

Summary of Significant Changes to Proposed 5th Edition of *Standards for Parentage Testing Laboratories*

Chapter 1 - Organization

A new standard requires that the laboratory director have two years of experience in an accredited laboratory. Another new standard introduces the concept of a laboratory director designee, who must be under the direct supervision of the laboratory director. The designee can be a person with the appropriate doctoral degree and who is qualified by training, but not necessarily by experience. The designee may be trained until he or she has the required two years of experience, and then become eligible to be considered a laboratory director.

This edition of *Standards* also allows the laboratory director to delegate responsibility of tasks to other qualified individuals, however, ultimately responsibility lies with the laboratory director. Also, the term “technical supervisor” was changed to “laboratory supervisor” to avoid using a term that is used by CLIA but in a different context. In addition, when high-level staffing changes are communicated to the AABB National Office, evidence of qualification must now be provided, eg, a *curriculum vitae*. Lastly, there are new requirements for an annual management review of the quality system. Executive management must participate in this review of the quality system.

Chapter 2 – Resources

A new standard in this chapter requires that current job descriptions, including appropriate job qualifications, be maintained for all positions. Further, the training concept was expanded to require identification of training needs for staff and qualification of individuals performing critical tasks. The number of hours for continuing education was increased from 4 to 12. Personnel records for all employees are required, with more specific requirements for those authorized to perform or review critical processing steps. The standard on the expert witness was deleted because it was determined that this was a legal qualification beyond the scope of scientific/medical standards for parentage laboratories. (Laboratories should continue to list individuals who have previously qualified as experts. This aspect of service will be clarified in the forthcoming edition of the guidance document that accompanies these *Standards*.)

Chapter 3 – Equipment

This chapter requires that there be defined selection criteria for equipment. In addition, there are new equipment monitoring standards.

Chapter 4 – Supplier and Customer Issues

A new standard in this chapter requires management to be aware of supplier failures to meet specific requirements.

Chapter 5 – Process Control

In this chapter, there is an emphasis on validating new and/or revised processes and procedures. In this edition, successful participation in proficiency testing has been defined. Standards relating to unsuccessful participation in proficiency testing surveys are located in Chapter 7, where nonconformances are handled. The concept of inspection has been broadened to include final inspection, where the laboratory must ensure that the final parentage report or parentage testing service is acceptable before being delivered to customers, ie, clients or child support agencies. A new standard requires that biological materials be stored for 6 months after release of test report.

There are also new standards that are related to genetic privacy and confidentiality. There is a new section on nucleic acid amplification (NAT) for nucleotide sequence determination or single nucleotide polymorphism (SNP) analysis. In the calculation section, loci that are utilized for calculations must be statistically unlinked. In addition, the laboratory must validate methods used to convert NAT designation to serological equivalent types.

Chapter 6 – Documents and Records

In the 4th edition of *Standards*, the laboratory director was required to perform an annual review of all policies, processes, and procedures. In the proposed 5th edition, this task can be performed by an authorized individual. The standard requiring that documents have a version number was deleted. A new standard requires the laboratory to ensure that content is verified when a record has been copied and the original record will be destroyed, eg, microfiche. The computer standards were expanded and moved from Chapter 3, Equipment, to Chapter 6.

The record retention requirements have been incorporated into a chart. The chart lists the standard requiring that records be created, specific records be maintained, and relevant retention times.

Items to be included in parentage testing reports are also included in a chart. A new standard requires a statement that the calculation of parentage index compares the tested individual(s) to an unrelated person of the same racial/ethnic background.

Chapter 7 – Incidents, Errors, and Accidents and Nonconforming Parentage Test Reports, Materials, Samples, and Parentage Testing Services

This chapter has been expanded. When nonconformances are discovered, they must be evaluated, prevented from distribution (if they have not already been distributed), and, if they have been distributed, the laboratory must determine who received the parentage test reports or services. In the event of nonconforming proficiency test results, new standards apply.

Chapter 8 – Assessments: Internal and External

The requirement for self-assessments was deleted because it is linked to Standard 1.2.2, which requires an annual management review. When any assessment takes place (internal or external), the results need to be reviewed and appropriate corrective action taken.

Chapter 9 – Process Improvement Through Corrective and Preventive Action

Standards 9.121, 9.122, and 9.123 were deleted because they are covered by Standard 7.1. Standard 9.123 was moved Chapter 7 (Standard 7.1.3). The corrective action concept in Chapter 9 requires analysis of a summary of nonconformances while the intent of Chapter 7 is to analyze the specific nonconformance at the time in which it happens. Accordingly, these standards are better placed in Chapter 7 when the nonconforming sample, test result, or interpretation is discovered.

Chapter 10 – Facilities and Safety

There are no significant changes in this chapter.

PROPOSED *Standards for Parentage Testing Laboratories,*
5th Edition

DRAFT

1.0 ORGANIZATION



The Parentage Testing Laboratory (hereinafter referred to as the laboratory) shall have a ~~defined~~ structure that clearly defines and documents the parties responsible for the provision of parentage test reports and parentage testing services and the relationship of individuals responsible for key quality functions.

1.1 Executive Management

The laboratory shall have a defined executive management. Executive management shall have (1) responsibility and authority for the laboratory's operations; (2) the authority to establish or make changes to the laboratory's quality system; (3) the responsibility for compliance with these Standards and applicable laws and regulations; and (4) participation in management review of the quality system.

1.1.1 Laboratory Director Responsibilities

The laboratory shall have a ~~laboratory~~ director who has a doctoral degree in a biologic science, and is qualified by sufficient advanced training and/or practical experience in those methods the laboratory employs for parentage testing. The laboratory director shall have responsibility and authority for all policies, processes, and procedures ~~used for parentage testing.~~

-  1.1.1.1 The laboratory director shall have two years of experience in an accredited laboratory performing parentage testing.
-  1.1.1.2 The laboratory director or a designee who has a doctoral degree in a biologic science and is qualified by sufficient advanced training and is under the direct supervision of the laboratory director shall review critical test results, worksheets that record interpretations and conclusions, and case reports.
- 1.1.1.3 The laboratory director may delegate responsibility to another qualified individual; however, the laboratory director shall retain ultimate responsibility for laboratory director duties.
- 1.1.1.4 The laboratory shall communicate staffing changes and provide evidence of qualification for the laboratory director and ~~technical~~ laboratory supervisors within 30 days of the change to the American Association of

Blood Banks (AABB).

1.2 **Quality System**

A quality system shall be defined, documented, implemented, and maintained. All personnel shall be trained in its application. ~~The laboratory director shall establish and maintain a quality system and ensure that the quality policy is understood and implemented by personnel at all levels.~~

1.2.1 **Quality Representative**

The quality system shall be under the supervision of a designated person who reports to executive management.



1.2.2 **Management Reviews**

Management shall assess the effectiveness of the quality system through annual management reviews.

1.3 **Policies, Processes, and Procedures**

Quality and operational policies, processes, and procedures shall be developed and implemented to ensure that the requirements of these Standards ~~and the quality system~~ are satisfied. All such policies, processes, and procedures shall be in writing or captured electronically and shall be followed.

2.0 **RESOURCES**

The laboratory shall have policies, processes, and procedures to ensure the provision of adequate resources to perform, verify, and manage all activities in the laboratory.

2.1 **Human Resources**

The laboratory shall have a process to ensure the employment of an adequate number of qualified (by education, training, or experience) individuals. Current job descriptions shall be maintained and shall define appropriate qualifications for each job position.

2.1.1 **Training**

The laboratory shall have a process for identifying training needs and shall provide for the training of all personnel performing activities affecting quality. Personnel performing critical tasks shall be qualified on the basis of appropriate education, training, and/or experience.

2.1.1.1 **Continuing Education**

All employees performing critical steps ~~Technical staff~~ shall participate in at least ~~4~~ 12 hours of relevant continuing education on an annual basis.

2.1.2 **Competence**

Evaluation of continued competence shall be performed at specified intervals.

2.1.2.1 **Competency Testing**

All employees performing critical steps ~~Technical staff~~ shall successfully participate, three times a year, in the testing of proficiency or shared competency samples for every methodology in which they participate.

2.1.3 **Personnel Records**

Personnel records for each employee shall be maintained. For those authorized to perform or review critical processing steps, records of names, signatures, initials or identification codes, and inclusive dates of employment shall be maintained.

Expert Witness

~~A qualified person shall be available to act as an expert witness in the event that legal testimony related to test results is required. (2.300)~~

3.0 **EQUIPMENT**

The laboratory shall identify the ~~instruments, measuring devices~~ equipment that is critical to the provision of parentage test reports and parentage testing services. The laboratory shall have policies, processes, and procedures to ensure that calibration, maintenance, and monitoring of equipment conforms to these Standards and other ~~applicable regulations and requirements~~ specified requirements.

3.1 **Selection of Equipment**

The laboratory shall have a process to define the selection criteria for equipment.


3.1.1 All equipment shall be qualified for its intended use.

3.2 **Unique Identification of Equipment**

Critical equipment shall have unique identification.

3.3 **Control of Critical Equipment**

The laboratory shall have a process for scheduled monitoring of all critical equipment.

 3.3.1 Monitoring of critical equipment shall include the following elements:

- 1) Calibration and adjustment of equipment prior to use and at prescribed intervals, with equipment having adequate accuracy and precision.
- 2) The calibration process shall include details of equipment type, unique identification, location, frequency of checks, check method, acceptance criteria, action to be taken for unsatisfactory results, labeling of equipment showing calibration status, and where applicable, safeguards to prevent equipment from adjustments that would invalidate the calibration setting.
- 3) Assessment of the conformance of parentage testing reports and parentage testing services provided when equipment is found to be out of calibration.

3.3.2 Laboratory personnel shall be alerted of equipment malfunction.

[3.300 “Computer Software Use for Process Control” was moved to Chapter 6.]

4.0 SUPPLIER AND CUSTOMER ISSUES

The laboratory shall have policies, processes, and procedures to ~~ensure~~ evaluate the ability of providers of critical materials and services to meet agreed upon requirements consistently.



4.1 Supplier Qualification

The laboratory shall evaluate and participate in the selection of suppliers prior to acceptance ~~preparation~~ of an agreement ~~purchase order~~.

4.1.1 When a supplier fails to meet specified requirements, it shall be reported to management with contracting authority.

4.1.2 Services Tests required by these Standards ~~and performed in another laboratory~~ shall be performed in a laboratory either accredited by the American Association of Blood Banks (AABB) or other equivalent accrediting body.

4.2 Agreements

Agreements, or changes to those agreements, to obtain or provide materials, parentage test reports, and parentage testing services shall define supplier and customer expectations and shall reflect agreement.



4.2.1 Agreement Review

Agreements shall be reviewed, and changes shall be incorporated as needed.



4.3 Incoming Receipt, Inspection, and Testing

Incoming materials and samples shall be received, inspected, and tested, as necessary, prior to acceptance or use.

4.3.1 Upon receipt of a sample, the laboratory shall verify package integrity. [Standard moved from Chapter 5, Process Control.]

4.3.2 Critical materials shall meet specified requirements.

5.0 **PROCESS CONTROL**

The laboratory shall have ~~a process to ensure that all processes and procedures are followed~~ policies and validated processes and procedures that ensure the quality of the parentage test reports and parentage testing services. The laboratory shall ensure that these policies, processes, and procedures are carried out under controlled conditions.

5.1 **General Elements**

5.1.1 **Change Control**

The laboratory shall have a process to develop new processes and procedures or change existing ones. This process shall include identification of specifications and verification that specifications have been met. Prior to implementation, the new or changed processes and procedures shall be validated. The laboratory shall have ~~a process to ensure development, verification, validation, and implementation of specifications of any new or changed services.~~

5.1.1.1 The laboratory shall ensure that the implementation of new or changed processes is controlled.

5.1.2 **Proficiency Testing Program**

The laboratory shall participate in a proficiency testing program, if available, for each genetic system used for reporting test results.

5.1.2.1 The laboratory shall successfully participate in a graded external proficiency testing program (1) three times a year for each genetic system routinely analyzed in the laboratory, and (2) once a year for any genetic system occasionally used in the laboratory. ~~If accredited by the AABB,~~ The laboratory shall designate the AABB to receive a copy of their individual evaluation from the sponsoring agency.

5.1.2.1.2 Participation in proficiency testing shall include 100% correct interpretation of exclusion or non-exclusion of an alleged father in the survey. Unsuccessful proficiency testing results shall be handled in conformance with Standard 7.2, Nonconforming Proficiency Testing Results.

5.1.2.2 When a formal proficiency testing program is not available, the laboratory shall participate, three times a year, in a sample

exchange program for determining the accuracy and reliability of test results.



5.1.3 Quality Control

A program of quality control that is sufficiently comprehensive to ensure that reagents, equipment, and methods function as expected shall be established. Results shall be reviewed and corrective action taken, where appropriate.

5.1.4 Use of Materials

All materials that are used to collect, store, and test samples shall be used in accordance with manufacturer's written instructions and shall meet specified requirements.



5.1.4.1 Reagents

Reagents that are prepared by the facility shall meet or exceed applicable criteria.



5.1.4.1.1

The reactivity and/or specificity of all critical reagents and/or controls shall be evaluated and deemed acceptable before use in recording test results. [Standard moved from Chapter 4.]

5.1.5 Identification and Traceability

The laboratory shall ensure that samples are identified and traceable from collection through testing and reporting of results.

5.1.6 Inspection

The laboratory shall have a process to ensure that samples are inspected at laboratory-defined stages to verify that specified requirements are met. The laboratory shall develop and implement testing processes and procedures for inspection and testing activities to ensure that the requirements of these Standards are met.

~~*Samples shall be inspected and reviewed at critical stages of processing and held until they pass specified in-process and final inspections. Records of sample inspection shall be maintained. (5.510)*~~

~~*The laboratory shall control receipt and dispatch of samples and reagents to and from storage areas. (5.512)*~~

5.1.6.1 Final Inspection

The laboratory shall have a process to ensure that finished parentage test reports and parentage testing services are acceptable prior to distribution or delivery.

5.1.7 Handling, Storage, Distribution, and Transportation

The laboratory shall have a process to ensure that samples and ~~critical materials~~ reagents are handled, stored, distributed, and transported in a manner that ~~ensures traceability~~ prevents damage and limits deterioration.

5.1.7.1 All remaining biological materials obtained from a tested individual shall be stored for a minimum of 6 months after the release of the parentage testing report.

5.1.8 Privacy and Confidentiality

The laboratory shall have a policy to ensure that the parentage testing process is private and confidential.

5.1.8.1 The laboratory shall not release test results for any purpose other than that relevant to the relationship testing without a court order or the written permission of the individuals who furnished the samples.

5.1.8.2 The laboratory shall not release an identifiable sample of an individual for any purpose other than that relevant to the actual testing for which the sample was submitted without a court order or the written permission of the individual who furnished the sample.

5.2 Sample Collection

The laboratory shall have processes and procedures for informed consent, collection, verification of sample collection, and acquiring and maintaining of identification records.



5.2.1 Informed Consent

Prior to ~~initiation of the~~ sample collection, informed consent, according to applicable law, shall be obtained from each tested person, or, in the case of a minor child or legally incompetent adult, from a legal guardian or conservator.



5.2.1.1 If a test participant has no legal authority to authorize

consent, it shall be obtained from one who has legal authority as determined by state law in the jurisdiction. Evidence ~~Proof~~ of legal authority shall be provided.



5.2.2 Collection

All biologic samples for testing shall be collected by persons with no interest in the test outcome. The person may be either:

- 1) An employee or agent of the laboratory, or a person who collects samples during the course of ordinary business.
- 2) A person capable of collecting the particular biologic sample. In this case, a second person with no interest in the test outcome shall serve to verify and witness the collection process.

5.2.2.1 Collection methods shall protect the safety of the person from whom the sample is taken, preclude contamination, and maintain integrity of the sample.

5.2.3 Verification of Sample Collection

The person collecting the sample and verifying the process, if applicable, shall confirm that (1) the identification of the tested person is accurate and the stated relationship is recorded; (2) informed consent was obtained; (3) the sample was collected from the intended person; (4) the label is accurate; and (5) the sample is packaged in a tamper-proof manner.

5.2.3.1 Each sample shall bear an affixed label containing the following information: (1) a unique identification for each sample collected, (2) date of collection, and (3) initials of the person collecting sample. The label shall not be obscured, altered, or removed.

5.2.3.2 The person who provides legal consent for the collection of the sample shall verify the accuracy of its affixed label in writing.

5.2.3.3 Collected samples that will be transported to the laboratory shall be sealed in a tamper-proof package by the person collecting or verifying the sample. Test participants shall not package or transfer samples.



5.2.4 Identification Records

The following records relating to each sample collected shall be

acquired and maintained.

5.2.4.1 Name, alleged relationship, date of birth, and the race/ethnic background of each parent/alleged parent.

5.2.4.2 Date of birth of the child.

5.2.4.3 Place and date of sample collection.

5.2.4.4 Printed name, signature, and contact information ~~Name and place of employment~~ of the person collecting the sample, if applicable.

5.2.4.5 Printed name, signature, and contact information ~~Name~~ of the person verifying collection process, if different from the person collecting the sample.

5.2.4.6 A history of transfusion in the preceding 3 months, or allogeneic hematopoietic progenitor cell transplantation.

5.2.4.7 Original or legible photocopies of one or both of the following items:

- 1) Government photo identification.
- 2) Photograph taken at the time of collection that is suitable for positive identification.

5.2.4.7.1 The original or legible photocopy of the identification record shall be signed and dated by the test participant or the person providing legal consent for the collection of the sample and shall be signed ~~initialed~~ by the person collecting the sample.

5.2.4.8 Special Circumstances

In rare circumstances, where verification of identity cannot be obtained according to these *Standards*, positive sample identification (eg, prenatal sample, coroner's sample, or samples provided by law enforcement agencies) shall be obtained.

5.3 Testing

The laboratory shall utilize a group of tests identified in these *Standards* that include multiple independent genetic systems as the basis for its findings. This group of tests shall, with rare exceptions, provide a nonexcluded alleged parent with a parentage index of at least 99.

5.3.1 A standard method of nomenclature for describing phenotypes in each genetic system shall be used. (See 5.4.1.4, Chart 6.4A)

5.3.1.1 For any apparent homozygote, only the observed result shall be listed.

5.3.2 Before the laboratory adds to its test battery a new test that is covered in these *Standards*, it shall first successfully complete validation studies. Records of validation results shall be sent to the AABB. The program shall meet the following criteria:

- 1) The validation protocol shall require the analysis of at least 20 biological test samples with consistency of test results within the laboratory (precision) and between the laboratory and other laboratories (accuracy).
- 2) The power of exclusion for the system and the typical parentage index in nonexclusion trios of the ethnicity typically seen in the laboratory shall be determined and compared with known values as part of the validation process.
- 3) The new method shall be used as a supplemental test only until the next regularly scheduled AABB assessment.

5.3.3 New test methods not covered by these *Standards* shall not be implemented, except as supplemental tests, and shall not be used as the basis for a finding of parentage until they are approved by the AABB Parentage Testing Standards Program Unit.



5.3.4 All cases shall be reviewed by two people, one of whom shall be the laboratory director or designee as defined by Standard 1.1.1.2.

5.3.5 The laboratory shall ensure that only those methods for which a confirmatory test is available are used.

5.4 Specific Testing Methods

Specific testing methods shall ensure that accurate results are produced. Appropriate test controls shall be conducted to ensure accurate results.



5.4.1 Serological Testing

When serological testing is performed, the laboratory shall use processes and procedures that are generally accepted by the scientific community. Two independent tests shall be performed per sample for each test system.

5.4.1.1 Red Cell Surface Antigens

Red cell typing reagents shall be shown to be reactive and specific on the day of use by the method employed.

5.4.1.1.1 Red cell typing controls shall be heterozygous when possible.

5.4.1.2 Red Cell Enzyme and Serum Protein Testing

When red cell enzyme and serum protein testing is performed, phenotype designations shall be based on two independent readings of the electrophoretic patterns.

5.4.1.2.1 Heterozygous phenotype controls shall be incorporated into testing programs, when possible.

5.4.1.3 Immunoglobulin Allotype Testing

Immunoglobulin allotyping shall be performed only on children over 6 months of age.

5.4.1.3.1 Appropriate positive and negative controls shall be incorporated to ensure accurate results.

5.4.1.4 HLA Antigen Testing

When serological testing for HLA antigens is performed, the laboratory shall type for at least HLA-A and HLA-B antigens officially recognized by the 1980 HLA Nomenclature Committee of the World Health Organization (WHO) and use terminology that is consistent with the current WHO nomenclature.

- 1) A standard complement-dependent lymphocytotoxicity method shall be used for serological HLA-A and HLA-B typing and shall incorporate the following appropriate controls to ensure accurate results.
- 2) Cell viability in the negative control well at the end of

incubation shall be recorded and shall be sufficiently high to ensure accurate results.

- 3) Each new batch of complement shall be tested to determine that it induces cytotoxicity in the presence of specific antibody at least one dilution beyond that selected for use but is not cytotoxic in the absence of specific antibody. Control procedures shall be established to ensure continued reactivity of stored complement.
- 4) Each typing tray shall include at least one complement-dependent positive control serum and at least one negative control serum (or serum pool) known to lack cytotoxic antibody.
- 5) Reagent specificity, both purchased and procured internally, shall be ensured through prospective analysis against well-defined cells, retrospective analysis of results from routine typing, and verification by an independent laboratory.
- 6) Each HLA-A and -B antigen shall be defined by at least two different operationally monospecific sera, by one mono-specific serum plus two multispecific sera, or by three partially nonoverlapping multispecific sera.
- 7) Assignment of each HLA antigen shall be based on reactions observed on two independent trays or tray sets. Each tray or tray set shall be read independently.



5.4.2 Nucleic Acid Testing (NAT) ~~(DNA Polymorphism Testing)~~

When NAT is used, the laboratory shall use processes and procedures that are generally accepted by the scientific community. The laboratory shall have a process that demonstrates reproducibility for each test result.

- 1) DNA loci used for parentage testing shall be validated by family studies to demonstrate that the loci exhibit Mendelian inheritance. The mutation frequency shall be documented and used appropriately for loci routinely used by laboratory ~~shall be known and shall be low.~~

- 2) The chromosomal locations of the polymorphic loci used for parentage testing shall be documented in the scientific literature ~~those recorded by the International Human Gene Mapping Workshop.~~
- 3) For systems dependent on accurate measurement of allele sizes, a human DNA control of known phenotype shall be tested with each analysis.
- 4) Alleles shall be identified by nomenclature consistent with the relevant scientific literature.
- 5) Appropriate stringency conditions shall be utilized to ensure accurate allele determination.

5.4.2.1 Restriction Fragment Length Polymorphism Testing

When restriction fragment length polymorphism (RFLP) testing is used, the process shall incorporate the following requirements:

- 1) The conditions of hybridization and size(s) of variable and constant fragments associated with each DNA locus shall be documented in the laboratory.
- 2) A method shall be available to ensure complete endonuclease digestion of DNA for testing.
- 3) ~~Sizing~~ Ladders composed of discrete fragments of known size shall span the range of allele sizes routinely encountered at DNA loci being tested. Size markers shall be used with sufficient frequency to accurately ~~size~~ determine allele fragment size.
- 4) DNA profiles shall be read and interpreted twice, independently.
- 5) Results involving closely spaced alleles at a single locus shall be evaluated using co-electrophoresis or other appropriate methods. ~~Co-electrophoresis of DNA from persons whose relationship is at issue shall be an integral part of quality assessments and shall be performed with rare exception.~~

- 6) The resolution of alleles shall be determined internally and considered appropriately in the calculations.

5.4.2.2 Nucleic Acid Amplification (NAT) for Short Tandem Repeat (STR) and Amplified Fragment Length Polymorphism (AMFLP) Analysis

When NAT testing is used for STR analysis, the process shall include the following requirements:

- 1) Results shall be read and interpreted twice, independently.
- 2) When electrophoresis is used, ladders composed of discrete fragments of known size or tandem repeat number shall encompass the range of allele sizes routinely detected at the locus in question. Flanking size markers shall be used with sufficient frequency to accurately determine allele size.
- 3) STR/AMFLP alleles shall be identified by repeat number as adopted by the International Society of Forensic Haemogenetics.
- 4) Results involving closely spaced alleles at a single locus shall be evaluated using co-electrophoresis or other appropriate methods.



~~Co-electrophoresis of DNA from persons whose relationship is at issue shall be an integral part of quality assessments and shall be performed with rare exception.~~

- 5) Negative control(s) shall be used to monitor for sample contamination and NAT product contamination.
- 6) Pre Post-amplification samples shall be prevented from contaminating post pre-amplification materials.
- 7) Appropriate positive controls shall be included to ensure the accuracy of the results.

5.4.2.3 Nucleic Acid Amplification (NAT) for Nucleotide Sequence Determination or Single Nucleotide


Polymorphism (SNP) Analysis

When NAT is performed for sequencing or SNP analysis, the process shall include the following requirements:

- 1) Alleles determined using a reverse dot blot method shall be read twice, independently.
- 2) Alleles determined using a reverse dot blot method shall only be called when allele intensity is greater than the sensitivity control.
- 3) The conditions for hybridization shall be controlled to ensure accurate allele determination.
-  4) When a site-specific oligonucleotide probe (SSOP) method is used for HLA allele determination, a control probe shall be used to ensure adequate target nucleic acid is available for analysis.
-  5) When a site-specific primer (SSP) method is used for HLA allele determination, a positive internal control primer shall be included to verify that amplification has occurred for each reaction.
- 6) SNP analysis utilizing fluorescent labeled reagents shall incorporate a control to ensure adequate target nucleic acid is available for analysis.
- 7) Alleles determined by sequence analysis shall confirm the nucleotide sequence by analysis of both strands of nucleic acid.
- 8) Negative control(s) shall be used to monitor for sample contamination and NAT product contamination.

5.5 Calculations

Calculation methods shall be validated. The loci utilized for calculations shall be shown to be statistically unlinked to be used independently.

-  **5.5.1** All calculations, including manual and computer-assisted analyses, shall be reviewed by a laboratory technical supervisor and/or laboratory director before serving as the basis for a final report.

5.5.2 Validation of Calculations

Formulas used for statistical calculations shall be specified and the databases used as sources of allele frequencies shall be validated.

5.5.2.1 The laboratory importing a valid database from another laboratory shall share sufficient samples with that laboratory to ensure accuracy in identifying alleles by size, repeat number, or sequence.

5.5.2.2 For the laboratory using RFLP methods where allele size or sequence is critical, a minimum of 20 samples shall be shared that include alleles encompassing 90% of the size range detected at that locus. For NAT methods, a minimum of 20 samples shall be shared that include alleles encompassing the size range of alleles as defined by sizing ladders.

5.5.2.3 The laboratory using a valid database for HLA-A and HLA-B alleles in which haplotype frequencies are based on serological equivalent types shall validate the method used to convert nucleic acid allele designations to serological equivalent types.

5.5.3 If only manual calculations are performed, they shall be performed in duplicate.



[Requirements for Testing Reports have been moved to Chart 6.4A]


6.0 DOCUMENTS AND RECORDS

The laboratory shall have policies, processes, and procedures to ensure that documents are identified, reviewed, approved, and retained and that records are created, stored, and archived in accordance with record retention policies.

6.1 Documents

The laboratory shall have a process for document control that includes the following elements:

- 6.1.1 A master list of documents, including policies, processes, procedures, labels, and forms that relate to the requirements of these Standards.
- 6.1.2 Use of a standardized format for all policies, processes, and procedures. Additional procedures (such as those in an operator's manual) may be incorporated by reference.
-  6.1.3 Review and approval ~~by the laboratory director~~ of new and revised documents prior to use.
-  6.1.4 Annual review of each policy, process, and procedure by an authorized individual ~~the laboratory director~~.
- 6.1.5 Use of only current and valid documents. Appropriate and applicable documents shall be available at all locations where activities essential to meeting the requirements of these Standards are performed.

~~Documents that are periodically revised shall be identified with a version number.~~
-  6.1.6 Identification and appropriate archival ~~and protection~~ of obsolete documents.

6.2 Records

The laboratory shall ensure identification, collection, indexing, access, filing, storage, and disposition of records.

6.2.1 Facility Records

Records shall be complete, retrievable in a period of time appropriate to the circumstances, and protected from accidental or unauthorized destruction or modification. Chart 6.2A, Retention of Records, applies.

~~The laboratory shall retain the following records for five years, or as required by applicable law:~~

~~Records related to each parentage case (6.310)~~

~~Superseded policies, processes, and procedures (6.320)~~

6.2.1.1 Copies

The laboratory shall have a process to ensure that copies of records are verified as containing the original content and shall be legible, complete, and accessible before the destruction of the original record.

6.2.2 A system designed to prevent unauthorized access and ensure confidentiality of all records shall be established and followed.

~~Prevention of unauthorized access to records, confidentiality of all parentage case records, and the privacy of the individual being tested shall be maintained.~~

[Genetic privacy and confidentiality are now in Standard 5.1.8]

6.2.1 Reports shall be released only to authorized individuals.

6.2.3 The record system shall make it possible to trace any parentage test report or parentage testing service from its source to final disposition and to review the records applying to the specific parentage test report or parentage testing service.

6.3 Computer Systems

The laboratory shall have a process to support the introduction of new software, hardware, or databases, or modifications of existing software, hardware, or databases relating to the requirements of these Standards.

This process shall include:

- 1) risk analysis, training, validation, implementation, and evaluation of post-implementation performance;
- 2) description of system maintenance and operation;
- 3) documentation that is written in language that is understandable to the user;
- 4) a system for display and verification of data before final acceptance when data are added or altered;

- 5) description of how modifications to the system are authorized and documented.



6.3.1 Computer Records

Records of the following shall be maintained:

- 1) Validation of system software, hardware, databases, and user-defined tables;
- 2) Fulfillment of life-cycle requirements for internally developed software;
- 3) Numerical designation of system versions, if applicable, with inclusive dates of use;
- 4) Monitoring of data integrity for critical data elements.

6.3.2 An alternative system that ensures continuous operation shall be available in the event that computerized data and computer-assisted functions are unavailable. The alternative system shall be tested periodically.

6.4 Testing Reports

When the tests have been completed, a report shall be generated and shall include the information required by Chart 6.4A, Requirements for Parentage Testing Reports.

6.4.1 Inconclusive or Unusual Findings

An opinion of nonparentage shall not be rendered on the basis of a single indirect exclusion or on the basis of an exclusion at a single DNA locus.

6.4.1.1 Results inconsistent with parentage testing detected at a single DNA locus shall be reported and shall be incorporated appropriately into statistical calculations.

RETENTION OF RECORDS – Chart 6.2A			
#	Standard #	Record to be Maintained	Minimum Retention Time (In Years)*
1	1.1.1.1	Laboratory director qualifications	5
2	1.1.1.2	Laboratory director or designee review of critical test results, worksheets that record interpretations and conclusions, and case reports.	5
3	1.2.2	Management review of effectiveness of the quality system	5
4	2.1	Current job descriptions	5
5	2.1.1	Training for personnel performing critical tasks affecting quality	5
6	2.1.1.1	Continuing education for employees performing critical steps	5
7	2.1.2	Competency evaluation of employees	5
8	2.1.2.1	Competency testing for employees performing critical steps	
9	2.1.3	Personnel records of employees. For those authorized to perform or review critical significant processing steps, maintain records of signatures, initials or identification codes for 10 years	5
10	3.2	Unique identification of critical equipment	5
11	3.3.1	Monitoring of critical equipment	5
12	4.1	Evaluation and participation in selection of suppliers	5
13	4.2.1	Review of agreements	5
14	4.3	Inspection of incoming materials and samples	5
15	5.1.1	Validation of new or changed processes and procedures	5
16	5.1.2	Participation in proficiency testing program	5
17	5.1.3	Review of quality control results for reagents, equipment, and methods	5
18	5.1.4.1	Reagents prepared by facility meet or exceed applicable criteria	5
19	5.1.4.1.1	Evaluation of reactivity and specificity of all critical reagents and/or controls prior to use	5
20	5.2.1	Informed consent from each tested person, legal guardian, or conservator. Include record in case file	5
21	5.2.1.1	Evidence of legal authority	5
22	5.2.2	Individual performing collection. Include record in	5

		case file	
23	5.2.4	<ol style="list-style-type: none"> 1. Name, relationship, date of birth, and the race/ethnic background of each parent/alleged parent. 2. Date of birth of the child. 3. Place and date of sample collection. 4. Printed name, signature, and contact information of the person collecting the sample, if applicable. 5. Printed name, signature, and contact information of the person verifying collection process, if different from the person collecting the sample. 6. A history of transfusion in the preceding 3 months, or hematopoietic progenitor cell transplantation. 7. Original or legible photocopies of one or both of the following items: <ol style="list-style-type: none"> a. Government photo identification. b. Photograph taken at the time of collection that is suitable for positive identification. 	5
24	5.3.4	Review of case by two people, including the laboratory director or designee	5
25	5.4.1	All records applying to serological testing, including red cell surface antigens, red cell enzyme and serum protein testing, immunoglobulin allotype testing, HLA antigen testing	5
26	5.4.2	All records pertaining to NAT testing, including RFLP, NAT for STR and AMFLP analysis, and NAT for nucleotide sequence determination or SNP analysis	5
27	5.4.2.3, #4	Control probe to ensure adequate target nucleic acid is available for analysis	5
28	5.4.2.3, #5	Control primer to evaluate amount of amplification	5
29	5.5.1	Technical supervisor and/or laboratory director review of calculations	5
30	6.1.3	Review and approval of new and revised documents prior to use	5
31	6.1.4	Annual review of policies, processes, and procedures	5
32	6.1.6	Archival of obsolete documents	5
33	6.3.1	Validation of computer system software, hardware, databases, and user-defined tables; fulfillment of life-cycle requirements for internally developed software; numerical designation of system versions, if	5

		applicable with inclusive dates of use; monitoring of data integrity for critical data elements	
34	7.1	Evaluation of nonconforming parentage test reports, materials, samples, and parentage testing services	5
35	7.2	Evaluation of nonconforming proficiency test results	5
36	8.2	Management of assessment results	5
37	9.1.4 9.2.3	Results of follow-up action to corrective and preventive actions	5
38	10.2	Monitoring of adherence to biological, chemical, and radiation safety standards and regulations	5
39	10.3	Discard of biological samples	5

*Applicable federal, state, or local law may supersede this time period.

6.4A - Requirements for Testing Reports	
A. Identifiers	
1	Date of collection for each sample
2	Name, address, and telephone number of the laboratory and its accession or case number, if assigned
3	Name of each person tested and his/her <u>alleged relationship to the child</u>
4	Racial/ethnic background(s) <u>used assigned</u> by the laboratory <u>for computations to the known and alleged parent(s)</u>
5	Phenotypes established for each person in each genetic system examined
6	The original signature of the laboratory director <u>or designee as defined by Standard 1.1.1.2</u>
7	The identity of any subcontracting laboratory(ies) and that portion of the report for which it bears responsibility <u>if applicable.</u>
B. Findings	
1	A statement <u>opinion</u> as to whether the alleged parentage (or other alleged relationship) can be excluded
2	IF:
	THEN:
	A statement of non-parentage is rendered
	The basis for the finding shall be provided

	There is a failure to exclude	<p>The report shall include the following information:</p> <ol style="list-style-type: none"> a. The individual parentage index for each genetic system reported b. The combined parentage index c. The probability of parentage expressed as a percentage. The prior probabilities used to calculate the probability of parentage shall be stated. d. <u>A statement that the calculation of the parentage index compares the tested individual(s) to an unrelated person of the same racial/ethnic background</u>
	Results are inconclusive or unusual	An explanation as to the nature of the problem
3	<p>For DNA tested:</p> <ol style="list-style-type: none"> a. Name of DNA locus tested as defined by the <u>relevant scientific literature Nomenclature Committee of the International Human Gene Mapping Workshop</u> b. Name of the restriction endonuclease used, if applicable c. The name of the probe used, if applicable, for RFLP testing d. Phenotypes <u>consistent with these Standards</u> consisting of size (in basepairs or kilobasepairs) of reported allelic fragment(s) for RFLP testing or repeat number (if known) for STR/AMFLP systems. 	
4	<u>Identification Reports containing the results of any supplemental test methods not covered by these Standards</u>	

7.0 INCIDENTS, ERRORS, AND ACCIDENTS AND NONCONFORMING PARENTAGE TESTING REPORTS, MATERIALS, SAMPLES AND PARENTAGE TESTING SERVICES

The laboratory shall have policies, processes, and procedures to ensure the capture, assessment, investigation, and monitoring of events that deviate ~~deviations from or of failures to meet specified~~ ~~accepted policies, processes, or procedures or that fail to meet the requirements of the laboratory, these~~ ~~Standards, or applicable laws and regulations~~ requirements. The responsibility for review and authority for the disposition of nonconforming parentage test reports, materials, samples, and parentage testing services shall be defined.



7.1 Nonconformances

Upon discovery, nonconforming parentage test reports, materials, samples, and parentage testing services shall be evaluated and their disposition determined.

7.1.1 Discrepant parentage test reports that do not conform to specified requirements shall be prevented from unintended distribution or use.

7.1.2 The laboratory shall have a process for the quarantine, retrieval, and recall of nonconforming parentage test reports, materials, and samples.

7.1.3 The laboratory shall investigate and resolve all discrepant test results or interpretations. [Moved from Chapter 9]

7.1.4 The laboratory shall have a process for identifying and notifying recipients of distributed nonconforming parentage test reports.

7.2 Nonconforming Proficiency Testing Results

When nonconforming proficiency testing results are obtained, the laboratory shall evaluate and take appropriate action in response to results with unacceptable grades or deviations from consensus assignments.

7.2.1 No more than one response graded as unacceptable shall be received per proficiency test.

7.2.2 Two consecutive misinterpretations of phenotypes shall warrant sanctions from the accrediting agency, and the laboratory shall send a letter of explanation to the accrediting agency.

8.0 ASSESSMENTS: INTERNAL AND EXTERNAL

The laboratory shall have a process to ensure that external assessments (inspections, surveys) are obtained at appropriately defined intervals and that internal assessments of operations and the quality system are scheduled and ~~conducted performed~~. ~~Compliance with review recommendations shall be monitored and shall promote continuous improvement. Results shall be reported to management, and records shall be maintained.~~

8.1 External Assessments

External assessments shall be scheduled at a minimum of every 2 years.

Self Assessments

~~The laboratory shall establish a self-assessment program that includes a review of case records, performance thresholds, variance reports, and appropriate corrective action (8.200).~~



8.2 Management of Assessment Results

8.2.1 *The results of internal and external assessments shall be reviewed by personnel having responsibility for the area being assessed.*

8.2.2 *Follow-up action shall verify the implementation and effectiveness of corrective and preventive action.*

8.2.3 *The results of internal and external assessments and associated corrective and preventive action shall be reviewed by executive management.*

9.0 PROCESS IMPROVEMENT THROUGH CORRECTIVE AND PREVENTIVE ACTION

The laboratory shall have policies, processes, and procedures ~~to ensure the use of defined methods for identification~~ for data collection, analysis, and follow-up of issues requiring corrective and preventive action.

9.1 Corrective Action

The laboratory shall have a process for corrective action that includes the following elements:

~~*Discrepant results shall be resolved and corrective action shall take place.*~~

9.1.1 *Documentation of incident, error, and accident reports; reports of nonconformances; and complaints. ~~The effective handling of error and accident reports.~~*


9.1.2 *Investigation of the cause of nonconformances relating to process, quality system and recording of the results of the investigation, parentage test reports, samples, materials, and parentage testing services.*

~~All nonconforming samples shall be identified. Records shall be maintained. (9.121)~~

~~The laboratory shall evaluate nonconforming samples and determine whether they are acceptable. (9.122)~~

[Standard 9.123 from 4th edition of *Standards* moved to Chapter 7.]

9.1.3 *Determination of the corrective action needed to eliminate the cause of nonconformances and incidents, errors, and accidents.*

and  **9.1.4** *Evaluation to ensure Assurance ~~-that corrective action is taken~~ that it is effective.*


9.2 Preventive Action

The laboratory shall have a process for preventive action that includes the following elements:

~~*Preventive action shall be taken in response to identified and potential nonconformities in equipment, procedures, and the quality system. Records shall be maintained.*~~

9.2.1 The review of appropriate sources of information, including assessment results, proficiency testing results, quality control records, and complaints, to detect and analyze potential causes of nonconformances.

9.2.2 Determination of steps needed to deal with any potential problems requiring preventive action.

 9.2.3 Initiation of preventive action and application of controls to ensure that it is effective.

10.0 FACILITIES AND SAFETY

The laboratory shall have policies, processes, and procedures to ensure the provision of safe and adequate environmental conditions in the workplace.

10.1 Safe Environment

The laboratory shall have a process ~~in operation programs~~ to minimize environmentally-related risks to the health and safety of employees, donors, volunteers, and persons who seek parentage testing. Programs shall meet local, state, and federal regulations, where applicable. Suitable quarters, environment, and equipment shall be available to maintain safe operations.



10.2 Biological, Chemical, and Radiation Safety

The laboratory shall have a process for monitoring adherence to ~~provide for~~ biological, chemical, and radiation safety standards and regulations, where applicable ~~and a system for monitoring training and compliance~~. Standard 2.1.1 applies.



10.3 Discard of Biological Samples ~~Infectious Agents~~

Biological samples shall be handled and discarded in a manner that minimizes the potential for human exposure to infectious agents.