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Labstract – November 2020

Syphilis *(Treponema pallidum)* Serologic Testing Update - Changes to Rapid Plasma Reagin (RPR) Confirmatory Test and Algorithm

Audience

Healthcare providers who order syphilis serology testing.

Overview

Effective November 2020:

- Public Health Ontario's (PHO) laboratory is changing the syphilis confirmatory serology testing methodology on serum from manual Rapid Plasma Reagin (RPR) testing to an automated RPR test system utilizing the Gold Standard AIX1000 RPR analyzer.
- PHO's laboratory follows the reverse syphilis serologic testing algorithm. Currently a treponemal test, Chemiluminescent Micro-particle Immunoassay (CMIA) is used as the screening test followed by both a non-treponemal test (RPR) and a treponemal test, Treponema pallidum particulate agglutination (TPPA) for confirmation. PHO's laboratory is changing its syphilis confirmatory algorithm by performing RPR first followed by TPPA only for those samples that test RPR non-reactive.

Background Information

Syphilis is a disease caused by infection with the bacterium *Treponema pallidum* (TP). Route of transmission is primarily through sexual contact, but it can also be transmitted from mother to fetus, or rarely, through blood and blood product and/or organ transplant. Syphilis typically follows a progression of stages including primary, secondary, latent and rarely tertiary stages that can last for weeks, months or even years.
Serologic testing is the primary method for routine diagnosis and monitoring of treatment.

Change to Syphilis RPR Confirmatory Testing

As the number of syphilis cases continues to rise, the need to fully automate all steps in the **syphilis testing** algorithm increases, and RPR testing has become an excellent candidate for **lab automation**.

The Gold Standard Diagnostics AIX1000 Rapid Plasma Reagin (RPR) Automated Assay is a non-treponemal flocculation test for the qualitative and semi-quantitative determination of reagin antibodies in human serum or plasma to aid in the diagnosis of syphilis.

The advantages of the automated RPR system include:

- Results are interpreted by pattern recognition software which is objective and consistent
- Complete traceability from sample to result
- Archived images of results are linked to samples

Results will be reported as either 'Reactive', 'Non-reactive' or 'Invalid' for the detection of reagin antibodies.
As per the studies conducted by the manufacturer, precision and reproducibility are at 98.8 % and 100% respectively.

Change to the Syphilis Serology Test Algorithm

An initial screening (CMIA) with a treponemal serology test is followed by a non-treponemal Rapid Plasma Reagin (RPR) test. If RPR test fails to confirm a reactive screening result, a treponemal test, *Treponema pallidum* Particle Agglutination (TPPA) is performed. Samples from patients with previously confirmed TPPA results will be excluded from testing.

Interpretation of the Most Common Results Using the Revised Syphilis Algorithm

Screening Test (CMIA)	Confirmatory Test (RPR)	Confirmatory Test (TPPA)	Possible Interpretations/ Recommendations
Non-reactive	Not tested	Not tested	No confirmatory testing is performed if syphilis screen result is non-reactive • Early incubating syphilis can be non-reactive before antibodies have developed. • If clinical suspicion of early syphilis, suggest single repeat serology in 4 weeks if not repeated already.
Reactive	Reactive	Not tested	Consistent with recent or prior syphilis infection
Reactive	Non- reactive	Reactive	Consistent with recent or prior syphilis infection
Reactive	Non- reactive	Non- Reactive	• Results consistent with false reactive screening test. • Rare alternate interpretations include early syphilis, previously treated, or late latent syphilis. • Repeat serology in 4 weeks if not already repeated.
Reactive	Non-reactive	Indeterminate	Inconclusive syphilis serology results • Possible interpretations include false positive, or early, old treated or untreated syphilis. • Repeat serology in 4 weeks if not already repeated.

Screening Test (CMIA)	Confirmatory Test (RPR)	Confirmatory Test (TPPA)	Possible Interpretations/ Recommendations
Reactive	Reactive	Indeterminate	Consistent with recent or prior syphilis infection
Reactive	Invalid	Not tested	Inconclusive syphilis serology results • Advise Follow-up sample
Age < 12 Months Reactive	Reactive		• Maternal antibody (can be present in infant for up to 12 months) • Congenital infection • If congenital or early syphilis is suspected, consider ordering repeat serology at the recommended intervals according to the PHAC Canadian Guidelines on Sexually Transmitted Infections, Section 5-10, Table 8(b) (see references)
Age < 12 Months Reactive	Non- reactive	Reactive	• Maternal antibody (can be present in infant for up to 12 months) • Does not rule out congenital infection • If congenital or early syphilis is suspected, consider ordering repeat serology at the recommended intervals according to the PHAC Canadian Guidelines on Sexually Transmitted Infections, Section 5-10, Table 8(b) (see references)

Specimen Collection Requirements

Human serum is acceptable for syphilis serology testing. Whole blood should be allowed to clot. Serum separator tubes (SST) are acceptable. Liquid anticoagulants may have a dilution effect resulting in lower concentrations for individual patient specimens. Heat inactivated, haemolysed, icteric, lipemic or microbially contaminated sera are not recommended for testing.

Note: This document does not apply to testing for syphilis in primary lesions and cerebrospinal fluid (CSF). Syphilis testing information for primary lesions and CSF is available at:
publichealthontario.ca/en/ServicesAndTools/LaboratoryServices/Pages/Syphilis_Chancre_Direct_Fluorescence.aspx;
publichealthontario.ca/en/ServicesAndTools/LaboratoryServices/Pages/Syphilis_CSF.aspx

Testing Turnaround Time (TAT)

TAT may be up to 6 days.

References

1. Centers for Disease Control and Prevention. Sexually transmitted disease surveillance 2014 cdc.gov/std/stats14/ (Accessed on February 06, 2017)

2. Hicks CB, Clement M. Syphilis: Screening and diagnostic testing. In: UpToDate, Hynes NA, Mitty J (Ed), UpToDate, Waltham, MA. (Accessed on April 03, 2017)

3. Levett PN, Fonseca K, Tsang RSW, et al. Canadian Public Health Laboratory Network laboratory (CPHLN) guidelines for the use of serological tests (excluding point-of-care tests) for the diagnosis of syphilis in Canada. Can J Infect Dis Mid Microbiol 2015;26(Suppl A):6A-12A.

4. PHAC Canadian Guidelines on Sexually Transmitted Infections; Section 5-10: Management and Treatment of Specific Infections, Table 8(b) at canada.ca/en/public-health/services/infectious-diseases/sexual-health-sexually-transmitted-infections/canadian-guidelines/sexually-transmitted-infections/canadian-guidelines-sexually-transmitted-infections-27.html

For further information

- Contact the PHO Laboratory Customer Service Centre at 416-235-6556 or 1-877-604-4567 (toll-free), or by email at customerservicecentre@oahpp.ca
- For PHO Laboratory specimen collection information and previous Labstracts, refer to publichealthontario.ca/test directory
- The current version of the PHO Laboratory General Test Requisition and other forms are available at publichealthontario.ca/Requisitions
- To subscribe to future Labstracts, register on our website
- To register for Autofax and receive laboratory reports by fax directly from our laboratory information system as soon as they are released, contact the PHO Laboratory Customer Service Centre.