M37-A3
Vol. 28 No. 7
Replaces M37-A2
Vol. 22 No. 7

Development of *In Vitro* Susceptibility Testing Criteria and Quality Control Parameters for Veterinary Antimicrobial Agents; Approved Guideline—Third Edition

This document addresses the required and recommended data needed for selection of appropriate interpretive standards and quality control guidance for new veterinary antimicrobial agents.

A guideline for global application developed through the Clinical and Laboratory Standards Institute consensus process.

CLINICAL AND LABORATORY STANDARDS INSTITUTE®
(Formerly NCCLS)

Clinical and Laboratory Standards Institute

Advancing Quality in Health Care Testing

The Clinical and Laboratory Standards Institute (CLSI, formerly NCCLS) is an international, interdisciplinary, nonprofit, standards-developing, and educational organization that promotes the development and use of voluntary consensus standards and guidelines within the health care community. It is recognized worldwide for the application of its unique consensus process in the development of standards and guidelines for patient testing and related health care issues. Our process is based on the principle that consensus is an effective and cost-effective way to improve patient testing and health care services.

In addition to developing and promoting the use of voluntary consensus standards and guidelines, we provide an open and unbiased forum to address critical issues affecting the quality of patient testing and health care.

PUBLICATIONS

A document is published as a standard, guideline, or committee report.

Standard A document developed through the consensus process that clearly identifies specific, essential requirements for materials, methods, or practices for use in an unmodified form. A standard may, in addition, contain discretionary elements, which are clearly identified.

Guideline A document developed through the consensus process describing criteria for a general operating practice, procedure, or material for voluntary use. A guideline may be used as written or modified by the user to fit specific needs.

Report A document that has not been subjected to consensus review and is released by the Board of Directors.

CONSENSUS PROCESS

The CLSI voluntary consensus process is a protocol establishing formal criteria for:

- the authorization of a project
- the development and open review of documents
- the revision of documents in response to comments by users
- the acceptance of a document as a consensus standard or guideline.

Most documents are subject to two levels of consensus—"proposed" and "approved." Depending on the need for field evaluation or data collection, documents may also be made available for review at an intermediate consensus level.

Proposed A consensus document undergoes the first stage of review by the health care community as a proposed standard or guideline. The document should receive a wide and thorough technical review, including an overall review of its scope, approach, and utility, and a line-by-line review of its technical and editorial content.

Approved An approved standard or guideline has achieved consensus within the health care community. It should be reviewed to assess the utility of the final document, to ensure attainment of consensus (ie, that comments on earlier versions have been satisfactorily addressed), and to identify the need for additional consensus documents.

Our standards and guidelines represent a consensus opinion on good practices and reflect the substantial agreement by materially affected, competent, and interested parties obtained by following CLSI's established consensus procedures. Provisions in CLSI standards and guidelines may be more or less stringent than applicable regulations. Consequently, conformance to this voluntary consensus document does not relieve the user of responsibility for compliance with applicable regulations.

COMMENTS

The comments of users are essential to the consensus process. Anyone may submit a comment, and all comments are addressed, according to the consensus process, by the committee that wrote the document. All comments, including those that result in a change to the document when published at the next consensus level and those that do not result in a change, are responded to by the committee in an appendix to the document. Readers are strongly encouraged to comment in any form and at any time on any document. Address comments to the Clinical and Laboratory Standards Institute, 940 West Valley Road, Suite 1400, Wayne, PA 19087, USA.

VOLUNTEER PARTICIPATION

Health care professionals in all specialties are urged to volunteer for participation in CLSI projects. Please contact us at customerservice@clsi.org or +610.688.0100 for additional information on committee participation.

M37-A3
ISBN 1-56238-660-3
Volume 28 Number 7 ISSN 0273-3099

Development of *In Vitro* Susceptibility Testing Criteria and Quality Control Parameters for Veterinary Antimicrobial Agents; Approved Guideline—Third Edition

Jeffrey L. Watts, PhD, RM(AAM)
Michael Apley, DVM, PhD
Donald J. Bade
Steven D. Brown, PhD
Jeffrey T. Gray, PhD
Henry Heine, PhD
Rob P. Hunter, MS, PhD
Marilyn N. Martinez, PhD
Dik J. Mevius, DVM, PhD
Mark G. Papich, DVM, MS
Thomas R. Shryock, PhD
Peter Silley, PhD
Robert D. Walker, PhD
Ching Ching Wu, DVM, PhD
Gary E. Zurenko, MS

Abstract

CLSI document M37-A3—*Development of* In Vitro *Susceptibility Testing Criteria and Quality Control Parameters for Veterinary Antimicrobial Agents; Approved Guideline—Third Edition* offers guidance for developing agar disk diffusion zones of inhibition, dilution MIC breakpoints, and quality control limits for antimicrobial susceptibility testing of aerobic bacteria isolated from animals. It is intended to be used in establishing interpretive and quality control criteria for CLSI antimicrobial susceptibility testing standards for antimicrobial agents intended for veterinary use. Host-specific pharmacokinetics, *in vitro* drug characteristics, distributions of microorganisms, and correlation of test results with outcome statistics are addressed from the perspective of interpretation of test results. In addition, this document addresses clinical confirmation of interpretive criteria and quality control limits. For clinical confirmation, the "ideal" data set may not be obtained during development of a new drug. Users of this guideline should understand the limitations and work toward the best-educated conclusions.

Clinical and Laboratory Standards Institute (CLSI). *Development of* In Vitro *Susceptibility Testing Criteria and Quality Control Parameters for Veterinary Antimicrobial Agents; Approved Guideline—Third Edition*. CLSI document M37-A3 (ISBN 1-56238-660-3). Clinical and Laboratory Standards Institute, 940 West Valley Road, Suite 1400, Wayne, Pennsylvania 19087-1898 USA, 2008.

Suggested Citation

(CLSI. *Development of* In Vitro *Susceptibility Testing Criteria and Quality Control Parameters for Veterinary Antimicrobial Agents; Approved Guideline—Third Edition.* CLSI document M37-A3. Wayne, PA: Clinical and Laboratory Standards Institute; 2008.)

Proposed Guideline
December 1996

Approved Guideline
February 1999

Approved Guideline—Second Edition
May 2002

Approved Guideline—Third Edition
February 2008

ISBN 1-56238-660-3
ISSN 0273-3099

Committee Membership

Area Committee on Microbiology

Mary Jane Ferraro, PhD, MPH
Chairholder
Massachusetts General Hospital
Boston, Massachusetts

James H. Jorgensen, PhD
Vice-Chairholder
University of Texas Health Science Center
San Antonio, Texas

Donald R. Callihan, PhD
BD Diagnostic Systems
Sparks, Maryland

Freddie Mae Poole
FDA Center for Devices and Radiological Health
Rockville, Maryland

John H. Rex, MD, FACP
AstraZeneca
Cheshire, United Kingdom

Daniel F. Sahm, PhD
Eurofins Medinet
Herndon, Virginia

Fred C. Tenover, PhD, ABMM
Centers for Disease Control and Prevention
Atlanta, Georgia

John D. Turnidge, MD
Women's and Children's Hospital
North Adelaide, Australia

Michael L. Wilson, MD
Denver Health Medical Center
Denver, Colorado

Advisors

Ellen Jo Baron, PhD
Stanford University Hospital & Medical School
Stanford, California

Lynne S. Garcia, MS
LSG and Associates
Santa Monica, California

Richard L. Hodinka, PhD
Children's Hospital of Philadelphia
Philadelphia, Pennsylvania

Michael A. Pfaller, MD
University of Iowa College of Medicine
Iowa City, Iowa

Robert P. Rennie, PhD
University of Alberta Hospitals
Edmonton, Alberta, Canada

Jana M. Swenson, MMSc
Centers for Disease Control and Prevention
Atlanta, Georgia

Melvin P. Weinstein, MD
Robert Wood Johnson Medical School
New Brunswick, New Jersey

Matthew A. Wikler, MD, MBA, FIDSA
Pacific Beach BioSciences
San Diego, California

Gail L. Woods, MD
University of Arkansas for Medical Sciences
Little Rock, Arkansas

Subcommittee on Veterinary Antimicrobial Susceptibility Testing

Jeffrey L. Watts, PhD, RM(AAM)
Chairholder
Pfizer Animal Health
Richmond, Michigan

Mike Apley, DVM, PhD
Kansas State University
Manhattan, Kansas

Donald J. Bade
Microbial Research, Inc.
Fort Collins, Colorado

Steven D. Brown, PhD
The Clinical Microbiology Institute
Wilsonville, Oregon

Jeffrey T. Gray, PhD
Des Moines University
Des Moines, Iowa

Henry Heine, PhD
USAMRIID
Ft. Detrick, Maryland

Rob P. Hunter, MS, PhD
Elanco Animal Health
Greenfield, Indiana

Dik J. Mevius, DVM, PhD
Central Institute for Animal Disease Control-CIDC-Lelystad
Lelystad, Netherlands

Mark G. Papich, DVM, MS
North Carolina State University
Raleigh, North Carolina

Peter Silley, PhD
MB Consult Limited
Lymington, Hampshire, United Kingdom

Gary E. Zurenko, MS
Micromyx, LLC
Kalamazoo, Michigan

Advisors

Jo Abraham, DVM, MS
Bayer Health Care LLC
Shawnee Mission, Kansas

Melanie R. Berson, DVM
FDA Center for Veterinary Medicine
Rockville, Maryland

Diane M. Citron, M(ASCP)
R.M. Alden Research Laboratory
Santa Monica, California

Ronald N. Jones, MD
JMI Laboratories
North Liberty, Iowa

Cindy Lindeman
Pfizer Animal Health
Kalamazoo, Michigan

Jennifer Lorbach
Trek Diagnostic Systems, Inc.
Cleveland, Ohio

Carol W. Maddox, PhD
University of Illinois
Urbana, Illinois

Marilyn N. Martinez, PhD
FDA Center for Veterinary Medicine
Rockville, Maryland

Patrick McDermott, PhD
FDA Center for Veterinary Medicine
Laurel, Maryland

Contents

Foreword

CLSI document M37-A3 is intended to offer guidance for sponsors (corporate or individual) that want to list interpretive criteria and quality control information in CLSI document M31[1] (Table 1, Group A) for a new and/or approved veterinary antimicrobial agent. CLSI welcomes presentations for antimicrobial agents originating from any country, not just the United States. Data developed according to M37, using relevant testing methods in other CLSI documents, are used by the Subcommittee on Veterinary Antimicrobial Susceptibility Testing (VAST) as the basis for establishing interpretive and quality control criteria for inclusion in the CLSI standard M31.[1] As the word "guideline" implies, this is not a mandatory step-by-step detailed protocol to apply to all new agents. Rather, it is intended as a statement of philosophy for the types of data useful for and/or required for making better judgments on interpretive criteria. The extent to which the guideline is followed remains the combined responsibility of the sponsor submitting a new agent and the Subcommittee on VAST. The sponsor is encouraged to consult the chairholder at any time to ensure the completeness of the presentation. The intent is to ensure that a "level playing field" is maintained, independent of manufacturer, veterinary health care professional, or government agency, in data presented to the subcommittee and in subcommittee determinations based on those data. Since the *in vitro* testing of some antimicrobial agents may present unique unanticipated situations, the minimal criteria outlined in this document might need to be expanded as problems become apparent during the data collection process.

This edition of M37, originally adapted from CLSI/NCCLS document M23,[2] has been modified to address more veterinary-specific issues, including a new paradigm to establish primary interpretive criteria. However, it retains basic guidelines on topics such as testing methodology, and quality control criteria that are consistent with those used for human-use antimicrobial agents in the CLSI Subcommittee on Antimicrobial Susceptibility Testing (AST). It is important to note that M37-A3 is not an alternative guideline to CLSI/NCCLS document M23[2] for those sponsors that seek to establish interpretive criteria for human use antimicrobial agents. Users of the document are referred to the Statement of Policy of the Antimicrobial Susceptibility Testing (AST) Standing Subcommittee of CLSI 20 February 2007, which does not apply the VAST. With the concurrent update of CLSI document M31,[1] *in vitro* tests for measuring the susceptibility of bacterial pathogens to veterinary antimicrobial agents are now available. Also, in CLSI documents M42[3] and M49,[4] testing methods for pathogens of aquatic species are now in place, and it is anticipated that M37-A3 will be used to generate interpretive criteria for those pathogens.

M37-A3 includes new sections based on lessons learned from implementation of M37-A2. Specifically, M37-A3 contains a new appendix that provides more rationale for the process of establishing breakpoints and interpretive criteria. As noted in CLSI document M31,[1] the subcommittee will now review data packages for treatments such as skin and soft tissue infections or enteric disease applications of antimicrobial agents per the M37-A3 guidelines. In recognition of the many generic antimicrobial agents used in veterinary medicine (that have been listed in CLSI document M31[1] and whose interpretive criteria, based on human clinical data, imported into CLSI document M31[1] from CLSI document M100's[5] Table 1, Group B), a new process to establish veterinary-specific interpretive criteria for them has been implemented. This document outlines the information needed to facilitate the decision-making process. Unlike the previous version, there are no mandatory requirements because it is expected that drug sponsors are now aware of the value of presenting as much information as possible to the subcommittee to achieve approval of quality control ranges or interpretive criteria for their products. To facilitate data presentation to CLSI VAST, sponsors are encouraged to begin data collection as early as possible in the clinical development phase.

In closing, I would like to recognize the outstanding efforts of the Subcommittee on VAST that made this revision possible. I would like to particularly acknowledge the individual members of the Editorial Working Group. Their willingness to sacrifice significant amounts of their personal time for the editing process and to address controversial topics demonstrates a real commitment to the CLSI process and the advancement of the veterinary and microbiology professions. In particular, I thank Marilyn Martinez for

her leadership on drafting the appendix that outlines the process of establishing breakpoints, and Jo Abraham, Melanie Berson, Bob Walker, Jeff Watts, and Steve Yan for their contributions. I would like to express my sincere appreciation to the CLSI staff for their ongoing support with the countless revisions, meetings, phone calls, and e-mails necessary to produce this document.

Finally, I would like to thank CLSI and the many participants in the CLSI consensus process for allowing me the privilege of serving as the VAST Chairholder.

Thomas R. Shryock, PhD, Past Chairholder
Subcommittee on Veterinary
Antimicrobial Susceptibility Testing

Key Words

Animal, antimicrobial agents, breakpoints, interpretive criteria, pharmacokinetics/pharmacodynamics, PK-PD, standard dilution methods for bacteria that grow aerobically, standard disk diffusion test, susceptibility testing, veterinary

Mission Statement

To develop and promote performance standards and interpretive criteria for *in vitro* antimicrobial susceptibility testing of bacteria isolated from animals

Development of *In Vitro* Susceptibility Testing Criteria and Quality Control Parameters for Veterinary Antimicrobial Agents; Approved Guideline—Third Edition

1 Scope

This document offers guidance for the development of quality control (QC) limits and interpretive criteria for antimicrobial susceptibility testing (AST), performed by disk diffusion and dilution testing with bacteria isolated from animals, for subcommittee review and, upon approval, inclusion in CLSI document M31.[1]

The guidance in this document applies to therapeutic antimicrobial agents intended for the treatment or control of systemic or organ-specific infectious disease processes in domestic animals (terrestrial or aquatic). Antimicrobial agents used for growth promotion or prophylaxis (disease prevention) are not included in this document. (See the discussion in CLSI document M31[1] for more details regarding this issue.) However, the testing methodology described for the development of QC standards may be applicable for those antimicrobial agents that are tested for epidemiological survey or other purposes for which a validated test is required. The subcommittee recognizes that antimicrobial agents are used to treat a variety of enteric infections in animals; thus, a concerted attempt to include them within CLSI document M31[1] should be made to guide practitioners in the proper selection of agents. **NOTE:** The guidelines do not apply to directly applied topical antimicrobials such as lotions, cream, ointments, or eye drops.

Since not all antimicrobial agents have veterinary-specific breakpoints or interpretive criteria, the subcommittee has imported breakpoints and zone diameters from CLSI document M100[5] (ie, human treatments) into Table 1, Group B of CLSI document M31,[1] and designated them by gray shaded listing. Since these breakpoints and interpretive criteria have been developed for human treatment applications, there is uncertainty as to how they apply to specific animal species and disease treatments. To facilitate moving CLSI document M100[5] interpretive criteria to veterinary-specific approved status, the Working Group on Generics will provide a gatekeeper function to ensure that presentations to the full subcommittee conform as much as possible to M37 requirements. This will allow for a consistent approach to address those situations where veterinary-specific data are not readily available within the public domain or where sponsors (ie, manufacturers) are not able or willing to provide data on their products.

Additionally, should there be a need to reevaluate previously established breakpoints or interpretive criteria, a process is outlined in Section 3.6.

2 Definitions

Susceptibility Testing

agar dilution susceptibility test – an *in vitro* antimicrobial susceptibility test method conducted using serial concentrations of an antimicrobial agent incorporated into an agar growth medium in separate petri dishes that are inoculated with a bacterial suspension to determine the minimal inhibitory concentration (MIC).

agar disk diffusion susceptibility test – an *in vitro* antimicrobial susceptibility test conducted using disks impregnated with a specified single concentration of an antimicrobial agent applied to the surface of an agar medium that has been inoculated with the test organism. The diameter of the zone of growth inhibition that results from the diffusion of an antimicrobial agent from the disks is measured with calipers or ruler and recorded in millimeters.

