COLLEGE of AMERICAN PATHOLOGISTS

December 2024 Changes

Hematology and Coagulation Checklist

CAP Accreditation Program

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

12.26.2024

Disclaimer and Copyright Notice

CAP 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).

Using the Changes Only Checklist

This document contains new checklist requirements, major and minor requirement revisions, and changes to explanatory text. **Changes appear in a track changes format that compares the previous checklist edition to the December 26, 2024 edition.** Requirements with significant revisions will display a "Revised" flag. These changes may affect your laboratory operations. Requirements with minor revisions will not display a "Revised" flag. They are editorial changes that are not likely to affect your laboratory operations.

Information regarding requirements that are new or have been combined, moved, resequenced or deleted, as applicable, appears in table format below.

2024 CHECKLIST EDITION CHANGES
NEW, DELETED, MERGED, AND MOVED REQUIREMENTS *

2023 Requirement		Action Taken	2024 Requirement	
		New	HEM	36960

*Deleted – Removed the requirement from the checklist edition
*Merged – Combined the requirement with a similar requirement in the same or different checklist
*Moved – Relocated the requirement to another checklist or resequenced it within the same checklist

ON-LINE CHECKLIST DOWNLOAD OPTIONS

Participants of the CAP accreditation programs may download the checklists ~~from the CAP website (cap.org)~~ by logging into cap.org and going to e-LAB Solutions Suite - Accreditation Checklists. 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.

INTRODUCTION

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

Certain requirements are different for waived versus nonwaived tests. Refer to the checklist headings and explanatory text to determine applicability based on test complexity. The current list of tests waived under CLIA may be found at http://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfClia/analyteswaived.cfm.


Policy/Procedure icon - The placement of this icon next to a checklist requirement indicates that a written policy or procedure is required to demonstrate compliance with the requirement. The icon is not intended to imply that a separate policy or procedure is required to address individual requirements. A single policy or procedure may cover multiple checklist requirements.

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).*

HEMATOLOGY

MANUAL HEMATOCRIT

***REVISED** 12/26/2024*

HEM.32050 Microhematocrit Centrifuge **Phase I**

The speed of the microhematocrit centrifuge is checked at least annually.

NOTE: Relative centrifugal field (RCF) must be sufficient to achieve maximum packing of cells. ~~The~~It is recommended that the centrifuge ~~must~~ be capable of sustaining an RCF of 10,000 to 15,000 at the periphery for five minutes.

If the centrifuge speed cannot be checked by the user, the laboratory must annually compare centrifuge test results against another centrifuge with known speed and constant packing time. If the laboratory does not have such an instrument, another laboratory or an outside vendor may be used for this comparison.

Evidence of Compliance:

✓ Records of microhematocrit centrifuge speed checks

REFERENCES

1) Clinical and Laboratory Standards Institute. *Procedure for Determining Packed Cell Volume by the Microhematocrit Method; Approved Standard;* 3rd ed. CLSI document H07-A3. CLSI, Wayne, PA, 2000.

BODY FLUIDS

MANUAL CELL COUNT - BODY FLUID

HEM.35338 Background Checks - Manual Counts **Phase II**

The diluting fluid is checked for ~~non-specimen~~interfering background particulates and changed when indicated.

NOTE: Checking can be done by examining samples of these fluids under the microscope. The check must be performed each day of use for manual diluting methods. If commercial microdilution systems are used, daily checks are not required but each lot must be examined visually for uniformity of filling and clarity. If diluting fluids are prepared by the laboratory, they must be prepared aseptically; refrigeration is recommended to prevent contamination with microorganisms.

Evidence of Compliance:

✓ Records of background checks

AUTOMATED CELL COUNT - BODY FLUID

***REVISED** 12/26/2024*

HEM.35414 Background Checks - Automated Counts **Phase II**

Instrument background counts are performed each day of testing on the diluent fluid and lysing agent to check for contamination that might affect cell counts.

NOTE: This can be done by processing these fluids on the instrument used for cell counting and checking for the presence of significant background in the diluting fluids and lysing agents.

For any external diluting fluid not part of the instrument's reagent system, the laboratory must check for interfering background particulates each day of use. Checking can be done by examining samples of these fluids under the microscope. If commercial microdilution systems are used, daily checks are not required but each lot must be examined visually for uniformity of filling and clarity. If diluting fluids are prepared by the laboratory, they must be prepared aseptically; refrigeration is recommended to prevent contamination with microorganisms.

Evidence of Compliance:

✓ Records of background checks **OR** records of interfering background particulate checks on external dilution fluids/reagents

SEMEN ANALYSIS

Requisitions, Specimen Receipt and Results Reporting

HEM.35661 Azoospermic Specimen Result Reporting **Phase I**

For azoospermic and post-vasectomy seminal fluid specimens, the laboratory clearly communicates the findings of the assay and either employs a concentrating technique on seminal fluid or includes a comment in the patient report indicating that a concentrating technique was not performed.

NOTE 1: Without a concentration technique, the presence of both motile and non-motile sperm may not be detected. The method for detection of motile and non-motile sperm and the laboratory findings must be clearly communicated on the patient report so that the clinician can interpret the results in context to the method performed. The decision on the method used and extent of testing to be performed should be made in consultation with the medical staff served.

The American Urological Association (AUA) Vasectomy Guideline recommends a careful evaluation of an uncentrifuged specimen, and does not recommend centrifugation of the specimen for further assessment. The AUA Guideline also recommends reporting both the presence and absence of sperm and presence or absence of sperm motility on the patient report. If no sperm are seen in the uncentrifuged specimen, the guideline recommends reporting that the presence of sperm is below the limit of detection.

NOTE 2: If the laboratory only performs post-vasectomy checks for presence or absence of sperm, HEM.35661 is the only applicable requirement in this section.

Evidence of Compliance:

✓ Patient report with concentration findings or appropriate comment indicating that concentration was not performed

REFERENCES
1) Schlegel PN, Sigman M, Collura B, et al. Diagnosis and treatment of infertility in men: AUA/ASRM Guideline Part I. *J Urol. 2021;205(1):36-43.*
2) Vasectomy Update 2010. *Can Urol Assoc J*. 2010 October; 4(5):306-309

RESULTS REPORTING - HEMATOLOGY

***REVISED** 12/26/2024*

HEM.36820 Reference Intervals **Phase II**

Patient results are reported with accompanying reference intervals or interpretive ranges.

NOTE: For WBC differential counts, the CAP recommends that laboratories report absolute cell counts, along with their corresponding reference intervals. The CAP discourages the reporting of percent cell counts without absolute counts on WBC differentials. Laboratories reporting only percent cell counts must provide laboratory established reference intervals.

Under some circumstances it may be appropriate to distribute lists or tables of reference intervals ~~to all~~(printed copies or electronic data) to users and sites where reports are received. ~~This system~~The laboratory must ensure that such data is ~~usually fraught with difficulties, but if in place and rigidly controlled, it is acceptable~~up to date.

Reference interval citations from the manufacturer's insert or published literature citations may be used to determine the reference interval. However, reference intervals have not been published for many body fluid analytes and obtaining normal fluids to establish reference intervals may not be feasible. If reference intervals are not available, results must be accompanied by an appropriate comment such as, "The reference interval(s) and other method performance specifications are unavailable for this body fluid. Comparison of the result with concentration in the blood, serum, or plasma is recommended."

Evidence of Compliance:
✓ Patient reports

REFERENCES
1) Trost DC, *et al*. Probability-based construction of reference ranges for ratios of log-Gaussian analytes: an example from automated leukocyte counts. *Am J Clin Pathol*. 2002;117:851-856
2) Clinical and Laboratory Standards Institute (CLSI). *Defining, Establishing, and Verifying Reference Intervals in the Clinical Laboratory - Approved Guideline-Third Edition.* CLSI Document EP28-A3c. Clinical and Laboratory Standards Institute, Wayne, PA; 2010.
3) Etzell, JE. For WBC differentials reporting absolute numbers. *CAP Today.* 2010; 3:12
4) Richardson-Jones A, Twedt D, Hellman R. Absolute versus proportional differential leukocyte counts. *Clin Lab. Haem.* 1995:17(2), 115-123

COAGULATION

SPECIMEN COLLECTION AND HANDLING - COAGULATION

***REVISED** 12/26/2024*

HEM.36880 Fill Volume and Specimen Mixing - Coagulation **Phase I**

Instructions for the acceptable fill volume and mixing of specimen collection tubes for coagulation testing are defined and followed.

NOTE: The recommended proportion of blood to the sodium citrate anticoagulant volume is 9:1. Inadequate filling of the collection device will decrease this ratio, and may lead to inaccurate results for calcium-dependent clotting tests, such as the PT and aPTT. The effect on clotting time from under-filled tubes is more pronounced when samples are collected in 3.8% rather than 3.2% sodium citrate. The effect of fill volume on coagulation results also depends on the reagent used for testing, size of the evacuated collection tube, and citrate concentration. A minimum of 90% fill is recommended; testing on samples with less than 90% fill should be validated by the laboratory. It is unacceptable to combine the contents from separate, underfilled sodium citrate collection tubes.

~~*Manufacturer's instructions for specimen mixing must be followed.*~~

Samples should be gently inverted to prevent clotting, in keeping with the manufacturer's instructions and laboratory's specimen collection instructions as described in GEN.40100.

Evidence of Compliance:
✓ Records of rejected specimens

REFERENCES
1) Peterson P, Gottfried EL. The effects of inaccurate blood sample volume on prothrombin time (PT) and activated partial thromboplastin time. *Thromb Haemost.* 1982;47:101-103
2) Adcock DM, Kressin D, Mariar PA. Minimum specimen volume requirements for routine coagulation testing. Dependence on citrate concentration. *Am J Clin Pathol.* 1998;109:595-599
3) Reneke J, *et al*. Prolonged prothrombin time and activated partial thromboplastin time due to underfilled specimen tubes with 109 mmol/L (3.2%) citrate anticoagulant. *Am J Clin Pathol.* 1998;109:754-757
4) Clinical and Laboratory Standards Institute (CLSI). *Collection, Transport, and Processing of Blood Specimens for Testing Plasma-Based Coagulation Assays ~~and Molecular Hemostasis Assays; Approved Guideline—Fifth Edition~~.* 6th ed. CLSI ~~Document~~guideline H21~~-A5 (ISBN 1-56238-657-3).~~ Clinical and Laboratory Standards Institute, ~~940 West Valley Road, Suite 1400,~~ Wayne, PA ~~19087-1898 USA, 2008.~~, 2024.

HEM.36940 Specimen Handling for Plasma-based Testing - Coagulation Phase II

Coagulation tests are promptly performed on fresh plasma, or the platelet-poor plasma is frozen until testing can be performed.

NOTE: After blood collection, there is progressive degradation of the labile coagulation factors V and VIII, leading to increasing prolongation of the aPTT and PT. The allowable time interval between specimen collection and sample testing depends on the temperature encountered during transport and storage of the specimen. Allowable time intervals are as follows:

1. *PT specimens, uncentrifuged or centrifuged with plasma remaining in the capped tube above the packed cells, or as centrifuged plasma separated from the cells, should be kept at room temperature (18 to 24°C) and tested no longer than 24 hours from the time of specimen collection. PT specimens should not be refrigerated (during storage).*
2. *aPTT specimens ~~that are~~, uncentrifuged or centrifuged with plasma remaining in the capped tube above the packed cells, should be kept at room temperature (18 to 24°C) and tested no longer than 4 hours after the time of specimen collection.*
3. *aPTT specimens that are centrifuged and plasma separated from cells can be kept for 4 hours refrigerated (2 to 8°C) or at room temperature (18 to 24°C). Samples for unfractionated heparin testing should be centrifuged within one hour from the time of specimen collection*
4. *Samples for other coagulation factors (eg, thrombin time, protein C, factor V, factor VIII) have variable stability and should be kept in the same manner as aPTT samples*

If PT or aPTT testing cannot be performed within these times, platelet-poor plasma should be removed from the cells and frozen at –20°C for up to 2 weeks or at –70°C for up to 12 months. If a laboratory has established an allowable time interval different than that detailed above, data must be available to verify that coagulation testing is valid in the time interval established.

REFERENCES
1) Clinical and Laboratory Standards Institute (CLSI). *Collection, Transport, and Processing of Blood Specimens for Testing Plasma-Based Coagulation Assays ~~and Molecular Hemostasis Assays; Approved Guideline—Fifth Edition~~.* 6th ed. CLSI ~~Document~~guideline H21~~-A5 (ISBN 1-56238-657-3).~~ Clinical and Laboratory Standards Institute, ~~940 West Valley Road, Suite 1400,~~ Wayne, PA ~~19087-1898 USA, 2008.~~, 2024.
2) Adcock DM, *et al*. The effect of time and temperature variables on routine coagulation tests. *Blood Coag Fibrinolysis.* 1998;9:463-470
3) Neofotistos D, *et al*. Stability of plasma for add-on PT and aPTT tests. *Am J Clin Pathol.* 1998;109:758-763
4) Davis KD, *et al*. Use of different thromboplastin reagents causes greater variability in international normalized ratio results than prolonged room temperature storage of specimens. *Arch Pathol Lab Med.* 1998;122:972-977

*****NEW** 12/26/2024***

HEM.36960 Specimen Handling for Whole Blood-Based Testing - Coagulation Phase II

Specimens for whole blood-based coagulation testing are handled according to manufacturer's instructions or as validated by the laboratory.

NOTE: Specimens must not be:
- *Heated, refrigerated, or frozen*
- *Centrifuged - Centrifuged specimens must be rejected. Reconstitution of a centrifuged specimen by mixing is not adequate.*

For additional specimen handling for platelet function studies, refer to HEM.38350.

COAGULATION STUDIES

COAGULATION FACTOR ASSAYS (EXCLUDING FIBRINOGEN BY IMMUNOLOGIC METHODS)

HEM.37980 Factor Assay Criteria **Phase II**

Three or more dilutions are plotted for each functional factor activity assay to detect non-parallelism and report non-parallelism if detected.

NOTE: This requirement does not apply to chromogenic factor assays or fibrinogen assays.

When performing factor assays, at least three dilutions of patient plasma in buffer are prepared either by the instrument or off the instrument. Multiple dilutions of test plasma are required to evaluate the extent of parallelism between test results and those of the reference plasma. This is necessary to be able to detect whether ~~a factor~~an inhibitor is present.

Criteria for demonstration of non-parallelism (or non-specific inhibitor effect) may vary between laboratories and instrument types. For example, in some laboratories, individual results of each dilution are reviewed and should agree within 20% of each other to be considered linear or parallel. In this instance, the average of all three results may be reported. Some coagulation instruments perform this determination automatically based on criteria programmed into the instrument.

Non-specific inhibitors often demonstrate a "dilution effect" characterized by non-parallelism of results with increasing dilutions. An example of non-parallel results is as follows: the 1:10 dilution yields 30% activity, the 1:20 dilution 50%, and the 1:40 dilution 75% activity. Further dilutions should be performed as needed and in accordance with the laboratory's practice and instrument capability, at least until the factor activity falls within the reference interval. In situations of non-parallelism, the highest value obtained with dilution should be recorded with a comment about dilution effect made in the laboratory report. In this instance, the mean result should not be reported nor should the value of the least dilute result.

Use of at least three patient dilutions enhances accuracy by minimizing dilutor error, and allows for detection of inhibitors or anticoagulants. To be valid, at least one value must fall within the upper and lower limits of the standard curve used for the calculation of the result.

The goal is to provide clinically useful data when a non-specific inhibitor activity is detected, (eg, a lupus anticoagulant or an anticoagulant drug like heparin). A comment like "inhibitor pattern detected" along with reporting the activity obtained at the highest dilution or over serial dilutions clarifies the result.

Functional fibrinogen assays may be subject to interference by certain anticoagulants; for guidelines on fibrinogen assays refer to COM.40500 and HEM.37165.

Evidence of Compliance:

✓ Records or worksheets showing patient data analyzed at three or more dilutions

REFERENCES
1) Clinical and Laboratory Standards Institute. *Determination of Coagulant Factor Activities Using the One-State Clotting Assay; Approved Guideline*. 2nd ed. CLSI document H48-ED2. Clinical and Laboratory Standards Institute, Wayne, PA, 2016.
2) Clinical and Laboratory Standards Institute. *Procedure for the Determination of Fibrinogen in Plasma; Approved Guideline;* 2nd ed. CLSI document H30-A2. CLSI, Wayne, PA, 2001.

PLATELET FUNCTION STUDIES

***REVISED** 12/26/2024*

HEM.38300 Platelet Function Studies **Phase II**

Platelet functional studies (platelet aggregation or initial platelet function test) are performed within an appropriate period after venipuncture.

NOTE: Following venipuncture, platelets continue to activate in vitro, so that platelet functionality becomes abnormal after a period of several hours. ~~The laboratory must ensure that platelet functional studies (platelet aggregation or initial platelet function test) are completed between 30 minutes and four hours from the time of phlebotomy, or erroneous results could be obtained.~~ Manufacturer's instructions for specimen stability must be followed for FDA-cleared/approved platelet function study assays~~.~~ (platelet function is generally not stable past four hours, although certain manufacturers may have more stringent requirements).

~~PRP (platelet rich plasma) should be used within three to four hours of platelet donation. The effects of time are related to changes in pH, which are directly related to the escape of CO_2 from the PRP sample tube. Platelets may be refractory to epinephrine when using PRP samples tested within 30 minutes of venipuncture; this is cited as the rationale for not testing PRP until at least 30 minutes after phlebotomy. There is evidence to suggest that this initial platelet refractoriness and subsequent gain of function occurs because centrifugation releases ADP from red blood cells and platelets. Specimens collected for whole blood aggregometry should be stored capped at room temperature and tested within four hours.~~

Evidence of Compliance:

✓ Records of testing completed within the defined time period

REFERENCES

1) Clinical and Laboratory Standards Institute (CLSI). *Platelet Function Testing by Aggregometry; Approved Guideline*. CLSI document H58-A. Clinical and Laboratory Standards Institute, 940 West Valley Road, Suite 1400, Wayne, PA 19087-1898 USA, 2008.
2) Winokur R, Hartwig JH. Mechanism of shape change in chilled human platelets. *Blood*. 1995; 85:1796-1804.
3) Mani H, Kitchmayr K, Klaffling C, et al. Influence of blood collection techniques on platelet function. *Platelets*. 2004;15(5):315-318.
4) Kattlove HE, Alexander B. The effect of cold on platelets. I. Cold-induced platelet aggregation. *Blood.* 1971;38(1):39-48.
5) Kattlove HE, Alexander B, White F. The effect of cold on Platelets. II. Platelet function after short-term storage at cold temperatures. *Blood*. 1972;40(5):688-695.