Cleveland Clinic

Cleveland Clinic Laboratories

# Syphilis Serological Testing

## Background Information

Syphilis testing can be divided into two categories: Treponemal assays (specific) measure antibodies that directly react with the syphilis-causing organism *T. pallidum subsp. pallidum*, while non-tremponemal assays, such as RPR testing, measure antibodies against non-specific antigens, including cardiolipin, lecithin, and cholesterol, released during treponemal infections.

In the traditional testing algorithm for diagnosing syphilis, patient serum is initially tested with a non-treponemal test, followed by confirmation with a specific/confirmatory treponemal test. This algorithm was popular because of the technical ease of performing the RPR relative to fluorescent treponemal antibody absorption (FTA-ABS) or *Treponema pallidum* particle agglutination (TP-PA) assays. However, because the RPR test does not recognize treponemal-specific antibodies, a number of clinical situations could result in false-positive RPR results, including autoimmune diseases,[1] acute or chronic viral infections, recent immunizations, pregnancy, or drug addiction.[2–3] Most importantly, because RPR reactivity is a feature of active syphilis infection, the test could give false negative results in latent or late syphilis. It may also test non-reactive during primary syphilis, potentially leading to mother-to-child transmission. Therefore testing pregnant women using the traditional algorithm may culminate in catastrophic consequences.

The CDC recognizes another testing algorithm – the reverse algorithm – in which the patient's blood is initially tested using a specific treponemal test and confirmed with a non-treponemal test.[4] The algorithm shown here represents Cleveland Clinic's recommended screening for syphilis serology testing.

## Clinical Indications

A reactive syphilis total/screen test result with a non-reactive RPR and confirmed by EIA (see algorithm), indicates that a person has been exposed to *T. pallidum subsp. pallidum* at some point in their life. However, this testing may remain reactive for life in the majority of people who have had syphilis, even if they have received appropriate treatment. Therefore, a positive result does not indicate that the person currently has untreated syphilis and the result should be confirmed with a non-treponemal test, such as RPR, to assess current disease activity. That being said, during the first few weeks post-infection, both treponemal assays may test positive while RPR remains non-reactive.

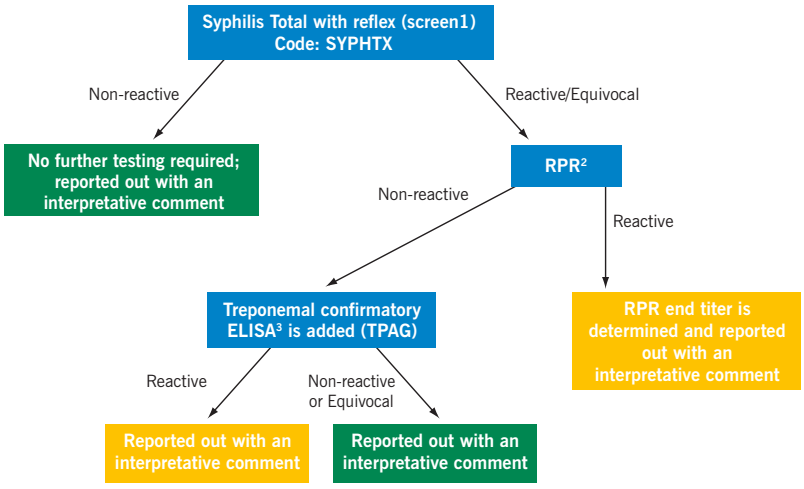
Most patients become seronegative on non-treponemal tests following adequate treatment; however, some patients have a low RPR titer for extended periods when they present with late latent or tertiary disease, despite being adequately treated in the past.[5] These patients are referred to as being "serofast."

Venereal disease research laboratory (VDRL) testing is used for diagnosis of neurosyphilis, otosyphilis, and ocular syphilis using CSF specimens. It is not included in this algorithm and must be ordered separately where clinically warranted.

## Limitations of the Assay

- Infants up to 18 months may have a reactive syphilis total/screen test result. This may also be seen with RPR; however, it shows faster clearance kinetics and usually

**Recommended Algorithm for Syphilis Serology Testing**



disappears in 4-6 weeks postnatally only where there is no congenital syphilis. The best approach would be to compare maternal and neonatal RPR titers collected at the same time, and where there is at least 4-fold higher titer seen in the neonate, it should significantly raise suspicion for congenital syphilis. It is still important to order SYPHTX to ensure the reactive RPR results are in the context of the reactive Treponemal result.

- Samples with very high antibody concentrations may produce false negative results for the RPR test due to the prozone effect. This has always remained a concern among clinicians, however, in the lab, the so-called "rough" RPR results are blindly diluted to rule out prozone phenomenon. Therefore such possibility remains exceedingly rare.

## Methodology

Multiplex flow immunoassay (MFIA) method for syphilis Total Ab. Agglutination method for RPR and enzyme immunoassay for the confirmatory assay (TPAG).

## References


1. Catteral RD. Collagen disease and the chronic biological false positive phenomenon. *QJ Med*.1961;117:41.
2. Harris A, Brown L, Portnoy J, Price EV. Narcotic addiction and BFP reactions in tests for syphilis. *Public Health Rep.* 1962;77:537.
3. Kaufman RE, Weiss S, Moore JD. Biologic false positive serological tests for syphilis among drug addicts. *Brit J Vener Dis.* 1974;50:350.
4. Pope, V., Use of Syphilis Test to Screen for Syphilis. *Infect Med.* 2004;21(8):399-404.
5. Pettit DE, Larsen SA, Harbec PS. Toluidine red unheated serum test, a non-treponemal test for syphilis. *J Clin Micro.* 1983;18:1141.
6. CDC, MMWR, Vol. 60 (5):133-140, 2011.


## Test Overview

| | |
|---|---|
| **Test Name** | Syphilis Total with reflex |
| **Ordering Mnemonic** | SYPHTX |
| **Reference Range** | Nonreactive |
| **Patient Preparation** | None |
| **Specimen Requirements** | 1.0 mL serum |
| **Test Ordering Information** | SYPHTX |
| **Reflex Information** | If Syphilis Total is Equivocal/Reactive, RPR and/or TPAG may be ordered and billed, depending on the algorithm. |
| **CPT Codes** | 86780 |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