MAYO CLINIC LABORATORIES

Test Definition: RPRS

Rapid Plasma Reagin Screen with Reflex, Serum

Overview

Useful For

Aiding in the diagnosis of recent or past *Treponema pallidum* infection

Rapid plasma reagin screening when *T pallidum* antibody screen is positive

This test is **not useful** as a screening or confirmatory test for blood donor specimens.

Testing Algorithm

If the rapid plasma reagin (RPR) screen is reactive, then the RPR titer will be performed at an additional charge.

If the RPR screen is nonreactive, then syphilis antibody *Treponema pallidum* particle agglutination testing will be performed at an additional charge.

See Syphilis Serology Algorithm

Special Instructions

- Syphilis Serology Algorithm

Method Name

Only available as a reflex test. For more information, see SYPHT / Syphilis Total Antibody with Reflex, Serum.

RPRS: Multiplex Flow Immunoassay
RRPRQ: Flocculation/Agglutination
RTPPA: Particle Agglutination

NY State Available

No

Specimen

Specimen Type

Serum

Specimen Required

Only available as a reflex test. For more information see SYPHT / Syphilis Total Antibody with Reflex, Serum.

Container/Tube:
Preferred: Serum gel

Acceptable: Red top
Submission Container/Tube: Plastic vial
Specimen Volume: 0.5 mL
Collection Information: Centrifuge and aliquot serum into plastic vial.

Specimen Minimum Volume
0.4 mL

Reject Due To

Gross hemolysis	Reject
Gross lipemia	Reject
Heat-inactivated specimen	Reject

Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Serum	Refrigerated (preferred)	14 days	
	Frozen	14 days	

Clinical & Interpretive

Clinical Information
Syphilis is a disease caused by infection with the spirochete *Treponema pallidum*. The infection is systemic, and the disease is characterized by periods of latency. These features, together with the fact that *T pallidum* cannot be isolated in culture, mean that serologic techniques play a major role in the diagnosis and follow-up of treatment for syphilis.

Historically, the serologic testing algorithm for syphilis included an initial non-treponemal screening test, such as the rapid plasma reagin (RPR) or VDRL tests. Because these tests measure the host's antibody response to non-treponemal antigens, they lack specificity. Therefore, a positive result by RPR or VDRL requires confirmation by a treponemal-specific test, such as the fluorescent treponemal antibody-absorbed (FTA-ABS) or microhemagglutination assay (MHA-TP). Although the FTA-ABS and MHA-TP are technically simple to perform, they are labor intensive and require subjective interpretation by testing personnel.

As an alternative to the traditional syphilis screening algorithm as described above, many laboratories utilize the reverse syphilis screening algorithm. This algorithm starts with an automated treponemal assay, such as an enzyme immunoassay and multiplex flow immunoassay (MFI), to detect antibodies specific to *T pallidum*. If the screening assay is positive, the sample is reflexed to a RPR assay, which, if positive, is reported with a titer and is indicative of active or recent syphilis infection. If the RPR is negative, the sample is reflexed to a second treponemal assay, such as the *T pallidum* particle agglutination (TP-PA) assay. If the TP-PA is positive, this would indicate previously treated or late stage syphilis infection. Alternatively, if the TP-PA is negative, the initial positive screen is interpreted as a false positive result.

Syphilis screening at Mayo Clinic is performed by using the reverse algorithm, which first tests sera for *T pallidum* specific IgG/IgM antibodies using an automated MFI. A positive treponemal test suggests infection with *T pallidum*, but does not distinguish between recent or past, or treated and untreated infection. This is because treponemal tests may remain reactive for life, even following adequate therapy. Therefore, the results of a non-treponemal assay, such as RPR, are needed to provide information on a patient's disease state and history of therapy.(Table 1)

In some patients, the results of the treponemal screening test and RPR may be discordant (eg, syphilis IgG/IgM positive and RPR negative). To discriminate between a falsely reactive screening result and past syphilis, a second treponemal-specific antibody test is recommended using a method that is different from the initial screen test (eg, -TP-PA).

In the setting of a positive syphilis IgG/IgM screening result and a negative RPR, a positive TP-PA result is consistent with either 1) past, successfully treated syphilis, 2) early syphilis with undetectable RPR titers, or 3) late/latent syphilis in patients who do not have a history of treatment for syphilis. Further historical evaluation is necessary to distinguish between these scenarios.(Table 1)

In the setting of a positive syphilis IgG/IgM screening result and a negative RPR, a negative TP-PA result is most consistent with a falsely reactive syphilis IgG/IgM screen.(Table 1) If syphilis remains clinically suspected, a second specimen should be submitted for testing.

Table 1. Interpretation and follow-up of reverse screening results:

		Test and result			
Patient history	**Syphilis total antibody by MFI**	**RPR**	**TP-PA**	**Interpretation**	**Follow-up**
Unknown history of syphilis	Nonreactive	NA	NA	No serologic evidence of syphilis	None, unless clinically indicated (eg, early/acute/ primary syphilis)
Unknown history of syphilis	Reactive	Reactive	NA	Untreated or recently treated syphilis	See CDC treatment guidelines
Unknown history of syphilis	Reactive	Nonreactive	Nonreactive	Probable false-positive screening test	No follow-up testing, unless clinically indicated (eg, acute/ primary syphilis)
Unknown history of syphilis	Reactive	Nonreactive	Reactive	Possible syphilis (eg, early or latent) or previously treated syphilis	Historical and clinical evaluation required
Unknown history of syphilis	Equivocal	NA	NA	NA	Unknown history of syphilis
Known	Reactive	Nonreactive	Reactive or	Past, successfully treated	None

history of syphilis			NA	syphilis	

MFI - multiplex flow immunoassay
NA - not applicable
RPR - rapid plasma reagin
TP-PA - *Treponema pallidum* particle agglutination

Reference Values

Only available as a reflex test. For more information see SYPHT / Syphilis Total Antibody with Reflex, Serum.

Nonreactive

Interpretation

Nonreactive:
Treponema pallidum-particle agglutination (TP-PA) has been ordered to distinguish between infection with *T pallidum* (syphilis) versus a falsely reactive treponemal antibody result

Reactive:
Specimen reflexed to determine rapid plasma reagin (RPR) titer value

Cautions

Despite active syphilis, serologic tests may be negative in severely immunosuppressed patients such as those with AIDS.

In very early cases of primary syphilis, serology tests for syphilis may be negative.

In cases of untreated, late, or latent syphilis, the result of rapid plasma reagin may be negative. However, the syphilis screening test multiplex flow immunoassay and *Treponema pallidum* particle agglutination should be positive. A thorough clinical and historical evaluation should be performed to determine if treatment for latent syphilis is required.

Results should be considered in the context of all available clinical and laboratory data.

Clinical Reference

1. Centers for Disease Control and Prevention (CDC). Discordant results from reverse sequence syphilis screening-five laboratories, United States, 2006-2010. MMWR Morb Mortal Wkly Rep. 2011;60(5):133-137
2. Radolf JD, Tramont EC, Salazar JC: Syphilis *(Treponema pallidum*). In: Bennett JE, Dolin R, Blaser MJ, eds. Mandell, Douglas, and Bennett's Principles and Practice of Infectious Diseases. 9th ed. Elsevier; 2020:2865-2892
3. Binnicker MJ, Jespersen DJ, Rollins LO: Direct comparison of the traditional and reverse syphilis screening algorithms in a population with a low prevalence of syphilis. J Clin Microbiol. 2012 Jan;50(1):148-150. doi: 10.1128/JCM.05636-11

Performance

Method Description

The BioPlex 2200 Syphilis Total and RPR kit employs *Treponema pallidum* fusion protein (rTP47/rTP17) and cardiolipin antigen-coated fluoromagnetic beads with unique fluorescent signatures to identify the presence of IgG and IgM antibodies to *T pallidum* and nontreponemal reagin antibodies in a 2-step assay format. Dyed beads are coated with recombinant *T pallidum* rTP47/rTP17 fusion protein or cardiolipin antigen. The BioPlex 2200 System combines an aliquot of patient sample, sample diluent, and bead reagent into a reaction vessel. The mixture is incubated at 37 degrees C. After a wash cycle, a mixture of murine monoclonal anti-human IgG and murine monoclonal anti-human IgM antibody conjugated to phycoerythrin (PE) is added to the dyed beads, and this mixture is incubated at 37 degrees C. The excess conjugate is removed in another wash cycle, and the beads are re-suspended in wash buffer. The bead mixture then passes through the detector. The identity of the dyed beads is determined by the fluorescence of the dyes, and the amount of antibody captured by the antigen is determined by the fluorescence of the attached PE. Raw data is calculated in relative fluorescence intensity (RFI).(Package insert: BioPlex 2200 Syphilis Total and RPR. Bio-Rad; 06/2017)

If the total antibody result is reactive, a rapid plasma reagin (RPR) screen is performed on the BioPlex 2200. If the RPR screen is reactive the RPR titer test is performed. The RPR titer test is a macroscopic screening assay done with unheated serum. Reagin reacts with nontreponemal antigen containing colloidal charcoal particles. This reaction results in a visual flocculation of the black particles against the white card background. The test yields a positive or negative result, and all positive samples are titered to determine the highest positive dilution.(Huber TW, Storms S, Young P, et al: Reactivity of microhemagglutination, fluorescent treponemal antibody absorption, Venereal Disease Research Laboratory, and rapid plasma reagin tests in primary syphilis. J Clin Microbiol. 1983 Mar;17[3]:405-409; Kaur G, Kaur P: Syphilis testing in blood donors: an update. Blood Transfus. 2015 Apr;13[2]:197-204)

If the RPR screen is negative, the Serodia *T pallidum* particle agglutination (TP-PA) test is performed. The TP-PA test is based on the agglutination of colored gelatin particle carriers sensitized with *T pallidum* (Nichols Strain) antigen. Serum samples are serially diluted in microplate wells. Sensitized gelatin particles are added to respective wells and the contents of the plate mixed. The mixture is incubated for 2 hours at room temperature. Serum containing specific antibodies will react with the antigen-sensitized colored gelatin particles to form a smooth mat of agglutinated particles in the microplate well. A compact button formed by the settling of the non-agglutinated particles characterizes negative reactions. The agglutination patterns are read visually to determine interpretation.(Package insert: Serodia TP-PA. Fujirebio Diagnostics, Inc; 04/2015)

PDF Report
No

Day(s) Performed
Monday through Saturday

Report Available
Same day/1 to 4 days

Specimen Retention Time
14 days

Performing Laboratory Location
Mayo Clinic Jacksonville Clinical Lab

Fees & Codes

Fees

- Authorized users can sign in to Test Prices for detailed fee information.
- Clients without access to Test Prices can contact Customer Service 24 hours a day, seven days a week.
- Prospective clients should contact their account representative. For assistance, contact Customer Service.

Test Classification

This test has been cleared, approved, or is exempt by the US Food and Drug Administration and is used per manufacturer's instructions. Performance characteristics were verified by Mayo Clinic in a manner consistent with CLIA requirements.

CPT Code Information

86593-Rapid Plasma Reagin Titer (if appropriate)
86780-Syphilis Antibody by TP-PA (if appropriate)

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
RPRS	RPR Screen w/ Reflex, S	20507-0

Result ID	Test Result Name	Result LOINC® Value
RPRS	RPR Screen w/ Reflex, S	20507-0