Technical Report No. 33 (Revised 2013)

Evaluation, Validation and Implementation of Alternative and Rapid Microbiological Methods
PDA Evaluation, Validation and Implementation of Alternative and Rapid Microbiological Methods

Technical Report Team

Authors

Michael J. Miller, Ph.D., Microbiology Consultants, LLC (Task Force Leader)
John Albright, bioMérieux, Inc.
Claude Anger, CBA MicroEnterprises
Dilip Ashtekar, Ph.D., Consultant
Peter Ball, Ph.D., Pall Life Sciences
Joseph Chen, Ph.D., Genentech Inc.
Steve Douglas, Hospira, Inc.
William Fleming, III., Ph.D., DosDocs Company, LP
Ren-Yo Forng, Ph.D., MedImmune, LLC
Gary Gressett, Excellent Pharma Consulting
Jianping Jiang, Instant BioScan, Inc.
Robert Johnson, Ph.D., Dialogue

David Jones, Ph.D., Rapid Micro Biosystems
Richard Levy, Ph.D., PDA
Daemon Lincoln, Ph.D., DL2 Limited
Patrick McCarthy, Millipore Corporation
Patrick McCormick, Ph.D., Bausch & Lomb, Inc.
Jeanne Moldenhauer, Excellent Pharma Consulting
Paul Newby, Ph.D., GlaxoSmithKline
Bryan Riley, Ph.D., US Food and Drug Administration
Miriam S. Rozo, Johnson & Johnson
Heather Wilson, Jubilant HollisterStier, LLC
Elizabeth Young, Formerly of Baxter Healthcare, Inc.
Pascal Yvon, AES – Chemunex, Inc.

Contributor

Oliver Gordon, Novartis Pharma Stein AG

DISCLAIMER: The content and views expressed in this Technical Report are the result of a consensus achieved by the authorizing Task Force and are not necessarily views of the organizations they represent.
Evaluation, Validation and Implementation of Alternative and Rapid Microbiological Methods

Technical Report No. 33 (Revised 2013)

ISBN: 978-0-939459-63-6
© 2013 Parenteral Drug Association, Inc.
All rights reserved.
Table of Contents

1.0 INTRODUCTION .........................................................1
  1.1 Scope and Purpose of the Technical Report .... 1
  1.2 Overview of Technical Report Structure .... 2

2.0 GLOSSARY OF TERMS ...........................................3

3.0 CLASSICAL MICROBIOLOGY AND THE MOVE TOWARD ALTERNATIVE AND RAPID METHODS .................................5
  3.1 Classical Microbiological Methods ..............5
  3.2 Introduction to Alternative and Rapid Microbiological Methods ..............6
  3.3 Regulatory Perspectives ..............................6
    3.3.1 United States ...................................7
    3.3.2 Europe ........................................8
    3.3.3 Japan and Australia .........................9
    3.3.4 Rest of World (ROW) .........................9
    3.3.5 When an RMM is Approved by One Regulatory Authority
         But Not Another ..........................10
  3.4 Business/Economic, Quality and Technical Considerations ..........................10
    3.4.1 Business and Economic Considerations 10
    3.4.2 Quality Considerations ...................11
    3.4.3 Technical Considerations .................11
  3.5 Risk Analysis .............................................12
  3.6 Vendors, Suppliers and Audits ..................12
  3.7 Automated Methods ..................................13

4.0 TECHNOLOGY REVIEW ............................................14
  4.1 Growth-based .............................................14
    4.1.1 Electrochemical Measurement ..............15
    4.1.2 Detection of Carbon Dioxide (CO2) ......15
    4.1.3 Utilization of Biochemical and Carbohydrate Substrates .........................15
    4.1.4 Digital Imaging and Auto-fluorescence of Micro-Colonies ...............15
    4.1.5 Fluorescent Staining and Laser Excitation of Micro-Colonies ..........15
    4.1.6 Use of Selective Media for the Detection of Specific Microorganisms ..16
    4.1.7 Measurement of Change in Head Space Pressure ..................16
    4.1.8 Microcalorimetry ................................16
  4.2 Viability-based ..........................................16
    4.2.1 Flow Cytometry ................................16
    4.2.2 Laser Scanning Solid Phase Cytometry ..16
    4.2.3 Direct Epifluorescence Filter Microscopy 17
  4.3 Cellular Component-based .........................17
    4.3.1 ATP Bioluminescence ..........................17
    4.3.2 Fatty Acid Profiling ............................17
    4.3.3 Matrix Assisted Laser Desorption Ionization Time of Flight (MALDI-TOF)
         Mass Spectrometry ........................17
    4.3.4 Surface Enhanced Laser Desorption Ionization Time of Flight
         (SELDI-TOF) Mass Spectrometry ..........18
    4.3.5 Fourier Transform-Infrared (FT-IR) Spectrometry ......................18
    4.3.6 Endotoxin Detection ...........................18
  4.4 Optical Spectroscopy ................................18
    4.4.1 Light Scattering/Intrinsic Fluorescence 19
    4.4.2 Raman Spectroscopy .........................19
  4.5 Nucleic Acid Amplification .........................19
    4.5.1 Polymerase Chain Reaction (PCR) ..........19
    4.5.2 Reverse Transcriptase (RT) PCR ..........20
    4.5.3 Ribotyping .....................................20
    4.5.4 Gene Sequencing ..............................20
    4.5.5 PCR and MALDI-TOF Mass Spectrometry ..........21
  4.6 Micro-Electro-Mechanical Systems (MEMS) ..................21
    4.6.1 Lab-On-A-Chip and Microfluidic Systems ...........21
    4.6.2 Microarrays ...................................21
    4.6.3 Other Technologies ............................21

5.0 THE VALIDATION PROCESS ...................................22
  5.1 Pre-Validation Activities ..................................22
    5.1.1 Proof of Concept (POC) ......................23
    5.1.2 Assessment of Supplier Capabilities/Supplier Audit ....................23
    5.1.3 Business Benefits or Return on Investment Considerations ..........24
  5.2 Validation of the Equipment, Software and Method ....................24
    5.2.1 Risk Assessment and Validation Planning ......................25
    5.2.2 User Requirements Specification (URS) ....................26
    5.2.3 Design Qualification (DQ) .....................26
    5.2.4 Functional Design Specification (FDS) ........27
    5.2.5 Requirements Traceability Matrix (RTM) ................27
    5.2.6 Standard Operating Procedures (SOPs) and Technology Training ..........27
    5.2.7 System Integration ................................27
    5.2.8 Installation Qualification (IQ) ..................28
    5.2.9 Operational Qualification (OQ) ..................28
5.2.10 Performance Qualification (PQ) ............... 28
5.2.11 On-going Maintenance and Periodic Reviews ............... 29
5.3 Establishment of Method Validation Criteria .. 29
  5.3.1 Accuracy .................................................. 31
  5.3.2 Precision .................................................. 32
  5.3.3 Specificity ................................................... 33
  5.3.4 Limit of Detection ......................................... 35
  5.3.5 Limit of Quantification .................................... 37
  5.3.6 Linearity ...................................................... 37
  5.3.7 Range ......................................................... 38
  5.3.8 Ruggedness .................................................. 38
  5.3.9 Robustness .................................................. 39
  5.3.10 Equivalence/Comparative Testing ............ 39
5.4 Suitability Testing ............................................. 41
  5.4.1 False Positive Testing .................................... 41
  5.4.2 False Negative Testing .................................. 42
5.5 Variability of Microbiological Methods: Additional Considerations ............................................. 43
  5.5.1 Preparation of Test Samples ......................... 43
  5.5.2 Sample Distribution Error ............................... 43
  5.5.3 Cellular Arrangement ..................................... 44
  5.5.4 Metabolic Activity ......................................... 44
5.6 Validation of Microbiological Methods: Additional Considerations ............................................. 44
  5.6.1 Alternative and Rapid Endotoxin Detection Methods .......................................................... 44
  5.6.2 Unique Methods Requiring Additional or Modified Validation Strategies .................................. 44
  5.6.3 Guidance on Changing Acceptance Criteria .......................................................... 45
5.7 Alternative and Rapid Microbial Identification Methods ..................................................... 46
5.8 Alternative and Rapid Methods for Mycoplasma Detection ..................................................... 46

6.0 IMPLEMENTATION: GUIDANCE ON SITE COMMISSIONING VERSUS INITIAL VALIDATION .................. 48
  6.1 Guidance for the Transfer of an Alternative or Rapid Method from an Originating Qualification Lab to a Separate Site/Manufacturing Facility 48
  6.2 Reduced Installation and Operational Qualification at the Site ...................................... 48
  6.3 Performance Qualification at the Site ............... 48
  6.4 Implementation of the Alternative or Rapid Method at the Site ....................................... 49

7.0 REFERENCES .............................................................. 50

FIGURES AND TABLES INDEX

Table 5.2-1 Validation Deliverables and Responsibilities ............... 25
Table 5.3-1 Method Validation Criteria ................................... 30
Microbiological testing plays an ever-increasing role in the pharmaceutical laboratory. In response to this, a variety of alternative and rapid methodologies that automate existing methods, make use of surrogate markers, or are based on wholly new technologies have emerged in recent years. These alternative methodologies offer significant improvements in terms of speed, accuracy, precision, and specificity over traditional, or classical, microbiology test methodologies.

The majority of testing performed today relies on century-old, conventional methods based on the recovery and growth of microorganisms using solid or liquid microbiological growth media. This is true in part because these methods can be appropriate for their intended use and have a long history of application in both industrial and clinical settings. They often are limited, however, by slow microbial growth rates, the unintended selectivity of microbiological culture, and the inherent variability of microorganisms in their response to culture methods. In spite of the limitations of classical culture methods, acceptance of alternative and potentially superior methods has only started to gain momentum within the pharmaceutical, biotechnology, and medical device industries. The Technical Report Team believes that the lack of clear guidance both on how to demonstrate the equivalence of alternative/rapid methods to existing methods in a manner acceptable to regulatory agencies and on how to validate the equipment associated with alternative/rapid methods is one impediment to the widespread adoption of these methods.

Considerable guidance can be found regarding the validation of chemical methods. Examples include USP General Informational Chapter <1225> Validation of Compendial Methods and the International Conference on Harmonisation (ICH) guideline Validation of Analytical Methods (1,2). These publications provide very specific instruction regarding the demonstration of alternative analytical chemistry methods and their equivalence to existing methods. Chapters introduced by the compendia, including USP General Information Chapter <1223> Validation of Alternative Microbiological Methods, and Ph. Eur. Informational Chapter 5.1.6 Alternative Methods for Control of Microbiological Quality, provide guidance on the steps needed to validate an alternative microbiological method (3,4). However, additional guidance is needed, as an understandable and holistic approach to the qualification and implementation of novel alternate microbiological methods, including rapid microbiological methods, still does not exist that would satisfy all regulatory agencies.

The original PDA Technical Report No. 33 was published in 2000 to fill this void. Industry, compendial, and regulatory developments since then, however, have necessitated this update to the guidance. The team believes that this revision is timely and will provide additional guidance to assist with the evaluation, validation, and implementation of the alternative microbiological methods.

This Technical Report was developed as a collaborative effort amongst representatives from alternative method suppliers and vendors, the pharmaceutical, biopharmaceutical and medical device industries, and regulatory agencies. It is intended to provide a comprehensive approach to the introduction of alternative microbiology methods in a government-regulated environment. It is anticipated that by providing agreed upon performance standards, the development, qualification and implementation of alternative microbiological methods will be greatly accelerated.

### 1.1 Scope and Purpose of the Technical Report

This Technical Report is intended to provide guidance for the successful evaluation, validation, and implementation of alternative and rapid microbiological methods needed by the pharmaceutical, biotechnology and medical device industries to assure product quality. Applications for these methods include, but are not limited to, the testing of microbial limits, sterility, and antimicrobial effectiveness; microbiological monitoring of clean rooms and other controlled environments and...