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Specifications for Immunological Testing for Infectious Diseases; Approved Guideline — Second Edition

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Abstract

Specifications for Immunological Testing for Infectious Diseases; Approved Guideline — Second Edition (NCCLS document I/LA18-A2) is intended for use by laboratorians who perform immunodiagnostic testing within clinical and reference laboratories. The document addresses the generic problems of preparation and characterization of antigens and antibodies; testing using these reagents; and understanding the results. *Specifications for Immunological Testing for Infectious Diseases; Approved Guideline — Second Edition* offers recommendations on specimen collection, handling, and storage, and performance criteria for the comparison of immunological test kits, as well as specifications for reference materials.

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Contents

Abstract i

Committee Membership v

Active Membership ix

Foreword xvii

1 Introduction 1

2 Scope 1

2.1 Standard Precautions 2

3 Definitions 2

4 Preliminary Information 7

4.1 Clinical Questions and Traditional Laboratory Methods 7
4.2 Immunoassays in Detection of Antigens from Infectious Agents 7
4.3 Immunoassays in Detection of Patient-Generated Antibodies 8

5 Immunochemical Methods 9

5.1 Tests for Antigen or Antibody in Liquid or Surface Specimens 9
5.2 Testing of Tissue Specimens (Primarily for Antigen) 10
5.3 Classes of Clinical Testing for Antigens and Antibodies 10

6 Qualification of Antigens 11

6.1 Types of Antigens 11
6.2 Ascertainment of Purity 12
6.3 Reporting by Manufacturers 12

7 Qualification of Antibodies; Immunochemical Specificity 12

7.1 Testing for Specificity 12
7.2 Causes of Nonspecificity 13
7.3 Monoclonal Antibodies 13
7.4 Reactivity with Immunoglobulin Classes and Subclasses 13
7.5 Methods 14
7.6 Antiserum Production 16
7.7 Nonspecificity, Undesirable Specificity, or Cross-Reactivity 17

8 Quantification of Antibodies (Titer) 18

9 Reference Materials, Proficiency Testing, and Challenge Panels 19

9.1 Calibrators, Controls, and Verifiers 19
9.2 Reference Materials 19
9.3 Proficiency Testing 19
9.4 Challenge Panels 21

10 Specimen Collection, Handling, and Storage 22

10.1 Specimen Collection 22
10.2 Specimen Handling 23
10.3 Specimen Storage 24

Contents (Continued)

11 Laboratory Interpretation of Tests 24

11.1 General Considerations 24
11.2 Criteria for Selection of Cut-Off Values 25
11.3 Clinical Predictive Value 25
11.4 Borderline Results 26
11.5 Evaluation of a Test 26
11.6 Reporting of Results 26

12 Evaluation of Test Results 27

13 Test Confirmation and Optimization 27

13.1 Precision vs. Accuracy 27
13.2 Physiologic Dynamic Range 28
13.3 Comparison of Quantitative Tests 28
13.4 Comparison of Qualitative Tests 28
13.5 Equivocal Results in Test Comparisons 28

14 Product Clearance; Recommendations for Package Inserts 29

14.1 Clearance by the Appropriate Regulatory Organization 29
14.2 Recommendations for Manufacturers of Kits and Reagents 29
14.3 Recommendations for the Clinical Laboratory 30
14.4 Recommendations for the Selection of Referral (Reference) Laboratories 31

References 33

Appendix. Interpretation of Laboratory Tests—Mathematical Concepts 36

Summary of Comments and Committee Responses 39

Related NCCLS Publications 41

Foreword

The intended audience for I/LA18-A2 — *Specifications for Immunological Testing for Infectious Diseases; Approved Guideline — Second Edition*, is clinical and reference laboratories that perform immunodiagnostic testing for infectious diseases, as well as the manufacturers of commercial test kits. To improve their positive and negative predictive values in diagnosis of disease, and to enhance interlaboratory comparability and performance, I/LA18-A2 promotes a better understanding of the requirements, capabilities, and limitations of these diagnostic tests.

The use of immunochemical methods for detection and quantification of agents of infectious disease, and of related antibodies, is increasing rapidly. Many of the assays in use, however, have significant problems with sensitivity and specificity. There are two basic types of immunodiagnostic tests for infectious disease: those that test for the presence of antigen(s) produced by the infectious agent(s), and those that measure antibody response to such antigens. Although the immunochemical methods are similar for both, potential problems with the two types differ with regard to sensitivity, specificity, correlation with the clinical stage of infection, and the like. Tests that are currently available vary greatly in reliability and accuracy.

Few, if any, guidelines or standards exist that address serological or immunological tests for infectious diseases. This document sets forth guidelines for the development and performance of immunodiagnostic tests for the detection of antigens from, and of antibody responses to, infectious agents. Also, the most commonly used immunological tests share basic specifications that apply to testing for many infectious agents and related antibodies; these specifications are the primary focus of this document. At the same time, it is recognized that there are exceptions, a few of which are addressed. Infections with the human immunodeficiency virus (HIV) and hepatitis viruses A-E, among others, have special requirements for testing and are not addressed in this document.

Key Words

Antibody, antigen, cross-reactivity, immunoassay, immunogen, sensitivity, specificity

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1 Introduction

Despite the great strides made in prevention and treatment, infectious diseases continue to exact a heavy toll from humankind.[1] While traditional pathogen-detection methods, such as culture, have established their credibility over time, they are often slow and relatively insensitive. More recently developed rapid immunoassay methods show great promise as adjuncts to the traditional methods used in clinical diagnosis. Immunoassay methods have been used for many years in the detection of antigen from infectious agents and immune response; newer methods have increased the speed (and often the specificity) of this type of testing as well. However, there are many problems—recognized and potential—that should be considered in the development and use of immunoassays for the detection of infectious disease.

2 Scope

The number of specific immunochemical tests, and modifications thereof, is already large and is increasing rapidly. Therefore, this document addresses testing only in a general manner; examples are given as appropriate. It is anticipated that many current and future tests will have characteristics and problems not addressed in this document. The basic concepts of sensitivity and specificity, from both a laboratory and clinical standpoint, will remain important in regard to future tests.

This document addresses several issues to which particular attention should be paid, including the following:

- Determination of purity and structure of antigens used as immunogens or as detection molecules in assays;
- Antibody specificity and quantification (titer or other measurement);
- The use of enhancing and amplifying agents;
- Interfering substances;
- Sensitivity and specificity criteria for test kits and procedures for assessing sensitivity and specificity;
- Patient preparation and sample collection and handling;
- Laboratory and clinical evaluation of test results;
- Specifications for reference materials;
- Storage conditions for samples and reference materials;
- Performance criteria for ensuring comparability of results among methods and laboratories; and
- Recommendations for product labeling and product literature, including limitations of the procedure(s).

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Related NCCLS Publications*

DI2-A2 **Immunoprecipitin Analyses: Procedures for Evaluating the Performance of Materials—Second Edition; Approved Guideline (1993) (Reaffirmed 1999)**. This document gives a description of, and procedures for, evaluating the performance of materials used in immunoprecipitin analysis, including a discussion of specificity.

DI3-A **Agglutination Analyses: Antibody Characteristics, Methodology, Limitations, and Clinical Validation; Approved Guideline (1993) (Reaffirmed 1999).** This document offers guidelines that describe specificities of antibodies and their required potency, labeling information, and characteristics and limitations of agglutination methods.

EP12-P **User Protocol for Evaluation of Qualitative Test Performance; Proposed Guideline (2000).** This document contains a protocol that optimizes the experimental design for the evaluation of qualitative tests, to better measure performance and provide a structured data analysis.

GP9-A **Selecting and Evaluating a Referral Laboratory; Approved Guideline (1998).** This guideline provides an outline of reasons and criteria for choosing a referral laboratory. A checklist for evaluating potential referral laboratories is included to assist in the decision process.

GP16-A **Routine Urinalysis and Collection, Transportation, and Preservation of Urine Specimens; Approved Guideline (1995).** This guideline describes routine urinalysis test procedures that address materials and equipment, macroscopic examinations, clinical analyses, and microscopic evaluation.

H3-A4 **Procedures for the Collection of Diagnostic Blood Specimens by Venipuncture; Approved Standard —Fourth Edition (1998).** This document provides procedures for the collection of diagnostic specimens by venipuncture, including line draws, blood culture collection, and venipuncture in children. It also includes recommendations on order of draw.

I/LA2-A **Quality Assurance for the Indirect Immunofluorescence Test for Autoantibodies to Nuclear Antigen (IF-ANA); Approved Guideline (1996).** This document offers guidelines for the development of reference sera of defined antibody specificity to ANA and standardization of the immunofluorescent test for ANA.

M29-A **Protection of Laboratory Workers from Instrument Biohazards and Infectious Disease Transmitted by Blood, Body Fluids, and Tissue; Approved Guideline (1997).** A consolidation of M29-T2 and I17-P, this document provides guidance on the risk of transmission of hepatitis viruses and human immunodeficiency viruses in any laboratory setting; specific precautions for preventing the laboratory transmission of blood-borne infection from laboratory instruments and materials; and recommendations for the management of blood-borne exposure.

* Proposed- and tentative-level documents are being advanced through the NCCLS consensus process therefore, readers should refer to the most recent edition.

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Related NCCLS Publications (Continued)

NRSCL8-A **Terminology and Definitions for Use in NCCLS Documents; Approved Guideline (1998).** This document provides standard definitions for use in NCCLS standards and guidelines, and for submitting candidate reference methods and materials to the National Reference System for the Clinical Laboratory.

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