Overview of Rapid Microbiological Methods

Jeanne Moldenhauer

Abstract

In recent years there have been significant advances in microbiology, achieved through the sister sciences of chemistry, molecular biology, and computer aided imaging. These have resulted in a significant increase in the methods available for the detection, enumeration, and identification of microorganisms in the laboratory. This chapter provides a brief overview of the types of technologies available and the premise of how they work.

1. Introduction

The typical microbiology laboratory is a complex operation and involves the performance of a variety of laboratory testing methods. Many of the methods typically used in a microbiology laboratory have their basis in the works of Drs. Lister, Pasteur, and Koch. Many of these methods are over a hundred years old. In many cases, these methods are called conventional or traditional methods. In recent years, there has been an emphasis on updating or revising these methods to provide more sensitive, more accurate, or faster methods. This has resulted in a number of alternative or rapid microbiological methods (RMMs). Some countries also refer to these types of methods as “modern methods.” While these methods are called rapid microbiological methods, it may be more correct to refer to them as alternative methods, i.e., a different way of performing the methods.

It should be noted that many of these methods may be more expensive than their conventional counterparts, either in the direct cost of the test or in the time it takes to prepare the sample for testing. It is believed by many vendors of this equipment and some users that obtaining results more quickly is worth the cost of these systems.

The tragic events of 9/11 resulted in a large amount of money being invested by the United States Department of Defense in the development of methods to detect bioterrorism agents. Significant advances have been made as a result of this financial investment. Many of the developers are actively looking for other opportunities to use these systems to generate income. Since these types of systems were designed to detect very low levels of contamination, many of them can be applied for use in a pharmaceutical manufacturing environment.

This chapter provides information on the history of rapid microbiological methods, some of the alternative (rapid) microbiological methods that may potentially be used to replace the conventional methods (overview of the technologies), and some information on identification systems. The terms “alternative microbiological methods” and “rapid microbiological methods

Jeanne Moldenhauer  •  Excellent Pharma Consulting, Mundelein, IL
are used synonymously” in this chapter. It should be noted that thousands of systems are in some stage of development; therefore, only a limited number of technologies could be described in this chapter.

2. A History of Rapid Microbiological Methods: Industry Reluctance to Accept These Methods

While science moved forward in development of rapid/alternative microbiological methods, the industry has been slow to accept and implement these methods. One of the greatest fears of industry came from a concern that regulators would not recognize or accept these methods in place of traditional methods. Another concern was that companies would not be allowed to change test limits based upon the test method, i.e., they would use a superior method that was likely to detect more organisms and not be allowed to adjust the limits to accommodate the sensitivity of the new method.

3. Types of Microbial Testing Performed

Current testing methods are divided into three basic categories:

- Is something there?—Qualitative Testing
- If there, how much is there?—Quantitative Testing
- If there, what is it?—Identification Testing

While there are newer methods in all of these categories, there is an ongoing debate on whether identification testing should be considered as an alternative method or in a separate category, due to the differences in validation methods. No one questions whether it is newer, potentially faster, or better.

4. Types of Rapid Microbiological Methods

The classification systems frequently used for alternative methods are based upon how the technology works, e.g., growth of microorganisms, viability of microorganisms, presence/absence of cellular components or artifacts, nucleic acid methods, traditional methods combined with computer-aided imaging (which might also be considered automation of an existing method), and combination methods.

4.1. Growth-Based Technologies

These methods are based upon the measurement of biochemical or physiological parameters that reflect the growth of the microorganisms. Examples of these types of methods include: ATP bioluminescence, colorimetric detection of carbon dioxide production, measurement of change in head space pressure, impedance, and biochemical assays.

4.2. Viability-Based Technologies

Viability-based technologies do not require growth of microorganisms for detection. Differing methods are used to determine if the cell is viable, and if viable cells are detected, they can be enumerated. Examples of this type of technology include solid phase cytometry, flow fluorescence cytometry, and optical imaging with NADH detection (Moldenhauer 2005).