# Test Definition: NSYPH

Syphilis IgG Enzyme Immunoassay, Serum

## Overview

### Useful For

An aid in the diagnosis of infection with *Treponema pallidum*

Routine prenatal screening

This test is **not useful** for diagnosis of congenital syphilis.

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

### Highlights

This testing should be used to assess for recent or past infection with *Treponema pallidum* or for routine prenatal screening.

Testing for syphilis is performed using the reverse screening algorithm at Mayo Clinic and Mayo Clinic Laboratories.

### Method Name

Enzyme Immunoassay (EIA)

### NY State Available

Yes

## Specimen

### Specimen Type

Serum

### Specimen Required

**Collection Container/Tube:** Serum gel
**Acceptable:** Red top
**Submission Container/Tube:** Plastic vial
**Specimen Volume:** 0.6 mL
**Collection Instructions:** Centrifuge and aliquot serum into plastic vial.

### Forms

If not ordering electronically, complete, print, and send Infectious Disease Serology Test Request (T916) with the specimen.

### Specimen Minimum Volume

0.5 mL

### Reject Due To

| | |
|---|---|
| Gross hemolysis | Reject |
| Gross lipemia | Reject |
| Heat-inactivated specimen | Reject |

### Specimen Stability Information

| Specimen Type | Temperature | Time | Special Container |
|---|---|---|---|
| Serum | Frozen | 14 days | |

## Clinical & Interpretive

### Clinical Information

Syphilis is caused by infection with the spirochete *Treponema pallidum* subspecies *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 the VDRL tests. Because these tests measure the host's immune response to nontreponemal 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-absorption (FTA-ABS) or microhemagglutination (MHA-TP) assay. Although the FTA-ABS and MHA-TP assays 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, many laboratories utilize the reverse syphilis screening algorithm. This algorithm starts with an automated treponemal assay to detect antibodies specific to *T pallidum*. If this screening assay is positive, the sample is reflexed for testing by RPR, 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 using the reverse algorithm, which first tests sera for *T pallidum* specific IgG antibodies using an automated enzyme immunoassay. A positive treponemal test suggests infection with *T pallidum* but does not distinguish between recent, past, treated, or untreated infection. This is because treponemal tests may remain reactive for life, even following adequate therapy. Therefore, the results of a nontreponemal assay, such as RPR, are needed to provide information on a patient's disease state and history of therapy.(Table)

In some patients, the results of the treponemal screening test and RPR may be discordant (eg, syphilis IgG 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 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, 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)

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

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

| | | Test and result | | | |
|---|---|---|---|---|---|
| **Patient history** | **Syphilis IgG Ab by EIA** | **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 Centers for Disease Control and Prevention 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 history of syphilis | Reactive | Nonreactive | Reactive or NA | Past, successfully treated syphilis | None |
| EIA, enzyme immunoassay; NA, not applicable; RPR, rapid plasma reagin; TP-PA, *Treponema pallidum* particle agglutination | | | | | |

## Reference Values

Nonreactive
Reference values apply to all ages

### Interpretation

Nonreactive:
No serologic evidence of exposure to *Treponema pallidum* (syphilis). Repeat testing may be considered in patients with suspected acute or primary syphilis.

Equivocal:
Recommend follow-up testing in 10 to 14 days if clinically indicated.

Reactive:
Results suggest infection with *T pallidum* at some point in time. Results do not distinguish between recent or past infection, or between treated and untreated syphilis as treponema-specific IgG may remain elevated despite appropriate therapy. Falsely reactive treponemal results may occur; additional testing by a non-treponemal assay is recommended if not previously performed on this sample.

### 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 may be negative.

In cases of untreated, late or latent syphilis, the result of rapid plasma reagin may be negative. However, the syphilis screening enzyme immunoassay (EIA) and *Treponema pallidum* particle agglutination (TP-PA) 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. Morb Mortal WKLY Rep. 2011 Feb 11;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; 2020:2865-289291
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

## Performance

### Method Description

Microtitration wells, coated with whole-cell sonicated *Treponema pallidum* (Nichols strain) antigens, are incubated with serum specimens which may contain specific antibodies to *T pallidum*. After incubation, unbound components in the test sample are washed away. IgG antibodies from the specimen that bound to *T pallidum* antigens are detected using monoclonal anti-human IgG secondary antibodies conjugated to horseradish peroxidase (HRP) during a second incubation period. Following a second wash cycle, the enzyme conjugate on bound secondary antibodies is detected

following addition of tetramethylbenzidine (TMB). The enzymatic reaction is stopped using 1 N sulfuric acid. The assay is measured spectrophotometrically to indicate the presence or absence of IgG treponemal antibodies relative to a cut-off calibrator.(Package insert: CAPTIA Syphilis (*T Pallidum*)-G. Trinity Biotek USA; 800-970-29 Rev H, 10/2013)

### PDF Report
No

### Day(s) Performed
Monday through Friday, Sunday

### Report Available
Same day/1 to 3 days

### Specimen Retention Time
14 days

### Performing Laboratory Location
Mayo Clinic Laboratories - Rochester Superior Drive

## 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
86780

### LOINC® Information

| Test ID | Test Order Name | Order LOINC® Value |
|---|---|---|
| NSYPH | Syphilis IgG EIA, S | 47238-1 |

| Result ID | Test Result Name | Result LOINC® Value |
|---|---|---|
| NSYPH | Syphilis IgG EIA, S | 47238-1 |