If the nontreponemal (lipoidal antigen) antibody test is performed to monitor treatment of patients with known syphilis infection (not as a screening tool), anti-treponemal antibody testing is not required. Because anti-treponemal antibodies persist after successful treatment, testing patients with previously diagnosed syphilis using a reverse algorithm approach is discouraged; therefore, laboratories should provide a clear option for providers to order nontreponemal (lipoidal antigen) titers directly for following serologic response to treatment.

This checklist requirement only applies to testing serum/plasma specimens. For testing CSF specimens, stand-alone anti-treponemal (eg, FTA-ABS or TPPA) and/or nontreponemal (lipoidal antigen) (eg, VDRL) tests may be used at the discretion of the laboratory director.

Evidence of Compliance:

✓ Records of confirmatory testing of positive screening antibody results with appropriate secondary assays

REFERENCES

1) Papp JR, Park IU, Fakile Y, Pereira L, Pillay A, Bolan GA, CDC laboratory recommendations for syphilis testing, United States, 2024. *MMQR Recomm Rep.*2024;73 (No. RR-1): 1-32.
2) Rhoads DD, et al. Prevalence of traditional and reverse-algorithm syphilis screening in laboratory practice. A survey of participants in the College of American Pathologists syphilis serology proficiency testing program. *Arch Pathol Lab Med.* 2017;141(1):93-97.

HIV PRIMARY DIAGNOSTIC TESTING

Inspector Instructions:

READ

- Sampling of HIV diagnostic testing policies and procedures
- Sampling of HIV result reports

IMM.41450 HIV Primary Diagnostic Testing - Supplemental and Confirmatory Testing Phase I

The laboratory follows public health recommendations or guidelines for HIV primary diagnostic testing, including primary screening and additional (supplemental and/or confirmatory) testing.

NOTE: If additional testing after a primary screening test is recommended by public health authorities, the laboratory:

- *Performs additional testing reflexively if the specimen is suitable and the test is performed in house, or*
- *Sends additional testing to a referral laboratory if the specimen is suitable, or*
- *Provides guidance to providers on submission of additional specimens, if needed for supplemental or confirmatory testing.*

The US Centers for Disease Control and Prevention (CDC) and Association of Public Health Laboratories (APHL) provide recommendations for HIV testing. Guidelines and recommended algorithms can be found on the CDC and APHL websites.

Evidence of Compliance:

✓ Patient reports with initial screening results and reflexive testing results and/or guidance

REFERENCES

1) Centers for Disease Control and Prevention and Association of Public Health Laboratories. Laboratory Testing for the Diagnosis of HIV Infection: Updated Recommendations. Available at http://stacks.cdc.gov/view/cdc/23447. Published June 27, 2014. Accessed 11/19/2019.
2) National Center for HIV/AIDS, Viral Hepatitis, and TB Prevention (US). Divisions of HIV/AIDS Prevention; Association of Public Health Laboratories. 2018 Quick reference guide: Recommended laboratory HIV testing algorithm for serum or plasma specimens, Available at: https://stacks.cdc.gov/view/cdc/50872. Published January 2018. Accessed 4/2/2023.

3) Association of Public Health Laboratories. Suggested Reporting Language for the HIV Laboratory Diagnostic Testing Algorithm. January 2019. Available at APHL Publications. Accessed 11/19/2019.

WESTERN BLOT ASSAYS

Inspector Instructions:

- Sampling of western blot policies and procedures

IMM.41500 Molecular Weight Markers **Phase II**

Known molecular weight markers are included and reviewed with each Western blot assay of patient samples.

Evidence of Compliance:

✓ Records of QC for known markers

IMM.41600 Western Blot Separations **Phase II**

Western blot separations are satisfactory with sufficient resolution (low background, clear signal, absence of bubbles, etc.) to interpret band size easily.

IMM.41700 Acceptable Limits - Controls **Phase II**

Acceptable limits are set for controls of procedures where the Western blot bands are quantified.

NOTE: The criterion to designate a Western blot test as positive is based on the detection of a certain combination of positive bands. The laboratory should define a minimum intensity that allows a band to be considered positive.

Evidence of Compliance:

✓ Records of defined acceptable limits for control range of each lot

IMM.41800 Interpretation **Phase II**

Objective criteria are defined for interpretation of Western blot.

REFERENCES

1) Clinical and Laboratory Standards Institute (CLSI). *Criteria for Laboratory Testing and Diagnosis of Human Immunodeficiency Virus Infection.* 2nd ed. CLSI guideline M53. Clinical and Laboratory Standards Institute, Wayne, PA; 2023.
2) Clinical and Laboratory Standards Institute. *Western Blot Assay for Antibodies to Borrelia burgdorferi; Approved Guideline*. CLSI document M34-A. CLSI, Wayne, PA, 2000.
3) Engstrom SM, Shoop E, Johnson RC. Immunoblot interpretation criteria for serodiagnosis of early Lyme disease. *J Clin Microbol* 1995;33:419-22
4) Dressler F, Whelan JA, Reinhart BN, Steere AC. Western blotting in the serodiagnosis of Lyme disease. *J Infect Dis* 1993;167:392-400