JOURNAL OF CLINICAL MICROBIOLOGY, Oct. 1980, p. 629-630
0095-1137/80/10-0629/02$02.00/0

Vol. 12, No. 4

Reactivity of Microhemagglutination, Fluorescent Treponemal Antibody Absorption, and Venereal Disease Research Laboratory Tests in Primary Syphilis

JOHN D. DYCKMAN,† SALLY STORMS, AND THOMAS W. HUBER*

Houston Health Department Laboratory, Houston, Texas 77030

Seroreactivity in 130 cases of primary syphilis was 91.5% by fluorescent treponemal antibody absorption test, 82.3% by microhemagglutination (MHA-TP test), and 68.5% by the Venereal Disease Research Laboratory (VDRL) test. The MHA-TP test generally became reactive earlier than the VDRL test and confirmed all reactive and most weakly reactive VDRL results.

Tests to detect antibodies against antigens of *Treponema pallidum* have an important role in the diagnosis of syphilis. Treponemal tests are used as confirmatory tests for serological screening procedures which employ nontreponemal lipid antigens, such as the Venereal Disease Research Laboratory (VDRL) and rapid plasma reagin tests. Patients who exhibit a reactive nontreponemal test are diagnosed as having syphilis or a "biological false-positive" reaction, based upon the results of the treponemal antigen test. It is important, therefore, that treponemal tests demonstrate high sensitivity and specificity.

The most commonly used treponemal antigen test is the fluorescent treponemal antibody absorption (FTA-ABS) test. Recently, however, microhemagglutination (MHA) tests have become popular. The simplicity and time-saving features of MHA tests are influencing laboratories to employ this procedure instead of the FTA-ABS test. Although MHA tests are equal in sensitivity to the FTA-ABS test in later stages of syphilis, some studies have indicated that they are less sensitive in the primary stage of the disease (2-4, 6). Two reports suggest that MHA tests may become reactive later in the disease course than nontreponemal screening tests (2, 6). If MHA tests are undersensitive early in the disease, some patients with early primary syphilis and a reactive nontreponemal test would be incorrectly classified as biological false-positive reactors.

In our large reference laboratory, performing over 11,000 FTA-ABS tests each year, we were concerned about misdiagnosing primary syphilis when considering a change from the FTA-ABS test to the MHA test for *T. pallidium* antibodies (MHA-TP; Ames Co., Elkhart, Ind.) A study was designed to compare the reactivity of the VDRL, FTA-ABS, and MHA-TP tests in patients with primary syphilis. The VDRL and FTA-ABS reagents were produced by the Texas Health Department Laboratory, except for FTA antigen and conjugate, which were obtained from Beckman Instruments, Inc., Fullerton, Calif.

Patients with a primary chancre which yielded a positive dark-field examination for *T. pallidum* were selected for study. Serum from these patients was tested by all three serological procedures. The performance of VDRL and FTA-ABS test materials was assured by testing positive and negative control sera as described in the *Manual of Tests for Syphilis* (5). Known positive and negative control sera were tested each time the MHA-TP test was performed. Reactivity within one dilution of the expected titer of the positive control serum, along with lack of hemagglutination with unsensitized cells and with negative control sera, was required to report test results.

The results for the three tests are presented in Table 1. The FTA-ABS test was the most sensitive test, with 91.5% reactivity. Two patients had borderline FTA-ABS test results. The data from these patients are not included in the tabulation of results because the validity of VDRL reactivity could not be confirmed by the equivocal borderline FTA-ABS reaction. The MHA-TP test was reactive in 82.3% of the patients, and the VDRL was the least sensitive test, with only 68.5% of the patients yielding a reactive or weakly reactive serum. These results suggest that the MHA-TP test becomes reactive earlier than the VDRL during the course of syphilis.

To make a more accurate determination of the sequence of seroconversion, MHA-TP and FTA-ABS reactivity was correlated with the strength of VDRL reactivity (Fig. 1). Forty-one

† Present address: Fisher Scientific Co., Diagnostics Division, Orangeburg, NY 10962.

TABLE 1. *Reactivity of three serological tests in 130 patients with primary syphilis*

Test	No. (%) of patients that were	
	Reactive	Nonreactive
FTA-ABS	119 (91.5)	11 (8.5)
MHA-TP	107 (82.3)	23 (17.7)
VDRL	89 (68.5)[a]	41 (31.5)

[a] 14 (10.8%) were weakly reactive.

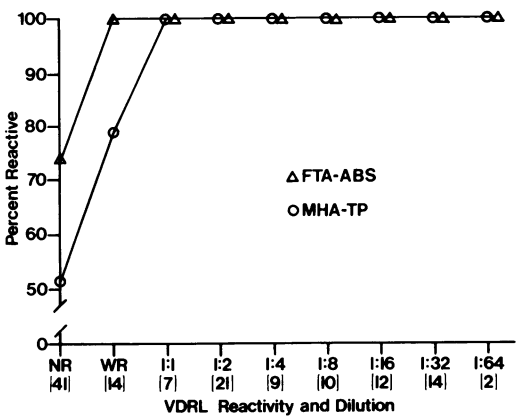

FIG. 1. *Correlation of VDRL titers with FTA-ABS and MHA-TP reactivity in primary syphilis. NR, Nonreactive; WR, weakly reactive. Numbers in parentheses indicate numbers of patients in each reaction category.*

patients were seen so early in the course of their infection that their VDRL reaction was negative. Seroreactivity in these 41 patients in the FTA-ABS and MHA-TP tests was 73% and 51%, respectively. By the time patients developed a weakly reactive VDRL test, their sera were uniformly reactive in the FTA-ABS test, and 78% were reactive in the MHA-TP test. Sera yielding a reactive VDRL result were uniformly reactive in both treponemal tests. No patient had a reactive MHA-TP or VDRL test with a nonreactive FTA-ABS test.

The results of this study indicate that the FTA-ABS is the first test to become reactive in the course of syphilis. The MHA-TP and VDRL tests become reactive somewhat later in the course of the disease. In the majority of cases in this study (21/24), the MHA-TP became reactive before the VDRL. However, a few patients (3/24) had a weakly reactive VDRL test with a nonreactive MHA-TP result. These findings are in excellent agreement with those of Alessi and Scioccati (1), who reported 62% reactivity for the VDRL and 83.3% reactivity for an MHA test in 90 patients with primary syphilis.

The MHA-TP test appears to be a reasonable alternative to the FTA-ABS test as a confirmatory test for syphilis as long as one keeps in mind that the MHA-TP is not as sensitive in the very early primary stage of the disease. However, in early primary syphilis, 78% of patients who have a weakly reactive and all who have a reactive VDRL will also have a reactive MHA-TP and will not be misclassified as having a false-positive nontreponemal antigen test. The failure of the MHA-TP to confirm VDRL reactivity occurred only in weakly reactive sera. In two of the three failures, +/− reactions occurred, suggesting that a low level of MHA-TP antibodies was present. A diagnosis could likely have been made in a subsequent serum sample collected a few days later. Clinical signs and symptoms should be evaluated along with laboratory data since the diagnosis of primary syphilis cannot be ruled out on the basis of reactive serological tests on a simple serum specimen.

LITERATURE CITED

1. **Alessi, E., and L. Sciocatti.** 1978. TPHA test: Experience at the Clinic of Dermatology, University of Milan, Br. J. Vener. Dis. **54**:151-154.
2. **Cox, P. M., L. C. Logan, and G. W. Stout.** 1971. Further studies of a quantitative automated microhemagglutination assay for antibodies to *Treponema pallidum*. Public Health Lab. **29**:43-50.
3. **Jaffe, H. W., S. A. Larsen, O. G. Jones, and P. E. Dans.** 1978. Hemagglutination tests for syphilis antibody. Am. J. Clin. Pathol. **70**:230-233.
4. **Logan, L. C., and P. M. Cox.** 1970. Evaluation of a quantitative automated microhemagglutination assay for antibodies to *Treponema pallidum*. Am. J. Clin. Pathol. **53**:163-166.
5. **Public Health Service.** 1969. Manual of tests for syphilis. Public Health Service publication no. 411. Superintendent of Documents, U. S. Government Printing Office, Washington, D.C.
6. **Rudolph, A. H.** 1976. The microhemagglutination assay for *Treponema pallidum* antibodies (MHA-TP), a new treponemal test for syphilis: where does it fit? J. Am. Vener. Dis. Assoc. **3**:3-8.