COLLEGE of AMERICAN PATHOLOGISTS

Master

Histocompatibility Checklist

CAP Accreditation Program

College of American Pathologists
325 Waukegan Road
Northfield, IL 60093-2750
www.cap.org

09.22.2021

Disclaimer and Copyright Notice

On-site inspections are performed with the edition of the Checklists mailed to a facility at the completion of the application or reapplication process, not necessarily those currently posted on the website. The checklists undergo regular revision and a new edition may be published after the inspection materials are sent.

For questions about the use of the Checklists or Checklist interpretation, email accred@cap.org or call 800-323-4040 or 847-832-7000 (international customers, use country code 001).

The Checklists used for inspection by the College of American Pathologists' Accreditation Programs have been created by the CAP and are copyrighted works of the CAP. The CAP has authorized copying and use of the checklists by CAP inspectors in conducting laboratory inspections for the Council on Accreditation and by laboratories that are preparing for such inspections. Except as permitted by section 107 of the Copyright Act, 17 U.S.C. sec. 107, any other use of the Checklists constitutes infringement of the CAP's copyrights in the Checklists. The CAP will take appropriate legal action to protect these copyrights.

Histocompatibility Checklist

TABLE OF CONTENTS

ON-LINE CHECKLIST AVAILABILITY AND RESOURCES

Participants of the CAP accreditation programs may download the checklists from the CAP website (cap.org) by logging into e-*LAB* Solutions Suite. They are available in different checklist types and formatting options, including:

- Master — contains ALL of the requirements and instructions available in PDF, Word/XML or Excel formats
- Custom — customized based on the laboratory's activity (test) menu; available in PDF, Word/XML or Excel formats
- Changes Only — contains only those requirements with significant changes since the previous checklist edition in a track changes format to show the differences; in PDF version only. Requirements that have been moved or merged appear in a table at the end of the file.

A repository of questions and answers and other resources is also available in e-LAB Solutions Suite under Accreditation Resources, Checklist Requirement Q & A.

SUMMARY OF CHECKLIST EDITION CHANGES
Histocompatibility Checklist
09/22/2021 Edition

The information below includes a listing of checklist requirements with significant changes in the current edition and previous edition of this checklist. The list is separated into three categories:

1. New
2. Revised:
   - Modifications that may require a change in policy, procedure, or process for continued compliance; or
   - A change to the Phase
3. Deleted/Moved/Merged:
   - Deleted
   - Moved — Relocation of a requirement into a different checklist (requirements that have been resequenced within the same checklist are not listed)
   - Merged — The combining of similar requirements

NOTE: The requirements listed below are from the Master version of the checklist. The customized checklist version created for on-site inspections and self-evaluations may not list all of these requirements.

NEW Checklist Requirements
None

REVISED Checklist Requirements

Requirement	Effective Date
HSC.21275	09/22/2021
HSC.29885	09/22/2021
HSC.29957	09/22/2021
HSC.30013	06/04/2020
HSC.38060	09/22/2021
HSC.38097	09/22/2021
HSC.38845	09/22/2021
HSC.39415	09/22/2021
HSC.39430	09/22/2021

DELETED/MOVED/MERGED Checklist Requirements

Requirement	Effective Date
HSC.29917	06/03/2020

INTRODUCTION

This checklist is used in conjunction with the All Common (COM) and Laboratory General Checklists to inspect a histocompatibility laboratory section or department.

Histocompatibility inspectors must be pathologists, clinical scientists or medical technologists who are actively involved with or have extensive experience in the practice of histocompatibility testing, are knowledgeable about current CAP Checklist and CLIA requirements, and have completed CAP Inspector Training. Inspectors should, to the greatest extent possible, be peers of the laboratory being inspected.

***Laboratories not subject to US regulations:** Checklist requirements apply to all laboratories unless a specific disclaimer of exclusion is stated in the checklist. When the phrase "FDA-cleared/approved test (or assay)" is used within the checklist, it also applies to tests approved by an internationally recognized regulatory authority (eg, CE-marking).*

DEFINITION OF TERMS

Common, intermediate and well-documented (CIWD) alleles - Common alleles have frequencies of at least 1 in 10,000; intermediate alleles are found at frequencies less than 1 in 10,000 but at least 1 in 100,000; well-documented alleles have been observed five or more times in unrelated individuals but not at the common or intermediate levels.

High resolution typing - A high-resolution typing is defined as an allele or a set of alleles (G or P groups) that encode the same protein sequence for the region of the HLA molecule called the antigen binding site, with the exception of common, intermediate, or well-documented null alleles (CIWD) version 3.0.0, Hurley CL, et al. *HLA.* 2020;95:516-531), which need to be resolved. The high-resolution genotype must include only one unambiguously assigned genotype; however, alternative genotypes can be listed if they do not contain common or intermediate alleles (CIWD) version 3.0.0, Hurley CK, et al. *HLA.* 2020;95:516-531).

Low resolution typing - A low-resolution HLA genotype result provides sufficient information to identify serological splits or their equivalent. In some cases, this may require two-field genotyping results. A list of serological splits can be accessed at: http://hla.alleles.org/nomenclature/index.html.

PROFICIENCY TESTING

Inspector Instructions:

- Are proficiency testing samples tested with the same cut-offs for clinical HLA antibody determination as clinical specimens?
- Are proficiency testing samples for HLA antigens tested to the same level of resolution as clinical specimens?

- Select a representative clinical report from each service area. Compare the extent of reporting for the relevant proficiency testing sample.

HSC.10475 PT Extent of Testing **Phase II**

Proficiency testing specimens are tested to the same extent as clinical specimens.

NOTE: Proficiency testing samples must be tested using the most comprehensive testing algorithm or pathway applied to patient samples. For example, if a laboratory has a written procedure that calls for both low and high-resolution HLA analysis for a certain patient population, then all PT samples are tested to the highest resolution level.

Evidence of Compliance:

✓ Comparison of patient and proficiency testing work records demonstrating identification to the same extent

QUALITY MANAGEMENT

PROCEDURE MANUAL

Inspector Instructions:

- Representative sample of procedures for completeness. Current practice must match contents of procedures.

HSC.20200 Procedure Manual **Phase II**

The procedure manual contains specific instructions for test performance, preparation of reagents, control methods, specimen requirements, limitations of the method, and criteria for accepting/rejecting runs and reporting of results for each of the following procedures, as applicable:

1. **Lymphocyte isolation**
2. **HLA serologic typing**
3. **HLA molecular typing**
4. **Crossmatching-T cells**
5. **Crossmatching-B cells**
6. **Antibody screening and identification**
7. **Engraftment monitoring**
8. **ABO grouping**
9. **Complement titration**
10. **Environmental control**

SPECIMEN COLLECTION AND HANDLING

Inspector Instructions:

	• Sampling of histocompatibility specimen collection/handling/tracking retrieval policies and procedures • Evaluation records (specimen collection containers/anticoagulants) for preservation of sample integrity
	• What are the specimen acceptability criteria for each specimen type? • What is your course of action when you receive unacceptable/sub-optimal histocompatibility specimens? • How does your laboratory ensure preservation of antibody integrity in recipient sera?
	• Review records of unacceptable specimens and follow up. Determine if practice matches procedure.

HSC.20982 Specimen Collection Procedures Evaluation **Phase II**

The laboratory evaluates its specimen collection procedures to ensure that the anticoagulant/preservation medium in use does not contribute to analytic interference in the assays to be performed, and that it preserves sample integrity as necessary.

NOTE: This may be done through some combination of direct testing by the laboratory, review of the clinical literature, and evaluation of information from manufacturers. It does not mandate exhaustive testing by each laboratory.

Evidence of Compliance:

✓ Records of the evaluation of specimen collection procedures and anticoagulants in collection containers

HSC.20988 Specimen Integrity - Flow Cytometry **Phase II**

There is a written procedure to verify the integrity of specimens for flow cytometry.

NOTE: The yield of T lymphocytes from blood samples is affected by a number of factors. If specimens are not processed immediately after collection, the laboratory should verify that its anticoagulant, holding temperature and preparation method maintain specimen integrity. Selective loss of cell subpopulations and/or the presence of dead cells may lead to spurious results. Routine viability testing is not necessary on specimens of whole blood that are analyzed within 24 hours of drawing. Analyses on older samples are possible if the laboratory has verified the absence of statistical differences between the fresh and aged specimen phenotype fractions being evaluated.

REFERENCES

1) Clinical and Laboratory Standards Institute. *Enumeration of Immunologically Defined Cell Populations by Flow Cytometry; Approved Guideline.* 2nd ed. CLSI Document H42-A2. Clinical and Laboratory Standards Institute, Wayne, PA; 2007.

HSC.21050 Recipient Sera **Phase II**

The most appropriate recipient sera are employed for final crossmatching or final selection of donor.

NOTE: There must be a written policy defining an appropriate specimen to utilize in transplantation or final donor selection that takes into consideration the potential recipient's past pregnancies, past transplants, recent blood transfusions, and sensitization history. The specimens must have been properly handled and appropriately stored to preserve antibody integrity.

REFERENCES
1) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.
2) Foundation for the Accreditation of Cellular Therapy (FACT) and Joint Accreditation Committee ISCT and EBMT (JACIE). FACT-JACIE International Standards for Cellular Therapy Product Collection, Processing, and Administration. 7th Edition, March 2018.

HSC.21130 Specimen Retrieval **Phase II**

Procedures are adequate to ensure that patient specimens are easily retrievable.

REFERENCES
1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):7170 [42CFR493.1278(a)(2)]

RESULTS REPORTING

Inspector Instructions:

	• Sampling of patient reports for completeness, use of appropriate nomenclature, and review prior to release • Sampling of referral laboratory accreditation records
	• How are urgent results communicated?

HSC.21250 Patient Report Requirements **Phase II**

Patient results are reported in a legible, easy to interpret format that clearly indicates the test method and delineates the clinical implications of the results.

NOTE: For patient test results that include an interpretative analysis narrative or statement, the name of the individual(s) responsible for the interpretation must be included.

***REVISED** 09/22/2021*
HSC.21275 Final Report **Phase II**

The final report includes the following:

- **Summary of the methods used**
- **Loci tested**
- **Objective findings***
- **Limitations of the methods, when applicable**
- **Interpretation.**

NOTE: For donor registries, aggregate reports may be provided for a group of donors.

** For high resolution HLA typing, there is no need to list unresolved non-common, intermediate, or well-documented (CIWD version 3.0.0) alleles or G and P group alleles or codes if stated in the report, client agreement, or client request in writing.*

REFERENCES
1) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. August 17, 2020.
2) Hurley CK, et al. Common, intermediate and well-documented HLA alleles in world populations: CIWD version 3.0.0. *HLA.* 2020;95(6):516-531.

HSC.21277 Nomenclature — Phase II

The HLA antigen and allele assignments and their written designation conform to the current World Health Organization nomenclature for histocompatibility antigens.

*NOTE: For example: Phenotype is HLA-A1,2; B51,B44; Cw3; DR1,4; DR53; DQ4,8. Genotype is HLA-A*01:01, *02:01; HLA-B*51:07, B*44:03, HLA-C*03:01; HLA-DRB1*01:01, DRB1*04:01; DRB4*01:01; DQB1*04:01, DQB1*03:02. Haplotypes should not be assigned unless all haplotypes can be identified, uniquely, by pedigree analysis. For example, solid organ transplant typings are reported as antigens compatible with UNOS/OPTN requirements and hematopoietic progenitor cell transplant typings are reported at the allele level compatible with National Marrow Donor Program (NMDP) requirements.*

Any newly discovered antigens not yet assigned by WHO Committee must be designated in such a manner as not to be confused with existing established terminology. All genotype and phenotype designations must also conform to WHO Committee recommendations. The laboratory must maintain a list of antigens and alleles defined by the reagents used.

Evidence of Compliance:

✓ Appropriate antigen and allele assignments to support the transplant program

REFERENCES
1) http://www.ebi.ac.uk/imgt/hla
2) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):[42CFR493.1278(b)(4)]

HSC.21281 Accreditation of Referral Laboratories — Phase II

Outside referral laboratories are accredited by appropriate histocompatibility agencies. US laboratories are CLIA certified or meet equivalent requirements as determined by the Centers for Medicare and Medicaid Services (CMS).

NOTE: Laboratories that are members of the United Network for Organ Sharing (UNOS) may only refer histocompatibility testing to other laboratories that are OPTN-approved.

Refer to GEN.41350 for additional information on requirements for referral laboratory selection.

Evidence of Compliance:

✓ Records verifying referral laboratory certification/accreditation in histocompatibility

REFERENCES
1) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.

HSC.21287 Result Review — Phase II

All laboratory results (excluding reports from outside referral laboratories) have two levels of independent review, including review by the section director (technical supervisor) or designee prior to release.

NOTE: The initial review may be performed by validated automated analysis or by a qualified individual. The data output results must be reviewed by a qualified individual before release.

HSC.21295 Critical Reporting **Phase I**

The laboratory has defined critical reporting situations (eg, an unexpected positive crossmatch or development of a de novo donor-specific antibody) and has a written procedure for the communication of the critical report. There is a record of this communication.

RECORDS

The records listed below must be kept to the extent of services provided by the laboratory.

Inspector Instructions:

	• Record retention policy • Sampling of stored specimen inventory records/log • Sampling of transplant donor and recipient records • Verification of patient data policy (interval of review is defined) • Sampling of patient histocompatibility data review and verification • Sampling of policies and procedures for donor confidentiality
	• How does your laboratory resolve inter-laboratory HLA typing discrepancies? • How do you store results for comparison with subsequent reports?
	• Review all records of a sampling of patient and donor histocompatibility results and reports to ensure completion of all steps in the process from specimen requisitions to final disposition. Determine if records provide an adequate audit trail of all activities.

HSC.21316 Record and Material Retention - Histocompatibility **Phase II**

A copy of each final report, all records of results, reagent lots, gel images, *in situ* hybridization slides, and histograms used for interpretation and determination of test results are retained in compliance with existing laws.

REFERENCES
1) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.

HSC.21332 Stored Specimen Log **Phase II**

A log of all stored specimens is maintained to enable prompt retrieval for further testing.

Evidence of Compliance:

✓ Electronic or paper inventory log of stored specimens

RECIPIENT AND DONOR INFORMATION RECORDS

HSC.21350 Clinical Transplant Registries and Transplant Data Retention — Phase II

The institution participates in and retains records of patient and donor transplant information in the United Network for Organ Sharing (UNOS) Clinical Transplant Registry or its equivalent.

NOTE: The laboratory and/or transplant coordinator must retain records on transplant recipients, including a history of prior transfusion, pregnancy, and prior transplants as well as HLA antibody history, date of transplant(s) and outcome. In addition, there should be records of donor and recipient age, race, sex, ABO, and HLA types. The source of the donor organ or donor hematopoietic stem cells should be recorded. This information can be retained as part of an institutional registry.

REFERENCES

1) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.

HSC.21366 Record Review — Phase II

There are records of periodic review and verification of patient histocompatibility data.

NOTE: Histocompatibility tests performed for organ transplantation (HLA typing, HLA antibody sensitization, unacceptable antigens during prior transplants or sensitization, and any pretransplant screening results) must be reviewed and verified when patients are placed on organ waiting lists. Changes or additions to the waiting lists must be verified. These records must be readily available for review, and retained for at least two years.

REFERENCES

1) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.

HSC.21382 Discrepancy Resolution — Phase II

There is a written procedure to resolve HLA typing discrepancies within and between laboratories.

NOTE: There must be records of the steps taken to resolve discrepancies.

REFERENCES

1) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.
2) Foundation for the Accreditation of Cellular Therapy (FACT) and Joint Accreditation Committee ISCT and EBMT (JACIE). FACT-JACIE International Standards for Cellular Therapy Product Collection, Processing, and Administration. 7th Edition, March 2018.

HSC.21390 Donor Confidentiality — Phase II

There are written policies and procedures to ensure confidentiality of all donor records, including releasing or sharing donor information for clinical purposes.

NOTE: For example, if identifiable donor information will be shared with the recipient, appropriate donor informed consent must be obtained, donor information must be redacted, or other appropriate action taken.

Refer to the Laboratory General Checklist for specific requirements on patient privacy and patient data accessibility.

REFERENCES
1) Foundation for the Accreditation of Cellular Therapy (FACT) and Joint Accreditation Committee ISCT and EBMT (JACIE). FACT-JACIE International Standards for Cellular Therapy Product Collection, Processing, and Administration. 7th Edition, March 2018.

REAGENTS

Inspector Instructions:

	• Reagent inventory log • Sampling of procedures for reagent and patient sample storage and handling • Sampling of typing/screening tray records for completeness • Validation studies for modified reagents
	• What are your laboratory's criteria for mixing components from one lot number of reagent kit with components from another lot number of kit? • How do you ensure that all reagents are acceptable and in date? • How does your laboratory manage and control reagent inventory?

Additional requirements are in the REAGENTS section of the All Common Checklist.

HSC.21612 Reagent Tracking **Phase II**

There is a written procedure for the recording of the specific reagent lot numbers and shipments used for each assay.

REFERENCES
1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2004(Oct 1): 1038 [42CFR493.1256(a)]

HSC.21675 Reagent Kit Components **Phase II**

Combinations of reagents from different lots are checked against old reagent lots or with suitable reference material before or concurrently with being placed in service.

Evidence of Compliance:
✓ Written policy defining allowable exceptions for mixing kit components from different lots

HSC.21800 Specimen Storage Procedure **Phase II**

There are written procedures for optimal reagent and patient sample storage.

NOTE 1: Written procedures must include storage requirements for frozen lymphocytes, frozen lymphocyte trays, and sera.

NOTE 2: Monitored alarms and back-up storage plans must be specified as applicable.

Evidence of Compliance:
✓ Written procedures defining requirements for specimen/reagent storage with backup plan in case of equipment failure

REFERENCES
1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):[42CFR493.1278(a)(1)]
2) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.

HSC.21805 Histocompatibility Reagent Confirmation of Acceptability **Phase II**

New typing reagents are checked using suitable reference materials prior to use.

NOTE: Suitable materials for checking typing reagents include the use of previously typed cells or known archived DNA. Suitable materials for checking reagents for engraftment monitoring include the use of previously tested or archived admixtures.

Evidence of Compliance:

- ✓ Written procedure including criteria for acceptability of new reagents **AND**
- ✓ Records of acceptability studies for new reagents prior to use

REFERENCES

1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):[42CFR493.1278(a)(4)]

HSC.21810 Specimen Handling - Typing/Screening Trays **Phase II**

If typing trays and antibody screening trays are prepared locally, the records indicate source, bleeding date, donor, identification, and available volume for sera and a means of identifying, locating and collecting fresh donor cells.

REFERENCES

1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):7170 [42CFR493. 1278(a)(3)], 7171 [42CFR492. 1278(d)(3)]

HSC.21835 Modified Reagent Use **Phase II**

If reagents are used in a manner different than manufacturer's instructions, there are records of validation studies.

Evidence of Compliance:

- ✓ Validation study data

CONTROLS

Inspector Instructions:

	• Sampling of QC policies and procedures • Sampling of lymphocyte preparation viability checks • Sampling of QC records
	• Control material (labeling)
	• How do you determine when QC is unacceptable and corrective actions are needed? • What is your course of action when QC for compatibility testing is not acceptable?

DISCOVER

- Review a sampling of QC data over the previous two-year period. Select several occurrences in which QC is out of range and follow documentation to determine if the steps taken follow the laboratory procedure for corrective action

HSC.21850 Daily Controls **Phase II**

Positive and negative controls are assayed daily, and there are positive controls for specific cell types (T cells, B cells, etc.), where available.

NOTE: Positive and negative controls must be run with each test procedure where appropriate. This must include daily positive controls for specific cell type (T cells, B cells, etc.), as well as appropriate antibody isotypes as needed for each assay. This must also include one positive control serum that is historically reactive to all Class I and/or Class II positive cells at the same dilutional titer as appropriate for the methodology utilized.

Evidence of Compliance:

✓ Written QC procedure for each test describing the quality control materials used **AND**
✓ Records of control results

REFERENCES

1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):7168 [42CFR493. 1256(d)(3)(iii)]
2) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):[42CFR493.1278(b)(6)] and [42CFR493.1278(c)]

HSC.21950 Viability Checks **Phase II**

Viability checks on lymphocyte preparations are performed by recording negative control results or by performing and recording a separate test each time they are used.

NOTE: For cytotoxicity procedures, cell viability after initial incubation should be greater than 80% in the negative control well.

Evidence of Compliance:

✓ Written procedure for evaluating viability of lymphocyte preparations

REFERENCES

1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):[42CFR493.1278(b)(6)(ii)]

HSC.22070 Compatibility Testing Controls **Phase II**

The laboratory includes control material for each phase of compatibility testing.

NOTE: Results of patient testing must not be reported until control values are reviewed and found acceptable.

Evidence of Compliance:

✓ Written QC procedure for compatibility testing that describes the quality control material used

REFERENCES

1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):7171 [42CFR493. 1278(c) and (e)(3)]

HSC.22140 QC Handling **Phase II**

Control specimens are tested in the same manner and by the same personnel as patient samples.

NOTE: QC specimens must be analyzed by personnel who routinely perform patient testing - this does not imply that each operator must perform QC daily, so long as each instrument and/or test system has QC performed at required frequencies, and all analysts participate in QC on a regular basis. To the extent possible, all steps of the testing process must be controlled.

Evidence of Compliance:

- ✓ Records reflecting that QC is run by the same personnel performing patient testing

REFERENCES

1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):7166 [42CFR493. 1256(d)(8)]

HSC.22150 Alternative Control Procedures — Phase II

If the laboratory performs test procedures for which control materials are not commercially available, there are written procedures for an alternative mechanism to detect immediate errors and monitor test system performance over time. The performance of alternative control procedures must be recorded.

NOTE: "Performance" includes elements of accuracy, precision, and clinical discriminating power. Examples of alternative procedures may include split sample testing with another method or with another laboratory, the testing of previously tested patient specimens in duplicate, testing of patient specimens in duplicate, or other defined processes approved by the laboratory director.

Evidence of Compliance:

- ✓ Written procedures for alternative quality control **AND**
- ✓ Records of alternative control procedures

REFERENCES

1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register*. 2003(Jan 24): [42CFR493.1256(h)].

HSC.22160 QC Confirmation of Acceptability — Phase II

The results of controls are reviewed for acceptability before reporting results.

NOTE: If the positive and negative controls do not give the expected outcome, the results are not reportable. The negative control serum is one that historically has been negative with all tested cells. The negative control may also originate from a non-sensitized male whose serum has been shown to be totally negative for cell death in cytotoxicity systems.

Evidence of Compliance:

- ✓ Written policy that controls are reviewed and acceptable prior to reporting patient results **AND**
- ✓ Records of control result approval

REFERENCES

1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):7166 [42CFR493. 1256(f)]

HSC.22170 QC Corrective Action — Phase II

There is a record of corrective action taken when control results exceed defined acceptability limits.

NOTE: Patient/client test results obtained in an analytically unacceptable test run or since the last acceptable test run must be re-evaluated to determine if there is a significant clinical difference in patient/client results. Re-evaluation may or may not include re-testing patient samples, depending on the circumstances.

Even if patient samples are no longer available, test results can be re-evaluated to search for evidence of an out-of-control condition that might have affected patient results. For example,

evaluation could include comparison of patient means for the run in question to historical patient means, and/or review of selected patient results against previous results to see if there are consistent biases (all results higher or lower currently than previously) for the test(s) in question.

REFERENCES

1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 1992(Feb 28):7176 [42CFR493.1445(e)]

HSC.22190 Monthly QC Review **Phase II**

Quality control data are reviewed and assessed at least monthly by the laboratory director or designee.

NOTE: QC data include control results of routine procedures and the reactivity of reagents. The review of quality control data must be recorded and include follow-up for outliers, trends, or omissions that were not previously addressed.

The QC data for tests performed less frequently than once per month should be reviewed when the tests are performed.

Evidence of Compliance:

✓ Records of QC review including follow-up for outliers, trends, or omissions

INSTRUMENTS AND EQUIPMENT

The checklist requirements in this section should be used in conjunction with the requirements in the All Common Checklist relating to instruments and equipment.

TEMPERATURE-DEPENDENT EQUIPMENT

Inspector Instructions:

	• Sampling of thermocycler monitoring logs • Sampling of alarm system checks • Sampling of LN2 monitoring records
	• What back-up options are available in the event of an electrical power failure? • How is the storage unit alarm system monitored? How was the response time validated? • How does the laboratory ensure the individual wells of the thermocycler are maintaining accurate temperature?

HSC.22531 Alarm System **Phase II**

There is an audible alarm for each sample or reagent storage unit, and the alarm is monitored 24 hours per day (in laboratory or remotely).

NOTE: All storage units must have an audible alarm with continuous monitoring (in laboratory or remote). The laboratory must be able to demonstrate how this system works, and that there is a process to ensure a timely response to an alarm.

REFERENCES

1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):[42CFR493.1278(a)(1)]

HSC.22562 Alarm System Checks **Phase II**

Alarm systems are checked at specified periodic intervals and results recorded.

NOTE: The alarm system must be checked at specified periodic intervals to ensure proper function.

HSC.22593 Power Failure Back-up **Phase II**

The alarms continue to function if the power is interrupted.

NOTE: Alarm systems must have a source of power separate from the house current, in order to allow proper monitoring during power failures. This can be accomplished by a separate circuit, power failure alarm, or battery power.

HSC.22625 Cell Freezers **Phase II**

Cell freezers that use liquid nitrogen are monitored.

NOTE: The system must ensure that an adequate supply of liquid nitrogen is present to maintain optimal cell storage temperature.

Evidence of Compliance:
- ✓ Written procedure defining method for monitoring liquid nitrogen (LN2) levels **AND**
- ✓ Records of monitoring of LN2 levels at defined frequency

HSC.22775 Thermocycler Temperature Checks **Phase II**

Individual wells (or a representative sample thereof) of thermocyclers are checked for temperature accuracy before being placed in service and every six months thereafter.

NOTE: If it is not physically possible to check individual wells, a downstream measure of well-temperature accuracy (such as productivity of amplification) must be substituted to functionally meet this requirement. On thermocycler models where such is possible, individual wells must be checked for temperature accuracy before being placed in service and every six months thereafter.

Evidence of Compliance:
- ✓ Written procedure for verification of thermocycler accuracy **AND**
- ✓ Records of thermocycler verification

REFERENCES

1) Saunders GC, *et al*. Interlaboratory study on thermal cycler performance in controlled PCR and random amplified polymorphic DNA analyses. *Clin Chem.* 2001;47:47-55

COLORIMETERS, SPECTROPHOTOMETERS, AND FLUOROMETERS

The following requirements apply to stand-alone instruments; they are not applicable to instruments embedded in automated equipment for which the manufacturer's instructions must be followed.

Inspector Instructions:

	• Spectrophotometer policy or procedure • Sampling of manufacturer required system checks
	• Filters (clean, not scratched or deteriorated)

HSC.23137 Absorbance/Linearity **Phase II**

Absorbance and/or linearity fluorescence is checked and recorded at least annually or as often as specified by the manufacturer, with filters or standard solutions.

Evidence of Compliance:
- ✓ Records of absorbance and linearity checks at required frequency

HSC.23324 Filter Photometers **Phase II**

Filters (filter photometers) are checked at least annually to ensure they are in good condition (eg, clean, free of scratches).

Evidence of Compliance:
- ✓ Records of filter checks at defined frequency

HSC.23511 Spectrophotometer Checks **Phase II**

Spectrophotometer (including ELISA plate readers) wavelength calibration, absorbance and linearity are checked at least annually (or as often as specified by the manufacturer), with appropriate solutions, filters or emission line source lamps, and the results recorded.

NOTE: Some spectrophotometer designs, eg, diode array, have no moving parts that can alter wavelength accuracy and do not require routine verification. The manufacturer's instructions must be followed.

Evidence of Compliance:
- ✓ Records of spectrophotometer checks at required frequency

HSC.23698 Stray Light **Phase II**

Stray light is checked at least annually with extinction filters or appropriate solutions, if required by the manufacturer.

Evidence of Compliance:
- ✓ Records of stray light checks, as applicable

FUME HOODS AND BIOLOGICAL SAFETY CABINETS

Inspector Instructions:

	• Biological safety cabinet certification records
	• Fume hood/chemical filtration unit (available)

HSC.24446 Fume Hood/Chemical Filtration Unit **Phase II**

A properly functioning fume hood (or chemical filtration unit) is available for any procedures using volatile chemicals.

HSC.24633 Biological Safety Cabinet **Phase II**

A biological safety cabinet (or hood) is available, when appropriate, and is certified at least annually to ensure that filters function properly and that airflow rates meet specifications.

Evidence of Compliance:

✓ Maintenance schedule of BSC function checks **AND**
✓ Records of testing and certification

REFERENCES
1) Department of Health and Human Services. *Biosafety in Microbiological and Biomedical Laboratories.* 5th ed. Washington Publishing No. (CDC) 21-1112; December 2009, DC: Publishing No. (CDC) 21-1112, December 2009 HHS.

ELECTROPHORESIS EQUIPMENT

Inspector Instructions:

	• Sampling of voltage check records
	• Electrophoresis equipment (clean, properly maintained)

HSC.25007 Electrophoresis Equipment **Phase II**

All electrophoretic apparatus in the laboratory is clean and properly maintained (electrodes and buffer tank intact, power supply electrodes fit snugly, no build-up of dried buffer).

HSC.25194 Voltage Reading **Phase II**

The displayed voltage reading is confirmed at least annually by a voltmeter or other suitable means for all power supplies in the laboratory.

Evidence of Compliance:

✓ Records of annual voltage checks

PROCEDURES AND TEST SYSTEMS

Inspector Instructions:

- If problems are identified during the review of the procedures and test systems, or when asking questions, further evaluate the laboratory's responses, corrective actions and resolutions
- Select a representative assay and follow the entire process from specimen receipt to final result reporting

LYMPHOCYTE ISOLATION

Inspector Instructions:

- Lymphocyte isolation policies and procedures

HSC.27438 Lymphocyte Source **Phase II**

The source of the lymphocytes is recorded.

NOTE: These may include blood, bone marrow, lymph nodes, spleen, or cultured cells.

SEROLOGICAL PROCEDURES

GENERAL

Inspector Instructions:

- Sampling of complement reagent validation records

- How does your laboratory ensure cell death is appropriately measured?

HSC.27625 Scoring System **Phase II**

There is a scoring system in place for measuring cell death in cytotoxicity tests.

NOTE: There must be established limits for defining positive and negative results by approximate percentage of cell death.

HSC.27812 QC - Complement **Phase II**

Each lot, batch and/or shipment of complement is checked for effectiveness before or during use for each specific target cell and each test method.

NOTE: Each lot, batch and/or shipment of complement must be evaluated to determine that it can mediate cytotoxicity when a specific antibody is present, and is not cytotoxic in the absence of a specific antibody. For each specific target cell (T cell, B cell, monocyte, etc.), complement cytotoxicity studies must be performed to determine optimal dilution for each type of cell tested by cytotoxicity. Two HLA antibodies should have variable antibody strengths when utilized in complement testing against 2 different known antigen-containing cells that are reactive to the antibodies. Alternatively, one antibody may be utilized at variable dilutions for complement testing.

Evidence of Compliance:

- ✓ Written procedure for the validation of new complement reagents **AND**
- ✓ Records of validation of complement reagents

REFERENCES
1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):7166 [42CFR493. 1256(e)]
2) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):[42CFR493.1278(a)(4)]

HLA CLASS I AND II ANTIGEN TYPING

Inspector Instructions:

- Sampling of HLA Class I & Class II Ag typing policies and procedures. Procedures should specify the level of resolution of HLA typing required for each tissue or organ transplanted.
- Sampling of HLA typing of solid organ and hematopoietic progenitor cell transplantation policies and procedures
- List of antigens defined by reagents used
- Sampling of typing reagent validation records
- Sampling of typing tray QC records

- How does your laboratory select target cells to ensure the detection of WHO recognized antigens?
- What is your laboratory's course of action when a donor cannot be reliably HLA typed?

HSC.28186 Serologic Typing - Class I **Phase II**

Target cells are defined for serological determination of HLA Class I antigens, and selected to permit typing the antigens officially recognized by the WHO Committee for which reagents are readily available.

NOTE: HLA Typing for all hematopoietic progenitor cell donors and recipients, and deceased organ donors must be performed by molecular methods. Serological determination of HLA Class I antigens should be performed on T cells or mononuclear cell preparations. Local serological typing reagents must be supported by appropriate documentation of HLA specificity, using cells of known HLA types. The test must detect WHO recognized specificities.

REFERENCES
1) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.
2) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):[42CFR493.1278(b)] and [42CFR493.1278(5)(iii)]

HSC.28373 Serologic Typing - Class II **Phase II**

The methodology for serological Class II antigen typing defines the proportion of B-cells needed for optimal testing, and the specificities that are officially recognized by the WHO Committee and for which reagents are readily available.

NOTE: The method should produce at least 80% B-cell enriched preparations. Documentation of B cell enrichment may not be necessary when procedural techniques already distinguish T- and B-lymphocytes, or when well-characterized antibodies are used that can only discriminate and identify Class II antigens. HLA typing for all hematopoietic progenitor cell donors and recipients, and deceased organ donors must be performed by molecular methods.

REFERENCES
1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):[42CFR493.1278(b)] and [42CFR493.1278(5)(iii)]
2) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.

HSC.28560 Class I Antigen Defined **Phase II**

The minimum number of Class I antisera used are defined by the laboratory.

Evidence of Compliance:
- ✓ Written procedure for performing serologic determination of HLA Class I

HSC.28747 Class II Antigen Defined **Phase II**

The minimum number of Class II antisera used is defined by the laboratory.

Evidence of Compliance:
- ✓ Written procedure for performing serologic determination of HLA Class II

HSC.28934 Typing Trays **Phase I**

The typing trays used for disease association testing permit characterization of at least those antigens accepted by the World Health Organization (WHO) for which sera are readily available.

REFERENCES
1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):7170 [42CFR493. 1278(a) through (c)]

HSC.28996 Controls - Typing Trays **Phase II**

Each typing tray contains both a positive and a negative control.

NOTE: The positive control must be known to react with all cells expressing the class of antigens being tested at a titer comparable with the typing reagents. Each typing tray must also contain one negative control. Cell viability in the negative control must be sufficient to accurately interpret the results. The use of sufficiently discriminatory positive and negative controls also applies to assays in which cell viability is not required.

REFERENCES
1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):[42CFR493.1278(a)(4)]

CYTOTOXICITY CROSSMATCH

Inspector Instructions:

- Sampling of cytotoxicity crossmatch policies and procedures including method sensitivity

HSC.29308 HLA Crossmatch Sensitivity — Phase II

The method used in the HLA crossmatch procedure is more sensitive than the basic complement dependent micro-lymphocytotoxicity crossmatch and is able to distinguish between reactions with T and B lymphocytes, such that crossmatches with donor T-lymphocytes identify Class I anti-HLA antibodies and crossmatches with donor B-lymphocytes identify Class I and Class II anti-HLA antibodies.

Evidence of Compliance:
- ✓ Records of method sensitivity **AND**
- ✓ Peer-reviewed published literature on method sensitivity

REFERENCES
1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):7170 [42CFR493.1278(e)(1)]

HSC.29495 Crossmatch — Phase II

The crossmatch procedure defines the patient sera and donor cells utilized for final crossmatch testing.

NOTE: Cellular targets for transplant crossmatches must include donor T-cells, and may include donor B-cells when appropriate.

REFERENCES
1) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.
2) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):7170 [42CFR493.1278(e)(2)]

HSC.29682 Specimen Handling — Phase II

Patient samples for crossmatch testing are used undiluted, and kept frozen for a defined time post-transplantation.

Evidence of Compliance:
- ✓ Written procedure for cytotoxicity crossmatch

REFERENCES
1) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.

HSC.29869 Final Crossmatch Results Availability **Phase II**

There is a written policy that defines when results of a final crossmatch are available before transplantation for renal transplant patients and for presensitized extrarenal transplant patients.

NOTE: Laboratories supporting solid organ transplants must be capable of performing prospective crossmatches and must have a written policy describing in what situations pre- or post-transplant crossmatching is performed for all types of solid organ transplants. Results of the final crossmatch must be available before a kidney transplant is performed. The policy for presensitized extrarenal transplant patients must describe if and when crossmatches are performed. Crossmatches may be physical or virtual crossmatches as defined in the policy.

REFERENCES
1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 1992(Feb 28):7170 [42CFR493.1278(f)]
2) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.

RED CELL TYPING

Inspector Instructions:

READ	• Sampling of blood type/antibody screen policies and procedures • Sampling of current typing sera/reagent package inserts, for consistency with written procedures • Sampling of typing sera/reagent cell reactivity/anti-D QC records • Sampling of patient records with forward and reverse grouping • Record retention policy • Sampling of historical record checks
DISCOVER	• If there has been an instance where the ABO and Rh typing results were not in agreement with the patient's historical record, further evaluate the laboratory's responses, corrective actions and resolutions

HSC.29877 Reagent Handling - Red Cell Typing Reagents **Phase II**

Typing sera and reagent cells are used according to the manufacturer's instructions; or, if alternative procedures are used, validation records confirm that they perform as intended.

NOTE: Testing methods used for ABO, Rh and antibody screening that are different from the manufacturer's instructions, are acceptable provided they are not prohibited by the manufacturer, have been demonstrated to be satisfactory, or, for laboratories subject to US regulations, have been approved by the Centers for Biologics Evaluation and Research (CBER).

Evidence of Compliance:

✓ Written procedure consistent with manufacturer's instructions

REFERENCES
1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):[42CFR493.1271(a)(1)]

***REVISED** 09/22/2021*

HSC.29885 Package Inserts/Manufacturer's Instructions - Red Cell Typing **Phase II**

Current package inserts/manufacturer's instructions are available for the red cell typing reagents used by the laboratory.

NOTE: The laboratory must have a procedure that assures that:

- *The most current manufacturer's instructions/package inserts are in use*
- *The relevant procedures are updated when changes to the instructions occur.*

Although it is not required to retain discontinued instructions, the laboratory must have a process to obtain expired package inserts from the manufacturer, if requested.

Evidence of Compliance:

✓ Written procedure consistent with manufacturer's instructions **OR** records of validation study if an alternative procedure is used

HSC.29893 Red Cell Typing **Phase II**

There is a test of each patient's blood sample with anti-A, anti-B, and anti-D, and serum/plasma is tested using A1 and B reagent red cells.

NOTE: The ABO and the Rh type of the patient's red blood cells must be determined by an appropriate test procedure. Tests on each sample must include forward and reverse grouping. Discrepancies between cell and serum groups must be resolved before ABO group is assigned.

Evidence of Compliance:

✓ Written procedure for ABO/Rh typing **AND**

✓ Logs or computer records with forward and reverse grouping

HSC.29901 A1 Red Cell Subgrouping **Phase II**

There is evidence of the specificity of A1 subgroup testing in the ABO system to distinguish A1 from other red cell subgroups.

NOTE: If the organ donor has been transfused with red blood cells in the past three months, ABO subgroup typing must be performed on a pretransfusion sample. This is due to the possibility of misinterpretation of ABO subgroup typing.

HSC.29909 Antisera/Reagent Red Cell QC **Phase II**

There are records of acceptable reactivity and specificity of typing sera and reagent red cells on each day of use, including a check against known positive and negative cells or antisera, or manufacturer's instructions for daily quality control are followed.

NOTE: Unless manufacturer's instructions state otherwise, the following apply:

- *Typing reagents, including antisera (eg, anti-D, anti-K, anti-Fy(a)) and reagent red cells must be checked for reactivity and specificity on each day of use. Typing antisera must be checked with known positive and negative cells; reagent red cells must be checked with known positive and negative antisera.*
- *Each cell used for antibody screening must be checked each day of use for reactivity of at least one antigen using antisera of 1+ or greater avidity.*
- *Anti-IgG reactivity of antiglobulin reagents may be checked during antibody screening and crossmatching.*

This checklist requirement can be satisfied by testing one vial of each reagent lot each day of testing.

REFERENCES
1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 1992(Feb 28):7171 [42CFR493.1256]

HSC.29925 Historical Record Check - Red Cell Typing **Phase II**

ABO, Rh, and antibody screen test results are compared against the same tests performed previously to detect discrepancies.

Evidence of Compliance:
- ✓ Written procedure for the historical record check for ABO/Rh/antibody screen results **AND**
- ✓ Records of historical result comparisons

HSC.29941 Results Reporting - ABO Antibody Titers **Phase I**

There are written procedures for the performance and interpretation of ABO antibody titers.

HSC.29949 Anti-D Controls **Phase II**

Appropriate control(s) are used for anti-D testing.

NOTE: If an anti-D reagent contains a potentiating diluent, the appropriate control is the diluent alone.

Evidence of Compliance:
- ✓ Written procedures defining controls used for anti-D testing consistent with manufacturer's instructions **AND**
- ✓ Records of anti-D QC

FLOW CYTOMETRY

INSTRUMENTATION AND PHENOTYPING

Inspector Instructions:

	• Sampling of flow cytometry policies and procedures • Sampling of QC policies and procedures (includes acceptable control type/frequency for each flow cytometric application) • Sampling of QC records • Sampling of optical alignment/laser output checks
	• How does your laboratory monitor instrument reproducibility? • How does your laboratory ensure each fluorochrome is appropriately calibrated? • How does your laboratory determine appropriate color compensation settings? • How does your laboratory ensure nucleic acid dye specificity?

*****REVISED** 09/22/2021***

HSC.29957 QC - Quantitative Assays **Phase II**

For quantitative assays (eg, CD4+, CD34+ cell concentrations), at least two levels of positive cellular controls are analyzed each day of patient testing or after an instrument

restart to verify the performance of reagents, preparation methods, staining procedures, and the instrument.

NOTE: One of the levels of these controls should be at (or near) clinical decision levels (eg, low CD34).Control testing is not necessary on days when testing is not performed.

Evidence of Compliance:

- ✓ Written procedure defining QC requirements for each test **OR** records of validation of an alternate/equivalent procedure when commercial controls are not available **AND**
- ✓ Records of QC results

HSC.29965 Optical Alignment — Phase II

There are written procedures for monitoring of optical alignment (where applicable) and instrument reproducibility on each day of use, and records of this monitoring.

NOTE: Instrument performance must be monitored under the same conditions used to run test samples.

REFERENCES

1) Clinical and Laboratory Standards Institute (CLSI). *Clinical Flow Cytometric Analysis of Neoplastic Hematolymphoid Cells; Approved Guideline—Second Edition*. CLSI document H43-A2 (ISBN 1-56238-635-2). Clinical and Laboratory Standards Institute, 940 West Valley Road, Suite 1400, Wayne, Pennsylvania 19087-1898 USA, 2007

HSC.29973 Fluorochrome Standards — Phase II

Appropriate standards for each fluorochrome, (eg, fluorescent beads), are run each day that the instrument is used as part of the calibration process; and the results are recorded for quality control purposes.

NOTE: These steps are necessary to optimize the flow system and the optics of the instrument.

Evidence of Compliance:

- ✓ Written procedure for calibration with fluorochrome standards

REFERENCES

1) Clinical and Laboratory Standards Institute (CLSI). *Enumeration of Immunologically Defined Cell Populations by Flow Cytometry; Approved Guideline—Second Edition*. CLSI document H42-A2 (ISBN 1-56238-640-9). Clinical and Laboratory Standards Institute, 940 West Valley Road, Suite 1400, Wayne, Pennsylvania 19087-1898 USA, 2007

HSC.29981 Color Compensation Settings — Phase II

Procedures are established for determining appropriate color compensation settings.

NOTE: For two or more color analysis there must be a procedure to ensure that cells co-labeled with more than one fluorescent reagent can be accurately distinguished from cells labeled only with one reagent. Cells stained with mutually exclusive antibodies bearing the relevant fluorochromes are the proper reference material for establishing appropriate compensation settings.

REFERENCES

1) Clinical and Laboratory Standards Institute (CLSI). *Clinical Flow Cytometric Analysis of Neoplastic Hematolymphoid Cells; Approved Guideline—Second Edition*. CLSI document H43-A2 (ISBN 1-56238-635-2). Clinical and Laboratory Standards Institute, 940 West Valley Road, Suite 1400, Wayne, Pennsylvania 19087-1898 USA, 2007

HSC.29989 Laser Performance — Phase I

For laser instruments, there are procedures in place to ensure acceptable and constant laser performance.

NOTE: For some instruments, current is a better gauge of laser performance than is power output, which may be relatively constant.

HSC.29997 Gating Techniques **Phase II**

Appropriate gating techniques are used to select the cell population for analysis.

NOTE: This may involve a combination of light scatter and/or fluorescence measurements. This is particularly important if the cell samples have a low lymphocyte count and/or a relatively high monocyte-granulocyte count. Lymphocyte gates may be validated using linear forward angle light scatter and 90-degree side scatter, and/or by using monoclonal antibodies to markers, such as CD45 and CD14.

REFERENCES

1) Clinical and Laboratory Standards Institute. *Enumeration of Immunologically Defined Cell Populations by Flow Cytometry; Approved Guideline.* 2nd ed. CLSI Document H42-A2. Clinical and Laboratory Standards Institute, Wayne, PA; 2007.

HSC.30005 Markers/Cursors **Phase II**

There is a procedure to set markers (cursors) to distinguish fluorescence negative and fluorescence positive cell populations.

NOTE: Each laboratory must have a set of objective criteria to define the appropriate placement of markers (cursors) to delineate the population of interest. Isotypic controls may not be necessary in all cases, and cursor settings for the isotype control may not be appropriate for all markers. Cursor settings must be determined based on the fluorescence patterns from the negative and positive populations for CD3, CD4, and CD8.

REFERENCES

1) National Institute of Allergy and Infectious Diseases/Division of AIDS flow cytometry guidelines, sec 3.09B and 5.03A
2) Sreenan JJ, *et al*. The use of isotypic control antibodies in the analysis of CD3+ and CD3+, CD4+ lymphocyte subsets by flow cytometry. Are they really necessary? *Arch Pathol Lab Med.* 1997;121:118-121

*****REVISED** 06/04/2020***

HSC.30013 Cellular Viability **Phase II**

There is a policy for determining when the percentage of viable cells in each test specimen should be measured.

NOTE: Selective loss of cell subpopulations and/or the presence of dead cells may lead to spurious results. This does not mean that all specimens with low viability must be rejected. Finding an abnormal population in a specimen with poor viability may be valuable but the failure to find an abnormality should be interpreted with caution. If specimen viability is below the established laboratory minimum, test results may not be reliable and this should be noted in the test report. Routine viability testing may not be necessary. However, viability testing of specimens with a high risk of loss of viability, such as disaggregated lymph node specimens, is required.

REFERENCES

1) Clinical and Laboratory Standards Institute (CLSI). *Clinical Flow Cytometric Analysis of Neoplastic Hematolymphoid Cells; Approved Guideline—Second Edition*. CLSI document H43-A2 (ISBN 1-56238-635-2). Clinical and Laboratory Standards Institute, 940 West Valley Road, Suite 1400, Wayne, Pennsylvania 19087-1898 USA, 2007

HSC.30021 Non-Specific Binding **Phase II**

There are written procedures to ensure that immunoglobulin binding is specific.

NOTE: Many cell types will bind serum immunoglobulin nonspecifically via Fc receptors, and steps may have to be taken to ensure that immunoglobulin staining detected by flow cytometry is specific rather than non-specific.

Evidence of Compliance:

✓ Written procedure defining method to ensure specific binding

REFERENCES

1) Clinical and Laboratory Standards Institute (CLSI). *Clinical Flow Cytometric Analysis of Neoplastic Hematolymphoid Cells; Approved Guideline—Second Edition*. CLSI document H43-A2 (ISBN 1-56238-635-2). Clinical and Laboratory Standards Institute, 940 West Valley Road, Suite 1400, Wayne, Pennsylvania 19087-1898 USA, 2007

HSC.30029 Cell Population Distinction **Phase II**

There is a procedure to distinguish fluorescence-negative and fluorescence-positive cell populations.

NOTE: This does not imply that a separate negative control sample must be run. It is possible to coordinate panels of monoclonal antibodies to compare the binding of monoclonal antibodies of the same subclass that typically have mutually exclusive patterns of reactivity of subsets of hematopoietic cells. In this way, test antibodies may also double as control reagents.

REFERENCES

1) Clinical and Laboratory Standards Institute (CLSI). *Clinical Flow Cytometric Analysis of Neoplastic Hematolymphoid Cells; Approved Guideline—Second Edition*. CLSI document H43-A2 (ISBN 1-56238-635-2). Clinical and Laboratory Standards Institute, 940 West Valley Road, Suite 1400, Wayne, Pennsylvania 19087-1898 USA, 2007

HSC.30037 Procedure Manual **Phase II**

The staining and analytical procedures described in the procedure manual are based upon established methodology (reference cited).

NOTE: Many different variables need to be controlled to ensure proper stoichiometry of dye binding to DNA. Therefore, it is essential that procedures adopted by a laboratory are based on published work.

HSC.30045 Specimen Treatment **Phase II**

Specimen treatment with nucleic acid dye includes treatment with RNAse if the dye is not specific for DNA.

NOTE: Certain dyes used to stain fixed cells, (eg, ethidium and propidium iodide) bind to RNA. Prior treatment with RNAse eliminates artifactual broadening of the DNA content distributions that would result from fluorescence of complexes of the dye with RNA.

Evidence of Compliance:

✓ Written procedure for specimen treatment with RNAse

REFERENCES

1) Shapiro HA. Practical flow cytometry. New York, NY: Alan R. Liss, 1985

FLOW CYTOMETRY CROSSMATCH

Inspector Instructions:

READ	• Sampling of flow cytometry crossmatch policies and procedures • Sampling of QC policies and procedures • Sampling of QC records • Sampling of positive cutoff validation records
ASK	• How has your laboratory established the cutoff for positive crossmatch results? • Are cutoffs for crossmatches reviewed with the clinical transplant service? • Have the cutoffs been correlated with signal strength or other measure of antibody concentration in the HLA antibody screen and detection methods used?

- How does your laboratory ensure separation of Class I & Class II antibodies?

HSC.30056 Crossmatch **Phase II**

The flow cytometry crossmatch identifies antibodies to T and B-cells.

NOTE: Two or multiple color techniques must be used to identify antibodies to T cells. Antibodies to B cells and other target cells must also be identified properly.

HSC.30243 IgG Antibody Identification **Phase II**

IgG antibodies are identified by appropriately labeled heavy chain-specific F(ab')2 reagents.

HSC.30430 Sensitivity **Phase II**

There is a record of the number of cells and volume of serum used for optimal sensitivity.

HSC.30617 Negative Control - Normal Human Serum **Phase II**

Normal human serum with demonstrated lack of reactivity against any potential target cell is used as a negative control.

Evidence of Compliance:
- ✓ Written procedure describing controls used for flow cytometry crossmatch **AND**
- ✓ Records of control results

HSC.30804 Positive Control - Diluted Human Serum **Phase II**

The positive control is an appropriately diluted human serum containing suitable HLA antibodies of appropriate immunoglobulin class known to react with lymphocytes from all donors.

Evidence of Compliance:
- ✓ Written procedure describing controls used for flow cytometry crossmatch **AND**
- ✓ Records of control results

HSC.30991 Antibody Reagents **Phase II**

The antibody reagents (anti IgG, IgM, IgA, etc.) are used at a selected dilution for optimal sensitivity and class specificity.

Evidence of Compliance:
- ✓ Written procedure describing controls used for flow cytometry crossmatch

HSC.31178 Positive Crossmatch Results Cut-off **Phase II**

The cut-off for positive crossmatch results is determined by testing an appropriate number of sera from non-alloimmunized individuals and established for all pertinent target cells (T-cells, B-cells, etc.).

Evidence of Compliance:
- ✓ Records for the validation of the positive cut-off

REFERENCES

1) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. March 1, 2016.

HSC.31552 HLA Class II Antibody Procedure **Phase II**

The procedure for HLA Class II antibodies readily separates Class I from Class II specificity.

HLA ANTIBODY SCREENING

Inspector Instructions:

	• Sampling of HLA antibody screening policies and procedures, including protocol for screening for each organ transplanted or hematopoietic progenitor cell recipient and the frequency of such screening • Agreement for reflex testing using more sensitive screening method, if applicable • Sampling of antibody identification QC records • Sampling of initial and subsequent recipient sera screening records
	• What is your laboratory's course of action for antibody identification/crossmatching for high risk patients? • How does the laboratory determine cutoffs for identification of HLA antibody based on the clinical programs supported? • How does the laboratory determine the assignment of unacceptable antigens for organ transplantation?

HSC.32487 Immunizing Event **Phase II**

There is a system to record any potential immunizing event that could cause sensitization in a patient.

NOTE: There must be a policy that encourages timely blood sample collection at 14 days after the potential immunizing event in a patient. This new sample should be available for use in antibody screening and crossmatch studies.

REFERENCES

1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):7170 [42CFR493.1278(d)(4)]
2) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.

HSC.32674 HLA Antibody Detection **Phase II**

The laboratory has the capability to detect HLA antibodies with sufficient sensitivity and to distinguish HLA antibodies from IgM autoantibodies or non-HLA antibodies.

NOTE: Methods to detect HLA antibodies must be more sensitive than the basic/NIH technique. There must be written procedures to differentiate HLA antibodies from autoantibodies.

REFERENCES

1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):7170 [42CFR493.1278(d)(1)]
2) Foundation for the Accreditation of Cellular Therapy (FACT) and Joint Accreditation Committee ISCT and EBMT (JACIE). FACT-JACIE International Standards for Cellular Therapy Product Collection, Processing, and Administration. 7th Edition, March 2018.

HSC.32861 Antigenic Diversity and Targets **Phase II**

There is sufficient antigenic diversity (individual antigens and/or crossreactive groups) for HLA Class I and II, and sufficient numbers of antigenic targets for optimal HLA antibody detection and specificity determination.

NOTE: There must be sufficient diversity for Class I and II HLA antigens and cross reactive groups, as well as sufficient numbers of well-characterized panel cells or HLA-purified protein targets for antibody detection and specificity determinations, and strength/avidity of the antibody when applicable.

Evidence of Compliance:

- ✓ Listing of antigenic targets for each panel cell

REFERENCES

1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):7170 [42CFR493.1278(d)]

HSC.33048 Antibody Detection and Specificity QC — Phase II

For HLA antibody detection and specificity determinations, positive and negative controls are used, and sera tested undiluted and diluted when appropriate.

Evidence of Compliance:

- ✓ Written procedure for QC for antibody detection and identification

REFERENCES

1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):7170 [42CFR493.1278(d)(6)]

HSC.33235 Target Source for Class I/II Antibody Determination — Phase II

There are records that the appropriate target sources are used for separate HLA Class I and II antibody determination including appropriate methods to distinguish antibody mixtures.

NOTE: There must be records that the appropriate target sources are used for HLA Class I and II antibody determination. The targets for HLA Class I antibody determination should be blood, spleen, lymph nodes, and cell lines. In addition, well-characterized purified HLA protein targets may also be used. Class II antibodies are best detected utilizing B-lymphocytes, B-lymphoblastoid cell lines, CLL cells or specific Class II purified HLA protein. Mixtures must be defined by methods shown to distinguish Class I from Class II reactivity.

Evidence of Compliance:

- ✓ Written procedure defining target sources used for HLA Class I/II antibody determination and to differentiate antibody mixtures

HSC.33422 Recipient Sera Screening — Phase II

All recipient sera are screened for HLA antibodies including, at least, an initial sample at the time of HLA typing, after sensitizing events and upon request.

Evidence of Compliance:

- ✓ Written procedure defining criteria for screening recipient sera **AND**
- ✓ Records showing initial screening of recipient sera and all subsequent screening results

REFERENCES

1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):7170 [42CFR493.1278(d)]
2) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.

HSC.33475 Antibody Identification/Crossmatching — Phase I

The laboratory has policies and procedures for antibody identification and crossmatching as defined by the transplantation programs supported by the laboratory (includes solid organ and hematopoietic progenitor cell transplantation).

MOLECULAR TESTING

If next generation sequencing methods are used for histocompatibility testing, the requirements in the Next Generation Sequencing section of the Molecular Pathology Checklist must be used in conjunction with these requirements for inspection.

Inspector Instructions:

	• Sampling of molecular HLA typing policies and procedures • Specimen storage/handling procedure • Sampling of QC records • Sampling of HLA typing and hematopoietic progenitor cell engraftment reports for completeness • Sampling of the following records: molecular weight marker, in-house probe labeling validation, nucleic acid measurement, electrophoretic gel interpretation, and chimerism measurement
	• Raw data (eg, gel images, sequencer histograms, flow microbead fluorescence intensity histograms) • Current databases of known sequences for all WHO-recognized alleles (available) • Pre/post amplification areas (adequate physical separation)
	• What is your laboratory's process for assessing the quality (intactness) of high molecular weight DNA or RNA? • How does your laboratory avoid cross-contamination when performing amplification procedures? • How does the laboratory ensure the level of HLA typing resolution is adequate for each transplant service or registry supported (eg, allele-level resolution for hematopoietic progenitor cell transplants)?

GENERAL REQUIREMENTS FOR MOLECULAR TESTING

The requirements in this section are intended to apply to all molecular-based histocompatibility testing.

HSC.34357 Nucleic Acid Extraction/Purification **Phase II**

Nucleic acids are extracted and purified by methods reported in the literature, or there is a record of the validation of a method developed in-house.

Evidence of Compliance:

✓ Records to support nucleic acid extraction/purification is performed by a validated method

REFERENCES
1) Sambrook J, *et al*. Molecular cloning: A laboratory manual, second edition. Cold Spring Harbor, NY: Cold Spring Harbor Laboratory Press, 1989:E.3-E.4
2) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):7170 [42CFR493.1278(5)(i)]

HSC.34544 Reverse Transcription **Phase II**

For RNA amplification methods, appropriate controls are used for reverse transcription.

HSC.34731 Specimen Handling/Storage **Phase II**

Handling and storage of nucleic acids are adequate to prevent degradation.

Evidence of Compliance:
- ✓ Written procedure for specimen storage and handling

REFERENCES
1) Rainen L, *et al.* Stabilization of mRNA expression in whole blood samples. *Clin Chem.* 2002;48:1883-1890

HSC.34760 Dedicated Pipettes **Phase II**

Dedicated pipettors are used for pre-amplification procedures.

HSC.34918 Nucleic Acid Quantity **Phase II**

The quantity of nucleic acid is measured, when appropriate.

Evidence of Compliance:
- ✓ Written policy defining conditions under which quantity of nucleic acid is measured **AND**
- ✓ Records of nucleic acid measurement

HSC.35105 High Molecular Weight DNA/RNA Quality **Phase II**

The quality (intactness) of the high molecular weight DNA (or RNA) is assessed by gel electrophoresis or comparable method, when appropriate.

Evidence of Compliance:
- ✓ Written procedure defining method for assessment of quality (intactness)

HSC.35292 Analytical Gels **Phase II**

Standard amounts of nucleic acid are loaded on analytical gels, when possible.

HSC.35479 Gel Images **Phase II**

Gel images are of sufficient resolution and quality (low background, clear signal, absence of bubbles, etc.) to permit the reported interpretation.

HSC.35666 Specimen Handling **Phase II**

There are written procedures to prevent specimen loss, alteration, or contamination.

HSC.36040 Enzymatic Amplification Procedures **Phase II**

Enzymatic amplification procedures (eg, PCR) minimize carryover (false positive results) by using appropriate physical containment and procedural controls.

NOTE: This item is primarily directed at ensuring adequate physical separation of pre- and post-amplification samples to avoid amplicon contamination. The extreme sensitivity of amplification systems requires that the laboratory take special precautions. For example, pre- and post-amplification samples should be manipulated in physically separate areas; gloves must be worn and frequently changed during processing; dedicated pipettes (positive displacement type or with aerosol barrier tips) must be used; manipulations must minimize aerosolization; following complete reagent addition to the reaction tubes, the patient samples should be added one at a time. The best way to avoid cross-contamination is to use the following order of preparation

within an amplification run: actual samples, followed by positive controls, followed by negative controls.

Evidence of Compliance:

- ✓ Written procedure that defines the use of physical containment and procedural controls as applicable to minimize carryover

REFERENCES

1) Kwok S, Higuchi R. Avoiding false positives with PCR. *Nature* 1989;339:237-238
2) Clinical and Laboratory Standards Institute. *Establishing Molecular Testing in Clinical Laboratory Environments; Approved Guideline.* CLSI document MM19-A. Clinical and Laboratory Standards Institute, Wayne, PA; 2011.

HSC.36414 Electrophoretic Gel Interpretation — Phase II

Electrophoretic gels are interpreted independently by at least two qualified readers using an objective method.

Evidence of Compliance:

- ✓ Written procedure for interpretation of electrophoresis gels **AND**
- ✓ Patient testing records or worksheets

HSC.36601 End-Point Amplification QC — Phase II

For end-point amplification assays such as sequence-specific priming, adequate internal controls are used, and criteria defined for a positive reaction.

HSC.36788 Daily Controls — Phase II

For qualitative and quantitative tests, positive and negative controls are included for each assay, where appropriate, in every run, and as specified in the manufacturer's instructions (as applicable) and laboratory procedure.

Evidence of Compliance:

- ✓ Written QC procedures **AND**
- ✓ Records of QC results, including external and internal control processes **AND**
- ✓ Manufacturer's product insert or manual, as applicable

REFERENCES

1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24): [42CFR493.1278(b)(6)], [42CFR493.1256(d)(3)(i), (ii)].
2) Clinical and Laboratory Standards Institute (CLSI). *Establishing Molecular Testing in Clinical Laboratory Environments; Approved Guideline.* CLSI document MM19-A. Clinical and Laboratory Standards Institute, Wayne, PA; 2011.

HSC.36795 Internal Controls - Nucleic Acid Amplification — Phase II

In all nucleic acid amplification procedures, internal controls are run to detect a false negative reaction secondary to extraction failure or the presence of an inhibitor, when appropriate.

NOTE: The laboratory should be able to distinguish a true negative result from a false negative due to failure of extraction or amplification. Demonstration that another sequence can be successfully amplified in the same specimen should be sufficient to resolve this issue. For quantitative amplification assays, the effect of partial inhibition must also be addressed.

The internal control should not be smaller than the target amplicon.

Evidence of Compliance:

- ✓ Written procedure defining use of internal controls **OR** records of assay validation and monitoring statistics for test result trends

REFERENCES

1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24): [42CFR493.1256(d)(3)(iv)(v)].

HSC.36975 DNA Contamination **Phase II**

There is a procedure to detect and control for DNA contamination.

NOTE: Contamination must be monitored in different areas by wipe tests using the regular detection for testing. There are records of the results of monitoring and corrective action taken when contamination is detected.

HSC.37162 Molecular Weight Markers **Phase II**

Known molecular weight markers that span the range of expected bands are used for each electrophoretic run.

Evidence of Compliance:

✓ Records of appropriate markers with each run

HSC.37349 Amplification Quality **Phase II**

For hybridization techniques, there are records that the different components and steps are monitored and acceptable, including the amount and integrity of amplified product and the signal intensity produced by each probe.

REFERENCES

1) Clark JR, Scott SD, Jack AL, et al. Monitoring of chimerism following allogeneic haematopoietic stem cell transplantation (HSCT): Technical recommendations for the use of short tandem repeat (STR) based techniques, on behalf of the United Kingdom National External Quality Assessment Service for Leucocyte Immunophenotyping Chimerism Working Group. *Br J Haematol.* 2015;168(1):26-37.

HSC.37536 Pre-Analytic Testing Requirements **Phase II**

The conditions (temperature, salt concentration, probe concentration, etc.) for pre-hybridization, hybridization, and solid-phase support systems are optimized to consistently produce accurate results.

HSC.37723 Probe Labeling Validation **Phase II**

The method of probe labeling is validated to detect the target sequence without a false positive signal for non-target sequences.

Evidence of Compliance:

✓ Records of in-house validation study data

HSC.37910 Re-Probing **Phase II**

If re-probing a solid-phase nucleic acid sample is performed, there are records of complete stripping of the previous probe before re-probing.

MOLECULAR HLA TYPING

*****REVISED** 09/22/2021***

HSC.38060 HLA Typing Level of Resolution **Phase II**

The level of resolution of HLA typing is adequate for the clinical programs, including donor registries, and the type of cell, tissue, or organ to be transplanted and meets the requirements of relevant accrediting agencies.

NOTE: Laboratories performing testing for NMDP donors must follow NMDP policies for resolution of typing ambiguities. Alternative allele combinations must be resolved when they contain one or more alleles in the common or intermediate categories of the CIWD 3.0.0 catalog.

For hematopoietic progenitor cell transplant, the laboratory must perform HLA typing at the level of resolution and including the loci required by the agreements with the transplant center and/or donor registry. For example, high resolution typing of HLA-A, B, C, DRB1 and DPB1 is mandatory for patient and unrelated donor matching per NMDP.

When performing HLA typing of deceased donors for the purpose of solid organ allocation in the United States, report the following loci as required by OPTN policies: A, B, Bw4, Bw6, C, DRB1, DRB3/4/5, DQA1, DQB1, and DPB1.

REFERENCES
1) Organ Procurement and Transplantation Network (OPTN) Policies. Policy 4: Histocompatibility. US Department of Health and Human Services. Effective Date: September 10, 2020.
2) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):7170 [42CFR493.1278(b)(2)]
3) Foundation for the Accreditation of Cellular Therapy (FACT) and Joint Accreditation Committee ISCT and EBMT (JACIE). FACT-JACIE International Standards for Cellular Therapy Product Collection, Processing, and Administration. 7th Edition, March 2018.
4) National Marrow Donor Program (NMDP)/Be The Match. US Transplant Center Participation Criteria. Document #A00228. Revised 2017.
5) National Marrow Donor Program. NMDP Policy for HLA Confirmatory Typing Requirements for Unrelated Adult Donors and HLA Typing Requirement for Patients. Document Number: P00079. Effective date: February 27, 2021.
6) Hurley CK, et al. Common, intermediate and well-documented HLA alleles in world populations: CIWD version 3.0.0. *HLA.* 2020;95(6):516-531.

*****REVISED** 09/22/2021***

HSC.38097 Sequence Based Typing **Phase II**

For sequence based typing, there are records of the following:

- **Templates with sufficient specificity for a locus or allele**
- **Appropriate monitoring of all steps**
- **Adequate electrophoretogram quality to support the sequence results**
- **Definition of a sequence following a procedure for accurate assignment of HLA alleles**

NOTE: Records must include the HLA locus and allele specificity of the template, the source of the sequence data base used (annually updated), and procedures to resolve ambiguous combinations. Assignment of alleles for HLA loci must be done by comparing the sequence data with the sequences of all alleles that are recognized by the WHO.

Laboratories must recognize ambiguous allele combination(s) and resolve these as appropriate for the clinical use as defined by the transplant agreement.

HSC.38100 Result Reporting - HLA Typing **Phase II**

For HLA typing, there is sufficient information available on the nature of all probes or primers used in an assay to permit interpretation and troubleshooting of test results.

NOTE: Items that must be defined are:

1. *HLA allele specificity of probes and primers*
2. *The HLA locus and allele designations recognized by the WHO for each combination of positive results (hybridization for SSOP and PCR product for SSP)*
3. *Ambiguous allele combinations*
4. *The HLA sequence database used*

REFERENCES
1) McAlpine PJ, *et al*. The 1991 catalog of mapped genes and report of the nomenclature committee. Human gene mapping 11(1991). *Cytogenet Cell Genet.* 1991, 58:5-102

HEMATOPOIETIC PROGENITOR CELL ENGRAFTMENT MONITORING

HSC.38120 Hematopoietic Progenitor Cell Engraftment **Phase II**

For hematopoietic progenitor cell engraftment, the polymorphic nature and independent segregation (eg, location on separate chromosomes) of the DNA system used is detailed and recorded in the literature.

REFERENCES
1) Clark JR, Scott SD, Jack AL, et al. Monitoring of chimerism following allogeneic haematopoietic stem cell transplantation (HSCT): Technical recommendations for the use of short tandem repeat (STR) based techniques, on behalf of the United Kingdom National External Quality Assessment Service for Leucocyte Immunophenotyping Chimerism Working Group. *Br J Haematol.* 2015;168(1):26-37.

HSC.38130 Chimerism **Phase II**

There are records of the accuracy of quantitative methods used to measure chimerism.

NOTE: The accuracy of quantitative methods used to measure chimerism must be verified at least annually by controlled blood mixing or other suitable method. If results on cell subpopulations are reported, there must be records of periodic testing of the purity of such cell subsets.

HSC.38140 Negative Control **Phase II**

A negative control is used and evaluated for non-specific background with each run.

REFERENCES
1) Clark JR, Scott SD, Jack AL, et al. (2015), Monitoring of chimerism following allogeneic haematopoietic stem cell transplantation (HSCT): Technical recommendations for the use of Short Tandem Repeat (STR) based techniques, on behalf of the United Kingdom National External Quality Assessment Service for Leucocyte Immunophenotyping Chimerism Working Group. *Br J Haematol.* 2015;168:26-37.

HSC.38150 Sensitivity Control **Phase II**

A sensitivity control is used and evaluated with each run.

NOTE: A low positive control may be used to meet this requirement.

HSC.38171 Internal Controls **Phase II**

For hematopoietic progenitor cell engraftment assays, internal controls are used to determine appropriate genotypes or at least to distinguish patient from donor(s) with each run.

NOTE: There must be criteria for the acceptance and rejection of the amplification of a particular genetic locus or individual sample.

Evidence of Compliance:
- ✓ Written procedure defining criteria for acceptance/rejection of amplification results

HSC.38180 Preferential Amplification **Phase II**

Reactions are optimized to avoid preferential amplification. The minimum amount of DNA is determined to obtain optimal sensitivity.

NOTE: Method validation must include a dilution study to evaluate the concentration of DNA to determine minimum sensitivity of the assay.

HSC.38190 Cell Subset Purity **Phase II**

If cell subset enrichment is performed, the patient report includes the actual or approximate purity of the cell subset.

NOTE: The determination of the actual or approximate purity of the cell subset does not imply that the purity determined in validation studies can be used without further evaluation. An actual measurement may be performed at the time of sample testing. Some isolation methods and cell subpopulations (eg, CD56) may not produce enough cells to test purity and run the monitoring engraftment test. At a minimum, the purity can be determined for each lot of reagent used to isolate the cell subset and then be reported as an approximate purity for that specific lot.

HSC.38200 Hematopoietic Progenitor Cell Engraftment Testing **Phase II**

For hematopoietic progenitor cell engraftment, samples from pre-transplant patient (recipient), pre-transplant donor(s), post-transplant patient, and an appropriate control are analyzed concurrently.

NOTE: Previously generated data from pre-transplant specimens may be used to compare to post-transplant results if a validated system is used to identify and link the appropriate data files for concurrent analysis.

Evidence of Compliance:
- ✓ Written procedure for hematopoietic progenitor cell engraftment testing

HSC.38205 Engraftment Analysis **Phase II**

Prior to evaluating post engraftment specimens, the laboratory evaluates a specimen from the donor(s) and a pre-transplant specimen from the patient to determine the number of informative loci to test in order to meet the minimum number of loci needed for calculations.

Evidence of Compliance:
- ✓ Written procedure for hematopoietic progenitor cell engraftment testing **AND**
- ✓ Records of hematopoietic progenitor cell engraftment testing

HSC.38208 Preferential Allele Amplification **Phase II**

Preferential allele amplification is considered in the interpretation of hematopoietic progenitor cell engraftment tests.

HSC.38220 Minimal Number of Informative Loci **Phase II**

For hematopoietic progenitor cell engraftment testing, a minimum of three informative loci are routinely used in the calculations.

NOTE: There are exceptions to this rule. Informative loci refer to loci that can distinguish between donor(s) and recipient. An exception for the number of informative loci used may occur in syngeneic twins (donor(s) and recipient) and rarely in closely related donor(s) and recipient.

HSC.38658 Result Reporting **Phase II**

For hematopoietic progenitor cell engraftment, the final report includes an appropriate summary of the methods, the loci tested, the number of informative loci used, the percent donor cells, an indication of any trace cells, and the sensitivity of the assay.

REFERENCES
1) Clark JR, Scott SD, Jack AL, et al. Monitoring of chimerism following allogeneic haematopoietic stem cell transplantation (HSCT): Technical recommendations for the use of short tandem repeat (STR) based techniques, on behalf of the United Kingdom National External Quality Assessment Service for Leucocyte Immunophenotyping Chimerism Working Group. *Br J Haematol.* 2015;168(1):26-37.
2) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.

ADDITIONAL MOLECULAR TESTING METHODS

HSC.38690 ABO and RhD Typing by Molecular Methods Phase II

ABO and RhD typing performed by molecular methods is used for presumptive ABO and RhD typing only. Donor-recipient ABO and RhD typing for transfusion and transplant compatibility evaluations is performed using FDA-cleared or approved serologic methods.

NOTE: Transplant donor registries often collect samples from potential donors using buccal swabs or saliva. These samples cannot be used for traditional serological ABO/RhD blood group typing because fresh intact red blood cells (RBCs) are not available. Molecular ABO and RhD typing may be performed to predict the presumptive ABO and RhD phenotype to aid in finding an appropriate donor. Because the ABO and Rh genes are complex, prediction of ABO and Rh phenotype by molecular methods is currently used in immunohematology red cell reference laboratories that focus on blood typing complications, for research, or for providing ***preliminary*** *information that can be confirmed by FDA-cleared or approved methods.*

The use of molecular based screening assays is not acceptable for ABO and RhD blood type assignment for the purposes of transfusion or transplantation. ABO and RhD typing by FDA-cleared or approved serologic methods must be used for the purpose of transfusion or donor and recipient ABO and RhD typing for transplantation.

Evidence of Compliance:
- ✓ Donor-recipient compatibility records with serologic ABO and RhD typing results **AND**
- ✓ Written policy for molecular ABO and RhD typing limiting use for non-clinical purposes

DONOR-RECIPIENT HISTOCOMPATIBILITY

Inspector Instructions:

READ	• Sampling of donor recipient histocompatibility policies and procedures • Sampling of cell typing records • Staffing/on call schedule
ASK	• What evidence, from family studies, does your laboratory use to support haplotype or other genetic reporting conclusions (four distinct haplotypes must be identified in a pedigree analysis for haplotype assignment)?

HSC.38845 HLA Identity Confirmation for Hematopoietic Progenitor Cell Transplantation **Phase II**

HLA identity is confirmed in both donor and recipient hematopoietic progenitor cell transplantation.

NOTE: For laboratories performing typing to support hematopoietic progenitor cell (HPC) transplants facilitated by NMDP, NMDP policy for patient typing and confirmatory typing of unrelated donors must be followed.

For laboratories performing typing to support HPC transplantation, repeat HLA typing of the transplant patient using a new sample is performed to verify the individual's HLA type prior to final donor selection for both related and unrelated transplants.

Similarly, repeat HLA typing of the related HPC donor is performed using a new sample prior to HPC collection. For purposes of verification testing, results from donor registry or another laboratory is acceptable as the first of the two samples required. For example, lower resolution is acceptable for verifying the original high-resolution typing of recipients and related donors.

Evidence of Compliance:

- ✓ Written procedure defining confirmation method(s) used for HLA identity

REFERENCES

1) Nunes E., *et al*. Definitions of histocompatibility typing terms: Harmonization of Histocompatibility Typing Terms Working Group. *Hum Immunol*. 2011; 72(12):1214-6; *Blood*. 2011; 118:e180-3
2) National Marrow Donor Program. NMDP Policy for HLA Confirmatory Typing Requirements for Unrelated Adult Donors and HLA Typing Requirement for Patients. Document Number: P00079. Effective date: February 27, 2021.

HSC.39032 Haplotype Reporting **Phase II**

Haplotype assignments are supported by sufficient evidence.

NOTE: When reporting haplotypes, homozygosity, blanks, recombination, or other genetic information, there must be sufficient evidence from family studies to support such conclusions. If probable haplotypes are reported, the report must indicate clearly that they are "probable". Reliable haplotype frequencies of the appropriate ethnic groups must be used.

If haplotypes are assigned, this can be done by testing both the parents or clearly defined segregation of the four haplotypes or may be based upon population frequencies. Haplotype assignments based on population frequency, must be clearly stated on the report and include the relevant source (including the version) or reference.

HSC.39219 Donor Typing for Solid Organ Transplant **Phase I**

Donor materials obtained pre-organ recovery are used whenever possible for donor HLA typing and recipient serum screening.

NOTE: Organ donors should be HLA-typed from any acceptable source of viable lymphocytes. Whenever possible, pre-organ recovery HLA testing and screening crossmatches should be done.

Evidence of Compliance:

- ✓ Written procedure defining criteria for organ donor typing **AND**
- ✓ Record of typing and screening on pre-organ donor materials, when possible

HSC.39406 Donor and Recipient HLA Typing **Phase II**

The HLA laboratory types all potential recipients and donors referred to the laboratory, and follows policies defining when HLA retyping and redefinition are required.

Evidence of Compliance:

- ✓ Written policies and procedures for typing potential recipients and donors with criteria for HLA retyping and redefinition **AND**
- ✓ Records of all HLA typing

REFERENCES

1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):7170 [42CFR493.1278]

HSC.39410 HLA Typing Post Transfusion **Phase II**

There is a procedure describing what to do when a donor or patient cannot be reliably HLA typed after transfusion.

NOTE: Transfusions may result in detection of additional HLA antigens and/or alleles in donor or patient samples. Alternative source typing material (eg, buccal swab, lymph node or spleen) should be considered if more than two antigens are detected for a locus or alleles cannot be discriminated. Typing should be performed at the level of resolution required for the transplant service being supported.

*****REVISED** 09/22/2021***

HSC.39415 HLA Typing Level **Phase II**

The laboratory performs HLA typing at least to the minimal resolution appropriate for the individual transplant program supported (*eg,* 2-field typing for hematopoietic progenitor cell transplantation, when appropriate, and the level of serological splits for solid organ transplantation).

Evidence of Compliance:

- ✓ Written procedures for HLA typing of solid organ and hematopoietic progenitor cell transplantation **AND**
- ✓ Patient reports and typing records

REFERENCES

1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):[42CFR493.1278(b)(2)]
2) Nunes E., et al. Definitions of histocompatibility typing terms: Harmonization of Histocompatibility Typing Terms Working Group. *Hum Immunol.* 2011; 72(12):1214-6; *Blood* 2011; 118:e180-3.
3) Organ Procurement and Transplantation Network (OPTN) Policies. Policy 4: Histocompatibility. US Department of Health and Human Services. Effective Date: September 10, 2020.
4) National Marrow Donor Program. NMDP Policy for HLA Confirmatory Typing Requirements for Unrelated Adult Donors and HLA Typing Requirement for Patients. Document Number: P00079. Effective date: February 27, 2021.

*****REVISED** 09/22/2021***

HSC.39430 Written Agreements **Phase II**

There are written agreements for histocompatibility testing with each transplant program, organ procurement organization (OPO), or donor registry served by the laboratory, unless clinical urgency prevents such an agreement.

NOTE: Written agreements must be reviewed biennially by the histocompatibility section director/ technical supervisor, and/or clinical consultant, and the clinical transplant program director, and be revised as necessary.

If the laboratory participates as a member of the United Network for Organ Sharing (UNOS), the written agreements must address all elements defined in the Organ Procurement and Transplantation Network (OPTN) Bylaws when applicable:

- *The sample requirements for typing and crossmatching*
- *The loci and level of resolution typed*
- *A process for requesting extended HLA typing*

- *A process for reporting and verifying HLA and unacceptable antigen data at the time of registration on the waiting list and any time there are changes*
- *A process for reporting HLA typing results to the OPTN Contractor*
- *A process for resolving HLA typing discrepancies and errors*
- *The maximum turnaround time from receipt of sample to reporting of results to the transplant program*
- *A process to obtain sensitization history for each patient*
- *The frequency of periodic sample collection*
- *The frequency of antibody screenings*
- *The criteria for crossmatching*
- *The assay format that will be used for antibody screening and for crossmatching*
- *The criteria for determining unacceptable antigens used during organ allocation*
- *The duration for which specimens need to be stored for repeat or future testing*
- *If desensitization is performed, a protocol for monitoring antibody levels*
- *If the laboratory registers candidates for the transplant process, a process for blood type verification*
- *If post-transplant monitoring is performed, a protocol for monitoring antibody levels.*

If the laboratory supports a program or donor registry that is accepted through the Foundation for the Accreditation of Cellular Therapy (FACT), the agreements must contain the requirements defined in the 7th edition of the FACT Standards.

If a laboratory supports a program or donor registry that is participating in the National Marrow Donor Program (NMDP)/Be The Match, the agreement must contain the provisions defined in the November 2017 NMDP U.S. Transplant Center Participation Criteria.

Agreements with OPOs must also include the following:

- *Process for prioritizing donors for histocompatibility testing*
- *All methods used for crossmatching, interpretation, and reporting of results if crossmatching is done by the OPO*

REFERENCES
1) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. August 17, 2020.
2) Foundation for the Accreditation of Cellular Therapy (FACT) and Joint Accreditation Committee ISCT and EBMT (JACIE). FACT-JACIE International Standards for Cellular Therapy Product Collection, Processing, and Administration. 7th Edition, March 2018.
3) National Marrow Donor Program (NMDP)/Be The Match. US Transplant Center Participation Criteria. Document #A00228. Revised 2017.

HSC.39450 Histocompatibility Testing Requests **Phase II**

Records exist of histocompatibility testing requests not covered by the transplant program support agreement.

NOTE: The laboratory has records of HLA testing requests which deviate from or are not covered in the existing transplant program support agreement (eg, the use of serum for a final crossmatch that is "too old" or "no final crossmatch" for a patient who would normally require a crossmatch within a previously defined time before transplant).

HSC.39499 Laboratory Coverage Plan **Phase II**

The laboratory coverage plan for staffing ensures that qualified testing personnel and key personnel are available to perform histocompatibility testing for organ transplantation and to facilitate organ acceptance and transplantation as needed.

NOTE: For laboratories that are members of the United Network for Organ Sharing (UNOS), the following staff availability requirements from the OPTN Bylaws apply:

- *Key personnel include the laboratory director, technical supervisor, general supervisor, and the clinical consultant*

- *The plan must include coverage at all times, including when changes occur in key personnel, and address coverage when key personnel serve more than one laboratory*
- *If the laboratory performs testing on deceased organ donors, key personnel and qualified testing personnel must be available 24-hours a day, seven days a week, unless an alternative coverage plan has been approved by UNOS/OPTN Membership and Professional Standards Committee*

Evidence of Compliance:

✓ Staffing schedule **OR** on-call schedule if 24-hour staffing is not available

REFERENCES

1) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.

NON-RENAL ORGAN TRANSPLANTS

Inspector Instructions:

- Sampling of non-renal organ transplant policies and procedures
- Sampling of antibody screening records
- Sampling of crossmatch records

HSC.39593 Recipient Screening — Phase II

Solid organ transplant recipients are screened for HLA Class I and Class II antibodies by a solid phase method.

NOTE: The frequency of testing is determined by the transplant program support agreement.

Evidence of Compliance:

✓ Written procedure for antibody screening of non-renal transplant recipients **AND**

✓ Records of antibody screening on transplant recipients

REFERENCES

1) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.
2) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2003(Jan 24):7170 [42CFR493.1278(d)(7)]

HSC.39780 Prospective Crossmatch — Phase II

Non-renal sensitized transplant recipients are prospectively crossmatched with their potential donor, whenever possible, before transplantation.

NOTE: The technique used for crossmatching in these patients must be one of enhanced sensitivity for antibody detection.

Evidence of Compliance:

✓ Written procedure for crossmatch of non-renal sensitized transplant recipients **AND**

✓ Crossmatch records

REFERENCES

1) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.

HSC.39850 CD34 Cellular Viability - Apheresis and Cord Blood Products — Phase II

The laboratory measures the viability of CD34 positive cells in samples aliquoted at the time of processing of hematopoietic progenitor cells, apheresis products and cord blood products.

NOTE: CD34 cell viability testing of cord blood products must be done on a sample aliquoted prior to the addition of cryoprotectant.

For any hematopoietic progenitor cell product, CD34 cell viability testing during or after storage should be considered as an additional quality control.

The viability dye 7-amino actinomycin-D (7-AAD) yields excellent results in this analysis. The viability assay must be performed using a flow cytometric method with the viability dye included in the same tube with the CD34 and CD45 monoclonal antibodies for the CD34+ viability determination. Estimates of total cellular viability (for example, trypan blue exclusion) may not be used as an alternative because the method can overestimate the viability of the CD34 stem cell population.

REFERENCES
1) Owens M, Loken M. Peripheral blood stem cell quantitation, In Flow Cytometry Principles for Clinical Laboratory Practice. New York, NY: Wiley-Liss, 1995:111-127
2) Keeney M., *et al.* Single platform flow cytometry absolute CD34+ cell counts based on the ISHAGE guidelines. *Cytometry.* 1998; 34:61-70
3) Hubl W., *et al.* Measurement of absolute concentration and viability of CD34+ cells in cord blood and cord blood products using fluorescent beads and cyanine nucleic acid dyes. *Cytometry.* 1998; 34:121-127
4) Gratama J., *et al.* Flow cytometric enumeration of CD34+ hematopoietic stem and progenitor cells. *Cytometry.* 1998;34:128-145
5) Lee S., *et al.* Post-thaw viable CD34+ cell count is valuable predictor of haematopoietic stem cell engraftment in autologous peripheral blood stem cell transplantation. *Vox Sang* Feb: 2008; 94:46-152
6) Riech-Slotky R., *et al.* Determining post-thaw CD34+ cell dose of cryopreserved haematopoietic progenitor cells demonstrates high recovery and confirms their integrity. *Vox Sang* 2008: May; 94(4):351-357
7) Clinical and Laboratory Standards Institute. *Enumeration of Immunologically Defined Cell Populations by Flow Cytometry; Approved Guideline.* 2nd ed. CLSI Document H42-A2. Clinical and Laboratory Standards Institute, Wayne, PA; 2007.

PERSONNEL

Inspector Instructions:

READ

- Records of section director/technical supervisor/testing personnel/clinical consultant education and experience
- Continuing education policy
- Sampling of continuing education records

HSC.40000 Section Director/Technical Supervisor Qualifications Phase II

The section director (technical supervisor) of the histocompatibility section has the following qualifications.

1. **MD or DO licensed to practice (if required) in the jurisdiction where the laboratory is located, or doctoral degree in chemical, physical, biological or clinical laboratory science from an accredited institution, AND**
2. **Laboratory training and experience: four years training and experience in histocompatibility, or two years training and experience in general immunology plus two years in histocompatibility. For section director/ technical supervisors supporting solid organ and/or hematopoietic progenitor cell transplantation, records of training or relevant experience in histocompatibility appropriate to the supported transplant program(s)**

NOTE: If there has been a change in the HLA Section Director (Technical Supervisor) in the last two years, the inspector must review the new section director's curriculum vitae and portfolio. The review should include at least 10 solid organ transplant cases from the portfolio for a

laboratory supporting a solid organ transplant program and at least 10 hematopoietic progenitor cell transplant cases representative of the program mix of related and unrelated transplants for a laboratory supporting hematopoietic progenitor cell transplantation.

If more stringent state or local regulations are in place for supervisory qualifications, including requirements for state licensure, they must be followed.

Evidence of Compliance:

- ✓ Records of section director/technical supervisor qualifications including diploma, transcript(s), primary source verification report, equivalency evaluation, board certification, or current license (if required) **AND**
- ✓ Work history in related field

REFERENCES
1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 1992(Feb 28):7180 [42CFR493.1449(o)]
2) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.

HSC.40100 Notification of Change in Key Personnel — Phase II

If the histocompatibility laboratory participates as a member of the United Network for Organ Sharing (UNOS), there is a policy to notify the CAP's Laboratory Accreditation Program when there is a change in key personnel, including the section director/technical supervisor, general supervisor, and/or clinical consultant.

NOTE: Notification must occur no later than 30 days prior to the change; or in the case of an unexpected change, no later than 2 working days afterwards. For changes in laboratory directorship, refer to GEN.26791.

REFERENCES
1) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.

HSC.45000 Testing Personnel Qualifications — Phase II

Personnel performing the technical work of histocompatibility have at least one year of training and/or experience in histocompatibility and qualify as high complexity testing personnel with a minimum of the following:

1. **Bachelor's degree in a chemical, physical, biological or clinical laboratory science or medical technology; or**
2. **Associate degree in a laboratory science or medical laboratory technology from an accredited institution, or equivalent laboratory training and experience meeting the requirements defined in the CLIA regulation 42CFR493.1489. The qualifications to perform high complexity testing can be accessed using the following link: CAP Personnel Requirements by Testing Complexity.**

NOTE: Persons with less than one year of training and/or experience must work under the supervision of persons who are qualified.

Evidence of Compliance:

- ✓ Records of section director/technical supervisor qualifications including diploma, transcript(s), primary source verification report, equivalency evaluation, board certification, or current license (if required) **AND**
- ✓ Work history in related field

REFERENCES
1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 1992(Feb 28):7175 [42CFR493.1423], 7183 [42CFR493.1489]
2) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.

HSC.46250 Supervisor Qualifications **Phase II**

Supervisor(s)/general supervisor(s) are qualified by experience in histocompatibility/ transplantation.

NOTE: The supervisor/general supervisor must have a minimum of three years' experience in histocompatibility/transplantation.

Evidence of Compliance:

- ✓ Records of work history in personnel file

REFERENCES
1) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.

HSC.47500 Continuing Education Program **Phase II**

There is a continuing clinical laboratory education program that addresses the areas of service offered by the laboratory, identifies the need for remedial training, where appropriate, and provides continuing education to improve skills in histocompatibility.

NOTE: The laboratory must have a complete continuing clinical laboratory education program that meets the needs of the various types of laboratory personnel and addresses the areas of service offered by the laboratory, including a predefined minimum number of contact hours annually. This program may be provided locally, regionally/nationally, through scientific article review and discussion, or some combination of the above.

Evidence of Compliance:

- ✓ Written policy for continuing education requirement for all personnel **AND**
- ✓ Records of continuing education

REFERENCES
1) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.

HSC.48750 Clinical Consultant Qualifications **Phase II**

The section director or other individual fulfills the responsibilities of clinical consultant.

NOTE: The clinical consultant must be an MD or DO licensed to practice medicine in the jurisdiction where the laboratory is located (if required) with appropriate training and experience in the interpretation of histocompatibility / transplantation immunology test results, or a doctoral level clinical scientist certified by a CLIA-approved specialty board.

Evidence of Compliance:

- ✓ Records of section director/technical supervisor qualifications including diploma, transcript(s), primary source verification report, equivalency evaluation, board certification, or current license (if required) **AND**
- ✓ Work history in related field **AND**
- ✓ Job description defining responsibilities of clinical consultant

REFERENCES
1) Department of Health and Human Services, Centers for Medicare and Medicaid Services. Clinical laboratory improvement amendments of 1988; final rule. *Fed Register.* 2002(Oct 1):1043 [42CFR493.1417] and 1038 [42CFR493.1405(b)
2) Organ Procurement and Transplantation Network (OPTN) Bylaws. Appendix C. Membership Requirements for Histocompatibility Laboratories. US Department of Health and Human Services. November 20, 2018.