organisms and antimicrobial agents described in the device manufacturer's approved package insert.

CLSI breakpoints may differ from those approved by various regulatory authorities for many reasons, including use of different databases, differences in data interpretation, differences in doses used in different parts of the world, and public health policies. Differences also exist because CLSI proactively evaluates the need for changing breakpoints. The reasons why breakpoints may change and the manner in which CLSI evaluates data and determines breakpoints are outlined in CLSI document M23.[4]

Following a decision by CLSI to change an existing breakpoint, regulatory authorities may also review data to determine how changing breakpoints may affect the safety and effectiveness of the antimicrobial agent for the approved indications. If the regulatory authority changes breakpoints, commercial device manufacturers may have to conduct a clinical trial, submit the data to the regulatory authority, and await review and approval. For these reasons, a delay of one or more years may be needed if a breakpoint and interpretive category change is to be implemented by a device manufacturer. In the United States, it is acceptable for laboratories that use US Food and Drug Administration (FDA)–cleared susceptibility testing devices to use existing FDA breakpoints. Either FDA or CLSI susceptibility breakpoints are acceptable to laboratory accrediting organizations in the United States. Policies in other countries may vary. Each laboratory should check with the manufacturer of its antimicrobial susceptibility test system for additional information on the breakpoints and interpretive categories used in its system's software.

Following discussions with appropriate stakeholders (eg, infectious diseases and pharmacy practitioners, the pharmacy and therapeutics and infection prevention committees of the medical staff, and the antimicrobial stewardship team), newly approved or revised breakpoints may be implemented by laboratories. Following verification, CLSI disk diffusion test breakpoints may be implemented as soon as they are published in M100. If a device includes antimicrobial test concentrations sufficient to allow interpretation of susceptibility and resistance to an agent using the CLSI breakpoints, a laboratory could choose to, after appropriate verification, interpret and report results using CLSI breakpoints.

Instructions for Use of Tables

These instructions apply to:

- **Tables 1A and 1B:** suggested groupings of antimicrobial agents that should be considered for testing and reporting by microbiology laboratories. These guidelines are based on antimicrobial agents approved by the US Food and Drug Administration (FDA) for clinical use in the United States. In other countries, placement of antimicrobial agents in Tables 1A and 1B should be based on available drugs approved for clinical use by relevant regulatory organizations.

- **Tables 2A through 2I:** tables for each organism group that contain:
  - Recommended testing conditions
  - Routine QC recommendations (also see Chapter 4 in M02[1] and M07[2])
  - General comments for testing the organism group and specific comments for testing particular agent/organism combinations
  - Suggested agents that should be considered for routine testing and reporting by medical microbiology laboratories, as specified in Tables 1A and 1B (test/report groups A, B, C, U)
  - Additional drugs that are appropriate for the respective organism group but would generally not warrant routine testing by a medical microbiology laboratory in the United States (test/report group O for "other"; test/report group Inv. for "investigational" [not yet FDA approved])
  - Zone diameter and minimal inhibitory concentration (MIC) breakpoints

- **Tables 1C and 2J:** tables containing specific recommendations for testing and reporting results on anaerobes and some of the information listed in the bullets above

- **Tables 3A to 3K:** tables describing tests to detect particular resistance types in specific organisms or organism groups

I. Selecting Antimicrobial Agents for Testing and Reporting

A. Appropriate Agents for Routine Testing

Selecting the most appropriate antimicrobial agents to test and report is a decision best made by each laboratory in consultation with the infectious diseases and pharmacy practitioners, the pharmacy and therapeutics and infection prevention committees of the medical staff, and the antimicrobial stewardship team. The recommendations for each organism group include agents of proven efficacy that show acceptable *in vitro* test performance. Considerations in the assignment of agents to specific test/report groups include clinical efficacy, prevalence of resistance, minimizing emergence of resistance, cost, FDA clinical indications for use, and current consensus recommendations for first-choice and alternative drugs. Tests on selected agents may be useful for infection prevention purposes.

B. Equivalent Agents

Antimicrobial agents listed together in a single box are agents for which interpretive categories (susceptible, intermediate, susceptible-dose dependent, or resistant) and clinical efficacy are similar. Within each box, an "or" between agents indicates agents for which cross-resistance and cross-susceptibility are nearly complete. Results from one agent connected by an "or" can be used to predict results for the other agent (ie, equivalent agents). For example, Enterobacterales susceptible to cefotaxime can be considered susceptible to ceftriaxone. The results obtained from testing cefotaxime could be reported along with a comment that the isolate is also susceptible to ceftriaxone. For drugs connected with an "or," combined major and very major errors are fewer than 3%, and minor errors are fewer than 10%, based on a large population of bacteria tested (see CLSI document M23[4] for description of error types). In addition, to qualify for an "or," at least 100 strains with resistance to the agents in question must be tested, and a result of "resistant" must be obtained with all agents for at least 95% of the strains. "Or" is also used for comparable agents when tested against organisms for which "susceptible-only" breakpoints are provided (eg, cefotaxime or ceftriaxone with *H. influenzae*). When no "or" connects agents within a box, testing of one agent cannot be used to predict results for another, owing either to discrepancies or insufficient data.

C. Test/Report Groups

1. **Group A antimicrobial agents,** as listed in Tables 1A, 1B, and 1C, are considered appropriate for inclusion in a routine, primary testing panel, as well as for routine reporting of results for the specific organism groups.

2. **Group B** includes antimicrobial agents that may warrant primary testing, but they may be reported only selectively, such as when the organism is resistant to agents of the same antimicrobial class, as in group A. Other indications for reporting the result might include a selected specimen source (eg, a third-generation cephalosporin for enteric bacilli from CSF or

Table 1A. (Continued)

Acinetobacter spp.	*Burkholderia cepacia* complex	*Stenotrophomonas maltophilia*	Other Non-Enterobacterales[i,w]
Group A: Includes antimicrobial agents considered appropriate for inclusion in a routine, primary testing panel, as well as for routine reporting of results for the specific organism group.			
Ampicillin-sulbactam	Levofloxacin[i]	Levofloxacin	Ceftazidime
Ceftazidime	Meropenem	Minocycline	Gentamicin
Ciprofloxacin Levofloxacin	Trimethoprim-sulfamethoxazole	Trimethoprim-sulfamethoxazole	Tobramycin
Doripenem Imipenem Meropenem			
Gentamicin Tobramycin			
Group B: Includes antimicrobial agents that may warrant primary testing but may be reported only selectively, such as when the organism is resistant to agents of the same antimicrobial class in Group A.[l]			
Amikacin	Ceftazidime	Ceftazidime[i]	Amikacin
Piperacillin-tazobactam	Minocycline		Aztreonam
Cefepime			Cefepime
Cefotaxime Ceftriaxone			Ciprofloxacin Levofloxacin
Doxycycline			Imipenem Meropenem
Minocycline			Piperacillin-tazobactam
Trimethoprim-sulfamethoxazole			Trimethoprim-sulfamethoxazole
Group C: Includes alternative or supplemental antimicrobial agents that may require testing in institutions that harbor endemic or epidemic strains resistant to several of the primary drugs, for treatment of patients allergic to primary drugs, for treatment of unusual organisms, or for reporting to infection prevention as an epidemiological aid.			
	Chloramphenicol[c,i]	Chloramphenicol[c,i]	Cefotaxime Ceftriaxone
			Chloramphenicol[c]
Group U: Includes antimicrobial agents that are used only or primarily for treating UTIs.			
Tetracycline[q]			Sulfisoxazole
			Tetracycline[q]

Abbreviations: CSF, cerebrospinal fluid; MIC, minimal inhibitory concentration; UTI, urinary tract infection.

Table 2A. Enterobacterales (Continued)

Test/Report Group	Antimicrobial Agent	Disk Content	Interpretive Categories and Zone Diameter Breakpoints, nearest whole mm				Interpretive Categories and MIC Breakpoints, µg/mL				Comments
			S	SDD	I	R	S	SDD	I	R	
CARBAPENEMS (Continued)											
B	Imipenem	10 µg	≥ 23	-	20-22^	≤ 19	≤ 1	-	2^	≥ 4	**(38)** Breakpoints are based on a dosage regimen of 500 mg administered every 6 h or 1 g every 8 h. **See comment (13).**
B	Meropenem	10 µg	≥ 23	-	20-22^	≤ 19	≤ 1	-	2^	≥ 4	**(39)** Breakpoints are based on a dosage regimen of 1 g administered every 8 h.
LIPOPEPTIDES											
(40) WARNING: Clinical and PK/PD data demonstrate colistin and polymyxin B have limited clinical efficacy, even if an intermediate result is obtained. Alternative agents are strongly preferred. Colistin and polymyxin B should be used in combination with one or more active antimicrobial agents. Consultation with an infectious diseases specialist is recommended. **(41)** Several species are intrinsically resistant to the lipopeptides (colistin and polymyxin B). Refer to Appendix B.											
O	Colistin or polymyxin B		- -	- -	- -	- -	- -	- -	≤ 2 ≤ 2	≥ 4 ≥ 4	**(42)** Colistin (methanesulfonate) should be given with a loading dose and maximum renally adjusted doses (see International Consensus Guidelines[8]). **(43)** Polymyxin B should be given with a loading dose and maximum recommended doses (see International Consensus Guidelines[8]). **(44)** When colistin or polymyxin B is given systemically, neither is likely to be effective for pneumonia. **(45)** For colistin, broth microdilution, CBDE, and CAT MIC methods are acceptable. For polymyxin B, broth microdilution is the only approved method. Disk diffusion and gradient diffusion methods should not be performed (see Table 3D).

Related CLSI Reference Materials[a]

EP23™ **Laboratory Quality Control Based on Risk Management. 1st ed., 2011.** This document provides guidance based on risk management for laboratories to develop quality control plans tailored to the particular combination of measuring system, laboratory setting, and clinical application of the test.

M02 **Performance Standards for Antimicrobial Disk Susceptibility Tests. 13th ed., 2018.** This standard covers the current recommended methods for disk susceptibility testing and criteria for quality control testing.

M02QG **M02 Disk Diffusion Reading Guide. 1st ed., 2018.** The Disk Diffusion Reading Guide provides photographic examples of the proper method for reading disk diffusion susceptibility testing results.

M07 **Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically. 11th ed., 2018.** This standard covers reference methods for determining minimal inhibitory concentrations of aerobic bacteria by broth macrodilution, broth microdilution, and agar dilution.

M11 **Methods for Antimicrobial Susceptibility Testing of Anaerobic Bacteria. 9th ed., 2018.** This standard provides reference methods for determining minimal inhibitory concentrations of anaerobic bacteria by agar dilution and broth microdilution.

M23 **Development of *In Vitro* Susceptibility Testing Criteria and Quality Control Parameters. 5th ed., 2018.** This guideline discusses the necessary and recommended data for selecting appropriate breakpoints and quality control ranges for antimicrobial agents.

M39 **Analysis and Presentation of Cumulative Antimicrobial Susceptibility Test Data. 4th ed., 2014.** This document describes methods for recording and analysis of antimicrobial susceptibility test data, consisting of cumulative and ongoing summaries of susceptibility patterns of clinically significant microorganisms.

M45 **Methods for Antimicrobial Dilution and Disk Susceptibility Testing of Infrequently Isolated or Fastidious Bacteria. 3rd ed., 2016.** This guideline informs clinical, public health, and research laboratories on susceptibility testing of infrequently isolated or fastidious bacteria that are not included in CLSI documents M02, M07, or M100. Antimicrobial agent selection, test interpretation, and quality control are addressed.

M52 **Verification of Commercial Microbial Identification and Antimicrobial Susceptibility Testing Systems. 1st ed., 2015.** This guideline includes recommendations for verification of commercial US Food and Drug Administration–cleared microbial identification and antimicrobial susceptibility testing systems by clinical laboratory professionals to fulfill regulatory or quality assurance requirements for the use of these systems for diagnostic testing.

[a] CLSI documents are continually reviewed and revised through the CLSI consensus process; therefore, readers should refer to the most current editions.

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