Interpretation of Serologic Tests for Syphilis

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INTERPRETATION OF SEROLOGICAL TESTS FOR SYPHILIS

Proper evaluation of serological test results in syphilis is often a confusing matter. Many physicians are confronted only rarely with problems in this regard. The following comments are provided as a matter of review. More extensive discussion may be found in the references listed at the end of this article. Several tests are in use. Most common of these are the RAPID PLASMA REAGIN CARD (RPRC) TEST, the VENEREAL DISEASE RESEARCH LABORATORY (VDRL) TEST, the MICROHEMAGGLUTINATION for TREPONEMA PALLIDUM (MHA-TP) TESTS and the FLUORESCENT TREPONEMAL ANTIBODY ABSORPTION (FTA-ABS) TEST.

Nontreponemal Antigen Tests

Nontreponemal antigen tests, including the RPRC and VDRL Tests, are designed to test serum for reagin, a heterogeneous group of antibodies which combine with a cardiolipin-lecithin antigen. This antigen, usually derived from beef heart, is a normal component of human tissue. The RPRC and VDRL Tests are sometimes described as standard tests for syphilis (STS).

The RPRC Test is quick, inexpensive, and easy to use; it is exceptionally useful in screening. Like its older nontreponemal counterpart, the VDRL, it may be quantitated. These tests are very sensitive; i.e., there are few false negatives, except as discussed below.

RPRC quantitative tests are reported as "nonreactive" or "reactive" at dilutions of 1:1, 1:2, 1:4, 1:8, 1:16, 1:32, 1:64, etc. While considerable confidence may be placed in the RPRC or VDRL Tests, it must be remembered that patients with active infectious primary syphilis frequently have nonreactive VDRL and RPRC Tests. This is a result of the patient's immune system lagging behind the disease process. At the time of appearance of the primary lesion, the chancre, only about 25% of the cases will have a reactive VDRL or RPRC Tests. After the chancre has been present for one week, some 50% will have a reactive test; after two weeks, the ratio rises to 75%. Almost all will have reactive serologies 3-4 weeks after appearance of the primary lesion; and virtually 100% of the cases will have a reactive serology by the time secondary syphilis develops. Although the development of secondary syphilis may take as long as six months, the rash or other signs may appear even before the primary lesion has "healed." Treatment with penicillin and other antibiotics will interrupt the development of subsequent stages of the disease if the dosage is adequate. If the dosage is NOT adequate, the development of clinical signs may be disrupted to some extent or delayed temporarily; the development of seroreactivity to the RPRC or VDRL Tests may also be masked or delayed by inadequate treatment.

Prior to treatment of any patient for syphilis, a quantitative VDRL or RPRC should be performed. The test should be repeated three months after therapy and on several occasions over the next two years—see Table.

Individuals who are recent contacts (within 90 days) of patients with infectious syphilis (primary, secondary, and early latent stages) and who have negative RPRC or VDRL findings still must receive prophylactic treatment for syphilis as an epidemiologic control measure.

In untreated primary syphilis, the seroreactivity usually reaches a titer of at least 1:4. Following treatment of primary syphilis, the reactivity may be expected to continue to rise for a few weeks but should revert to non reactivity within 6-12 months following treatment. Ninety-seven percent of patients will be nonreactive within two years.

In SECONDARY or EARLY LATENT syphilis, the VDRL and/or RPRC Tests are invariably reactive, usually with a titer of 1:32 or higher. While the titer may continue to rise immediately after successful treatment, the reactivity should gradually revert to non reactivity within 18 months following the completion of successful treatment. After 2 years, over 75% will be nonreactive. Twenty-five percent will have positive titers that have stabilized at or below the four-fold decrease needed to document adequate treatment. The majority of these titers will have a low reactive titer 1:4 or less. If the patient with secondary syphilis develops a VERY strong reactivity, the VDRL or RPRC Test could be read spuriously as nonreactive, due to the prozone phenomenon. The laboratory should, therefore, be asked to dilute the "nonreactive" serum and continue the titration in all cases wherein suspicious lesions or clinical findings are present.

LATE syphilis (syphilis of more than one year's duration) may be symptomatic or asymptomatic. A patient may have late syphilis, either acquired or congenital, and have a nonreactive VDRL or RPRC Test. Further evaluation by means of a test such as the MHA-TP is necessary in such persons suspected of having late stage manifestations of syphilis. Cerebrospinal fluid (CSF) studies are recommended to rule out neurosyphilis in these cases. Late syphilis must be adequately treated.

Treatment of late syphilis may have no effect on the titer of reactive RPRC or VDRL Tests, but after successful treatment, the titer usually decreases "four-fold" (by two "dilutions"; e.g., from 1:32 to 1:8). A stable or rising titer during one year of observation after treatment suggests a treatment failure, reinfection, or a diagnostic error.

In any treatment situation, failure of the highest titer achieved to decrease four-fold within one year suggests a treatment failure and warrants reevaluation of the case, (the highest titer may be reached a week or two after treatment is instituted).

In cases wherein the epidemiological and clinical information fails to support serological findings, the diagnosis of syphilis should be questioned. A "biologic false positive" basis for seropositivity should be sought.

Biologic false positivity, meaning a nonsyphilitic basis for reactive RPRC or VDRL Tests, must be established by the use of treponemal antigen test such as the MHA-TP. The RPRC and the VDRL are two among many nontreponemal antigen tests. Among the nontreponemal tests, "false positivity" or "false reactivity" occurs in at least 1% of persons tested. Barring laboratory error, the trepenomal antigen tests are seldom falsely positive. They are more complicated and expensive to perform, and there are relatively more false negative test results. Treponemal antigen tests, such as the MHA-TP and FTA-ABS, are inappropriate for use as screening tests. They also are not quantitative tests and cannot be used for following titers in response to therapy or progression of disease.

While most persons infected with HIV will have typical antibody responses to syphilis infections, some may not. All persons suspected of having syphilis should be referred for HIV counseling and testing. If syphilis is suspected in a person known or suspected to have HIV, then direct examination of lesions exudate by darkfield microscopy or direct fluorescent antibody tests should be done.

Treponemal Antigen Tests

Treponemal tests detect an antibody that is directed toward pathogenic members of the genus *Treponema.*

The Bureau of Laboratories, Texas Department of Health, uses the MICRO-HEMAGGLUTINATION FOR TREPONEMA PALLIDUM (MHA-TP) Test for routine confirmatory testing for syphilis. The specificity of the MHA-TP Test is as good as or better than the FTA-ABS Test. Usually, a nonreactive result on an MHA-TP Test will establish the "biologic false positive" diagnosis of a "positive" nontreponemal antigen test (RPRC or VDRL). Transient (acute) false reactivity of the RPRC or VDRL Tests occurs in some patients due to intercurrent viral and bacterial infections, when the serum titer of heterophile antibodies is high. Serum controls used in the laboratory identify this heterophile activity when it occurs in the MHA-TP, permitting further evaluation with the FTA-ABS Test. Infectious mononucleosis (Epstein-Barr viral infections) and viral hepatitis, as well as herpes simplex infections, chancroid, and lymphogranuloma venereum, may be accompanied by biologic false positive serological tests for syphilis. Long term (chronic) biologic false positivity may be present in Hansen's disease and collagen diseases, such as systemic lupus erythematosus and rheumatoid arthritis, as well as in narcotics addiction and in some forms of neoplasms. A determination that a patient has a biologic false RPRC or VDRL mandates a search for the etiology of the positivity.

The Bureau of Laboratories will perform the FTA-ABS Test, but only under the following circumstances:

1. In suspected cases of primary syphilis in which two nontreponemal tests performed five days apart have shown a static reactive titer and in which the MHA-TP Test performed on the second specimen was nonreactive. This must be documented when the specimen is sent for FTA-ABS testing.
2. In diagnostic problems arising from conflicts between the overall clinical impression and results from both treponemal and nontreponemal tests. Such conflicts sometimes occur in cases of late syphilis. A brief written description of the diagnostic problem must accompany specimens sent for FTA-ABS testing.

The FTA-ABS Test is performed weekly on Fridays. The RPRC Test is performed daily. The MHA-TP Test is performed on Tuesdays and Thursdays, but only on specimens that are reactive by our RPRC Test, or in cases where the results of nontreponemal tests are equivocal and in which information describing this situation accompanies the specimen. Additionally, since it is recognized that the RPRC Test can be nonreactive in active tertiary syphilis, the MHA-TP will be performed if this justification is indicated.

Unlike the nontreponemal antigen tests, the MHA-TP and the FTA-ABS do not revert to nonreactivity after successful treatment of syphilis. Once reactive, they almost always stay reactive. Therefore, ordering repeated MHA-TP tests to check on a patient's progress is not warranted. In these situations, the desired information would be obtainable through repetition of the RPRC quantitative test.

Patients treated for OTHER sexually transmitted diseases should receive a serological test for syphilis because such persons are at relatively high risk for exposure to syphilis. Adequate penicillin or ceftriaxone treatment for gonorrhea is probably also effective against incubating syphilis. In such situations, further serological study is generally not needed unless another exposure occurs or new clinical findings warrant additional evaluation. Following recommended treatment of gonorrhea with any drug other than one of the penicillins or ceftriaxone, a follow-up RPRC quantitative test should be obtained at 2-4 weeks.

A reactive STS determined on serum raises the question as to whether testing of cerebrospinal fluid (CSF) is indicated. A lumbar puncture is generally not indicated during the primary and secondary stages of this infection, unless syphilitic meningitis is suspected or unexplained neurological findings occur. Following proper penicillin therapy for syphilis, a favorable blood serologic response (four-fold titer drop) generally indicates that NO testing of the CSF is required. Final judgment must be based on careful evaluation of the serologic response to treatment while following the serum titer for no less than 12 months, or reversion to nonreactivity. (See table)

Tests of the CSF (VDRL quantitative) SHOULD be performed in:

... all cases of congenital syphilis

... any syphilis patients with neurologic signs or symptoms,

... patients with primary, secondary or early latent syphilis treated with ANY DRUG OTHER THAN PENICILLIN should have a CSF test for syphilis one year after treatment. Patients with untreated syphilis of greater than one year's duration should have CSF tests if a non-penicillin regimen is planned to see if such therapy is appropriate, and

... patients with syphilis of greater than one year's duration (late syphilis) ideally should have a CSF test. If this is not possible, then priority should be given to persons with signs of active syphilis (aortitis, gumma, iritis) or who are HIV positive.

A reactive VDRL performed on a sample of spinal fluid should be considered neurosyphilis until proven otherwise. A diagnosis of central nervous system syphilis is supported by CSF findings of lymphocytic pleocytosis (often <100 wbc/mm3 and normal to elevated protein. In general, interpretation of CSF results may be difficult. Often, adults with early syphilis may show CSF abnormalities but not develop neurosyphilis. Consultation with experts is often necessary.

If an RPRC performed on the cord blood of a newborn is reactive, it may represent the passive transfer of maternal antibodies or congenital infection of the newborn. Such infants should be treated at birth if maternal treatment was inadequate; or unknown; or did not include penicillin; or if adequate follow-up of the infant cannot be ensured.

If an infected pregnant woman received adequate penicillin treatment at least 30 days prior to delivery, risk to the infant is small. However, in all cases the infant should be tested at birth, 1 month, and every 3 months for the first 15 months, until non-treponemal serologic tests are negative. If a serologic test is positive at 3 months, the infant should be treated for congenital syphilis. After treatment a non-treponemal test for syphilis should be performed every 3 months until adequate treatment can be documented. The non-treponemal test may become non-reactive or remain stable at a low titer.

Infants with congenital syphilis should have a CSF examination before treatment to provide a baseline for follow-up. Regardless of CSF results, infants should be treated with regimen effective for neurosyphilis.

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TABLE - Summary of Recommended of Follow-up

Condition	Treatment	Follow-up Posttreatment	
		Serology	Discharge
Primary, Secondary, and Early Latent	Yes	3rd and 6th	End of 6 months[1]
Contact of Primary or Secondary within 90 days of exposure. (with negative RPRC or VDRL)	Yes	None	Post treatment
Late Syphilis of more than 1 years duration, cardio-vascular syphilis, gummas (Late benign syphilis)	Yes	6 months and 12 months	End of 1 yr.
Syphilis in pregnancy[2]	Yes	Monthly until delivery	6 months to 1 year
Subsequent pregnancies. No change in titer[2]	No	Initial visit monthly and until delivery.	
Congenital[1] - Seropositive untreated infants (suspect maternal transfer.)	N/A Only if RPR is positive at 3 months	1st, 2nd, 3rd, 6th, 12th, 15th months;	15 months
Congenital - Treated infants[3]	Yes	3 and 6 months	End of 6 months[1]
Neurosyphilis[4]	Yes	3 and 6 months	6 months
Syphilis in the HIV infected	Yes	1,2,3,6,9,12	End of 2 years[1]

1. Discharge patients should have either negative serologic tests for syphilis or fixed low titers. If a positive titer or an increasing titer is found, the patient may warrant further follow-up.
2. Retreatment is indicated if there is any doubt concerning adequacy of previous treatment.
3. An appropriate medical specialist should be consulted regarding treatment of the complications of congenital syphilis.
4. Neurosyphilis patients should have a spinal fluid examination at the 6-month follow up visit. If a positive titer is found in the CSF, the patient may warrant further follow up.