CLINICAL AND LABORATORY STANDARDS INSTITUTE®

4th Edition

MM01

Molecular Testing for Heritable Genetics and Specimen Identification

This guideline provides recommendations for detecting and reporting genetic variation associated with heritable conditions.

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

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Molecular Testing for Heritable Genetics and Specimen Identification

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Abstract

Clinical and Laboratory Standards Institute guideline MM01—*Molecular Testing for Heritable Genetics and Specimen Identification* provides general recommendations for all phases of the operation of a molecular diagnostic laboratory focused on detecting and reporting genetic variation associated with heritable conditions. It explains different types of genetic variation, along with the corresponding standardized nomenclature. Various methods of detecting and characterizing genetic variants are described, as well as test-specific requirements for validation and quality assurance. This guideline also discusses special considerations for analyzing and reporting genetic variation associated with heritable conditions.

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Foreword

Profound technical advances in data acquisition and data analysis have propelled clinical genetic testing to the forefront of laboratory medicine and delivered the era of precision medicine. As a result, many medical laboratories routinely interrogate the entire genome to screen for, monitor, and/or diagnose health disorders; assess well-being; and treat medical conditions. The success of modern genetic testing relies on the proper use of complex analytics and bioinformatics.

This edition of MM01 focuses exclusively on genetic variation associated with heritable conditions. It is intended for use by laboratory staff who oversee all clinical, scientific, and/or operational aspects of the molecular laboratory. Individuals in these roles are responsible for introducing, developing, validating, implementing, and interpreting laboratory tests in the safest, most efficient, and most cost-effective manner possible. To this end, MM01 includes guidance on maintaining the highest standards of quality through robust QC systems.

Overview of Changes

This guideline replaces the previous edition of the approved guideline, MM01-A3, published in 2012. Several changes were made in this edition, including:

- Removing information on oncology markers and somatic or acquired genetic signatures associated with nonhematological cancers
  - See CLSI documents MM05,[1] MM21,[2] and MM23[3] for information on these topics.
- Updating the types of heritable genetic variation and nomenclature, including mitochondrial and epigenetic disorders (see CLSI document MM09[4])
- Revising methods and technologies, including next-generation sequencing for detection of genetic variation
- Updating considerations for preexamination, examination, and postexamination processes, with a focus on challenges posed by rare, heritable diseases
- Adding special considerations for accurate reporting and interpretation of carrier screening and noninvasive prenatal testing
- Adding special considerations for bioinformatics and genetic counseling

NOTE: The content of this guideline is supported by the CLSI consensus process and does not necessarily reflect the views of any single individual or organization.

KEY WORDS

cytogenetics
epigenetics
genetic disease
genetic variation
methylation
molecular diagnostic test
next-generation sequencing
nucleic acid

Chapter 1

Introduction

Molecular Testing for Heritable Genetics and Specimen Identification

1 Introduction

1.1 Scope

This guideline has been revised by experienced molecular laboratory professionals to focus on germline genetic diagnostic testing. It provides information for laboratories implementing molecular assays for inherited disorders, including both distributed test systems authorized by applicable regulatory agencies and complex laboratory-developed tests (LDTs). This guideline is intended for use by experienced laboratory directors, supervisors, and manufacturers involved in assay development, validation, verification, and interpretation of molecular testing.

Because molecular diagnostics is multidisciplinary in nature, this guideline provides an overview of the subspecialties that use similar technology or performance specification details. New to this edition are clinical applications of genetic identity testing. These applications include monitoring bone marrow transplant engraftment in hematopoietic stem cell transplantation, tracing sample provenance, and detecting sample contamination. Owing to advances in preimplantation diagnostics and prenatal testing, descriptions of noninvasive and cell-free DNA (cfDNA) detection methods are included.

This guideline does not cover biochemical genetics, paternity and forensics testing, blood banking, or detection of bioterrorism agents that require biosafety levels 3 or higher.

1.2 Background

Molecular testing has become essential for assessing a growing number of heritable disorders and pharmacogenetic responses. Genotyping can provide useful indicators of disease diagnosis, prognosis, and progression; guide treatment selection and response; and interrogate targets for gene-specific therapies. Genetic testing can be used in the prenatal setting to screen for and diagnose either suspected or at-risk disorders. In the postnatal setting, genetic testing is routinely used for newborn screening and for diagnosis of suspected disorders. In the pediatric and adult settings, in addition to diagnostics, genetic testing is used for carrier screening, presymptomatic or predictive testing, or assessment of potential treatment response.

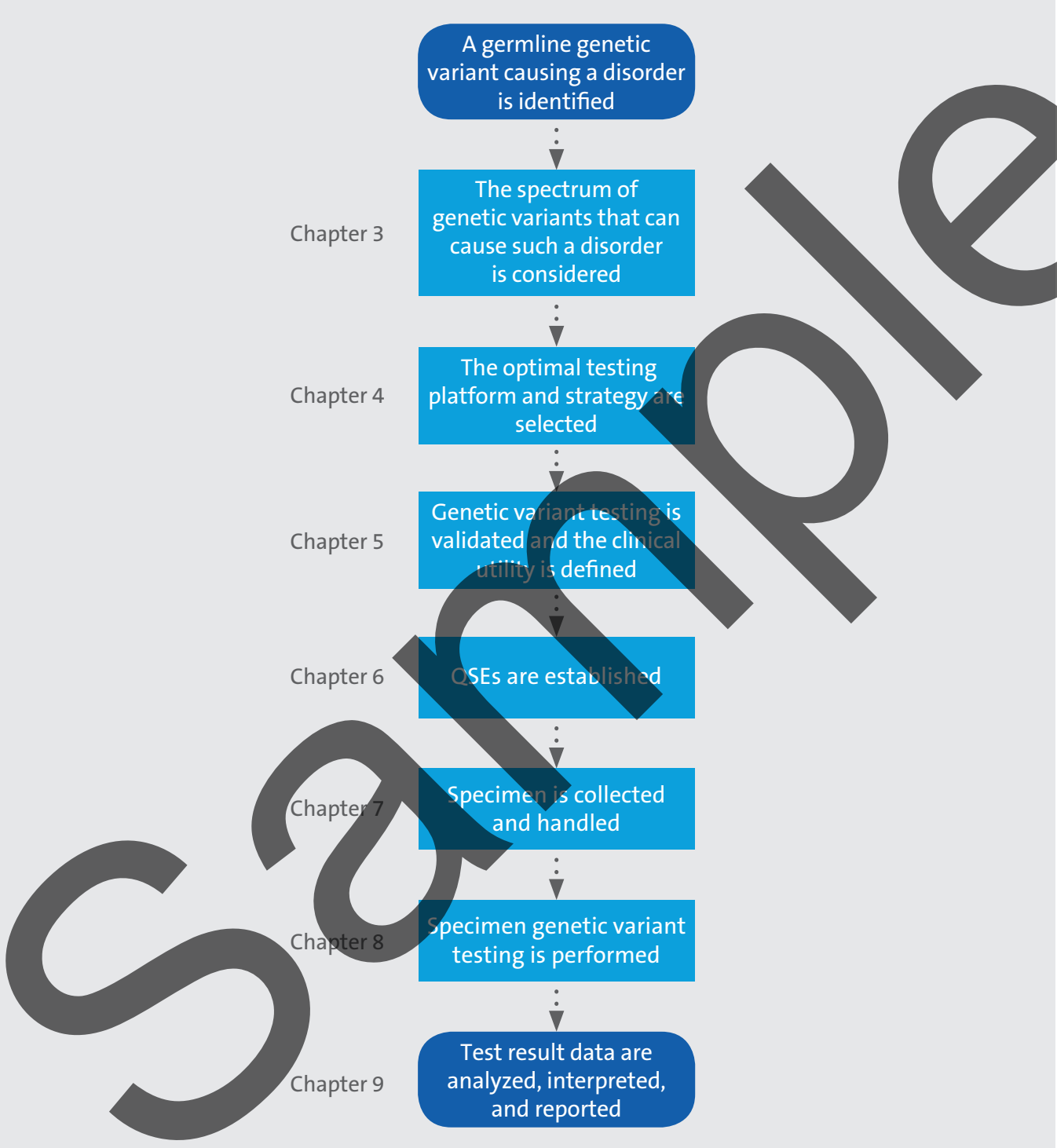
Although sensitivity, or threshold of detection, is essential to quantitative testing for genetic signatures of acquired diseases, this guideline primarily focuses on qualitative molecular testing. CLSI document MM06[5] discusses quantitative molecular methods that are applicable to detecting acquired genetic variations in neoplasias, such as linearity and measuring interval, lower limit of detection (LLoD) and lower limit of quantitation, results outside the measuring interval, and technologies used in molecular quantification.

When a molecular test is performed, laboratories should consider the preexamination, examination, and postexamination factors discussed in this guideline. In addition, it is important that the test methodology and results interpretation be based on the test's indication and application. Laboratories should also consider ethical concerns associated with genetic testing and the privacy of genetic information.

Securing qualified and competent personnel at all levels is key to producing accurate and timely medical laboratory test results. Specific qualifications and responsibilities vary from region to region. However, to ensure all personnel have the appropriate skills to perform their jobs competently, medical laboratories should have

2 Path of Workflow

Figure 1 depicts the processes of developing and performing molecular tests for heritable conditions.



Abbreviation: QSE, quality system essential.
[a] Three basic symbols are used in this process flow chart: oval (signifies the beginning or end of a process), arrow (connects process activities), box (designates process activities).

Figure 1. Processes for Developing and Performing Molecular Tests for Heritable Conditions[a]

Appendix A. (Continued)

Table A3. (Continued)

Risk Item Number	Mitigation (by design)	Mitigation (by process)	Mitigation (by use)	Is Risk Reduced to Lowest Possible Level (postmitigation)? (If no, additional mitigation measures applied)	Justification for Any Remaining Risk	Evidence of Effectiveness for All Mitigations Implemented
R-0003	• Analytical validation demonstrated 99.9% sensitivity and 99.6% specificity for all reported genotypes. • Batch QC includes positive synthetic QC samples for the 10 most prevalent mutations, a rotating set of 2 extracted DNA QC samples for the additional mutations, and a negative QC sample. • Outcomes are tracked.	N/A	N/A	• D = 5, unlikely before harm • O = 1, apparently random • S = 10, customer satisfaction, potential for permanent harm • Score = 50, intermediate risk	Risk cannot be completely eliminated, but mitigations have improved detectability and decreased likely occurrence.	6 months after assay launch, there has been 1 customer complaint of an inaccurate test result (0.001% of test volume), which is within analytical performance claims. In this case, an investigation determined, and an orthogonal method confirmed, that patient had a variation that affects annealing in upstream primer sequence, which likely caused a false-negative result. Mitigations were effective.

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