



3rd Edition

VET04

Performance Standards for Antimicrobial Susceptibility Testing of Bacteria Isolated From Aquatic Animals

This document includes updated tables for the Clinical and Laboratory Standards Institute veterinary antimicrobial susceptibility testing guideline VET03.

A CLSI supplement for global application.

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Abstract

The data in the tables are valid only if the methodologies in CLSI document VET03¹ are followed. This guideline contains information about disk and broth dilution susceptibility test procedures for bacteria isolated from aquatic animals. The clinical importance of antimicrobial susceptibility test results demands that these tests be performed under optimal conditions and that laboratories have the capability to interpret results based on the most current clinical breakpoint or epidemiological cutoff value interpretive categories.

The tables presented in VET04 represent the most current information for drug selection, interpretation quality control using the procedures standardized in VET03¹. Users should replace previously published tables with these new tables. Changes in the tables since the previous editions appear in boldface type. Users should consider the interpretive categories presented in these tables most useful to isolates of *Aeromonas salmonicida*, *Aeromonas hydrophila*, *Flavobacterium columnare*, and *Flavobacterium psychrophilum*. Careful extrapolations may be possible to other bacterial species and with other similar antimicrobial agents, but only after consulting CLSI document VET09.² Fish disease diagnostic laboratories that typically conduct susceptibility testing less often than once per week should consult this document for revised guidance for frequency of QC.

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Foreword

It is important for users of CLSI document VET03¹ and VET04 to recognize that the standard methods described in CLSI documents are reference methods. These methods may be used for routine antimicrobial susceptibility testing of bacteria isolated from aquatic animals. The Working Group on Aquatic Animals envisions adding more aquaculture pathogens and antimicrobial agents to these (clinical) breakpoint and epidemiological cutoff value (ECV) tables as the data become available. Data needed to develop more clinical breakpoints could include, for example, a clinical effectiveness report that may be correlated with minimal inhibitory concentrations and/or zone diameters for a suspected pathogen obtained using standard methods. **If such data are available, individuals are strongly encouraged to contact any member of the Working Group on Aquatic Animals.**

Breakpoints and ECVs (defined in Sections II and III) established by CLSI may differ from those approved by various authorities for many reasons, including the use of different susceptibility databases, differences in data interpretation, and different public health policies. Differences also exist because CLSI proactively evaluates the need for changing clinical breakpoints. The reasons why veterinary breakpoints may change and the manner in which CLSI evaluates data and determines veterinary breakpoints are outlined in CLSI document VET02.³

Following a decision by CLSI to change an existing breakpoint, regulatory authorities may also review data to determine how changing a breakpoint may affect the safety and effectiveness of the antimicrobial agent for the approved indications. If the regulatory authority changes a breakpoint, commercial device manufacturers may have to conduct a clinical laboratory trial, submit the data to the regulatory authority, and await review and approval. For these reasons, a delay of more than the suggested CLSI "tentative" period of one year may be needed if a breakpoint change is to be implemented by a device manufacturer.

Overview of Changes

This supplement replaces the previous edition of the supplement VET03/VET04-S2, published in 2014. This list includes the major changes in this document. Other minor or editorial changes were made to the general formatting and to some of the table footnotes and comments. Changes to the tables since the previous edition appear in boldface type. The following are additions or changes unless otherwise noted as a “*deletion.*”

- **General:**
 - Changed document code from VET03/VET04 to VET04 to differentiate it from the methods guideline, VET03¹
 - **NOTE:** The broth dilution and disk diffusion methods, formerly in separate CLSI guidelines (VET03 and VET04, respectively) have been combined into one methods guideline, VET03.¹
 - Harmonized language and common information on methods and QC with CLSI documents VET01,⁴ VET08,⁵ M02,⁶ the *M02 Disk Diffusion Reading Guide*,⁷ M07,⁸ and M100.⁹
 - Updated genera formerly included in the family *Enterobacteriaceae* reorganized to an order (Enterobacterales) containing seven families: *Budviciaceae*, *Enterobacteriaceae*, *Erwiniaceae*, *Hafniaceae*, *Morganellaceae*, *Pectobacteriaceae*, *Yersiniaceae*¹⁰
 - To align with the International Organization for Standardization, changed the name of the inoculum preparation method in all appropriate tables from growth method to broth culture method and changed direct colony suspension to colony suspension
 - Reorganized and reformatted tables:
 - Breakpoint table for *Aeromonas salmonicida* (Table 1)
 - Epidemiological cutoff value (ECV) tables (Tables 2A through 2D)
 - QC tables with minimal inhibitory concentration (MIC) QC ranges (Tables 4A through 4F)
 - QC tables with disk diffusion QC ranges (Tables 5A through 5D)
- **Summary of CLSI Processes for Establishing Breakpoints and Quality Control Ranges (p. xv):**
 - Added new section
- **CLSI Reference Methods vs Commercial Methods and CLSI vs Regulatory Authority (p. xvi):**
 - Added new section
- **CLSI Aquatic Animal–Specific Breakpoint Additions/Revisions Since 2008 (p. xvii):**
 - Added new table of breakpoint additions and revisions since 2008, organized in alphabetical order by antimicrobial agent
- **CLSI Aquaculture Pathogen–Specific Epidemiological Cutoff Value Additions/Revisions Since 2008 (p. xvii):**
 - Added new table of ECV additions and revisions since 2008, organized in order of appearance in the tables by organism group (2A, 2B, 2C, 2D), then in alphabetical order by antimicrobial agent
- **Subcommittee on Veterinary Antimicrobial Susceptibility Testing Mission Statement and Responsibilities (p. xviii):**
 - Added new section