NHANES 2001-2002 Data Release
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Documentation for Laboratory Results

NHANES 2001-2002 Public Release Data File
Laboratory 36 - Syphilis-IgG, Syphilis Rapid Plasma Reagin (RPR), and *Treponema pallidum* Particle Agglutination (TP-PA)

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(2) Documentation File Name- Laboratory 36 - Syphilis-IgG, Rapid Plasma Reagin (RPR), and *Treponema pallidum* particle agglutination (TP-PA)

(3) Survey Years Included in this File Release-2001-2002

(4) Component Description

Although there has been a marked decrease in the number of primary and secondary syphilis cases in the United States, there has been very little decrease in the number of reported cases of late latent and tertiary syphilis over the past 20 years. This suggests that there may be a large pool of infected but asymptomatic persons. Although the primary and secondary stages of syphilis are infectious and associated with fetal wastage and the congenital syphilis syndrome, the tertiary stage is associated with a vasculitis that may cause neurologic and cardiovascular manifestations and other chronic problems. Similarly, primary and secondary syphilis increase the risk of HIV acquisition and transmission while latent disease may be associated with progression of HIV disease to AIDS and more prominent neurologic disease in HIV-infected persons. Despite the importance of syphilis as a risk factor for both chronic disease and the progression of HIV infection, there has not been a population-based measure of syphilis prevalence for the United States since 1980. Because these are often asymptomatic stages of infection and may lead to severe neurologic or cardiovascular complications, it is important to document a decrease in the late stages of syphilis that have resulted from our extraordinary efforts to reduce primary and secondary syphilis. NHANES offers a unique opportunity to estimate the prevalence of reactive serologic tests as an estimate of the prevalence of syphilis infections in the general population, to identify and confirm risk factors for syphilis, to confirm the risk for HIV infection and HIV-related neurologic disease among Americans with syphilis, and to monitor trends in prevalence as syphilis detection and treatment programs are established and expanded.

(5) Sample Description:

5.1 Eligible Sample

Participants aged 18 to 49 years were tested.

(6) Description of the Laboratory Methodology

6.1 Syphilis-G enzyme immunoassay (EIA)

The Captia Syphilis-G enzyme immunoassay (EIA) is an indirect method for the detection of IgG antibodies to Treponema pallidum[a,b,c,d,e]. Currently, CDC recommends that the test be used in the clinical laboratory as a confirmatory test for the diagnosis of syphilis. However, the test may be used as a screening test and is FDA approved for such use in clinical laboratories and blood banks. *T. pallidum* antigens are coated onto the wells of a 96-well microtiter plate. A dilution of the patient's serum is added to the well to allow any *T. pallidum* specific antibodies present to bind to the treponemal antigens. Biotinylated anti-human IgG labeled with strepavidin-peroxidase is used to detect the patient's antibody. After rinsing off the excess antibodies, an enzyme substrate is added for detection. If the patient has antibodies to *T. pallidum*, a color reaction takes place. The intensity of the color development is proportional to the amount of antibody present. This color change can then be read using a plate reader, which eliminates subjective interpretation of the results.

6.2 Rapid plasma reagin (RPR)

The rapid plasma reagin (RPR) 18-mm circle card test is a macroscopic, nontreponemal flocculation card test used to screen for syphilis[f, g, h, i]. The antigen is prepared from a modified Venereal Disease Research Laboratory (VDRL) antigen suspension containing choline chloride to eliminate the need to heat inactivate serum, ethylene-diamine-tetra- acetic acid (EDTA) to enhance the stability of the suspension, and finely divided charcoal particles as a visualizing agent. In the test, the RPR antigen is mixed with unheated or heated serum or with unheated plasma on a plastic-coated card. The RPR test measures IgM and IgG antibodies to lipoidal material released from damaged host cells as well as to lipoprotein-like material, and possibly cardiolipin released from the treponemes[j,k]. The anti lipoidal antibodies are antibodies that are produced not only as a consequence of syphilis and other treponemal diseases, but also in response to nontreponemal diseases of an acute and chronic nature in which tissue damage occurs[l] If antibodies are present, they combine with the lipid particles of the antigen, causing them to agglutinate. The charcoal particles coagglutinate with the antibodies and show up as black clumps against the white card. If antibodies are not present, the test mixture is uniformly gray. The test can be purchased in kit form or in component parts from many commercial sources. Without some other evidence for the diagnosis of syphilis, a reactive nontreponemal test does not confirm *T. pallidum* infection.

6.2 *Treponema pallidum* particle agglutination (TP-PA)

The Serodia *Treponema pallidum* particle agglutination (TP-PA) test is a treponemal test for the serologic detection of antibodies to the various species and subspecies of pathogenic Treponema, the causative agents of syphilis, yaws, pinta, bejel, and endemic syphilis. The test is a passive agglutination procedure based on the agglutination of gel particles sensitized with *T. pallidum* antigens by antibodies found in the patient's serum[m, n, o] The test is intended as a confirmatory test to replace the microhemagglutination assay for antibodies to *T. pallidum* (MHA-TP).

Serum containing antibodies to pathogenic treponemes react with gel particles sensitized with sonicated *T. pallidum*, Nichols strain (the antigen), to form a smooth mat of agglutinated gel particles in the microtiter tray well. If antibodies are not present, the particles settle to the bottom of the tray well, forming a characteristic compact button of unagglutinated particles. The unsensitized gel particle control well for each serum should also show this compact button, or the absence of agglutination.

The TP-PA test is used to confirm the reactive results[n,o] of a nontreponemal screening test for syphilis, such as the Venereal Disease Research Laboratory (VDRL) slide test, or as a diagnostic test in patients with a nonreactive nontreponemal test but with signs or symptoms suggestive of late syphilis.

(7) Testing algorithm for NHANES specimens

All serum specimens were tested for IgG antibody by the EIA assay. If results of this assay were positive or equivocal, the specimens were tested using the RPR test. If the RPR test was negative, the TP-PA test was performed on the sample.

(8) Suggested interpretation of laboratory results.

LBXSY1= Syphilis-G enzyme immunoassay (EIA)
LBDSY3= Rapid plasma reagin (RPR)
LBDSY4= *Treponema pallidum* particle agglutination (TP-PA)

Recent positive syphilis infection:
(LBXSY =1 or LBXSY1=3) and (LBDSY3 >=8)

Remote positive syphilis infection:
(LBXSY1 =1 or LBXSY1= 3) and (0<= LBDSY3 <8) and (LBDSY4=1)

No infection:
LBXSY1=2

OR

(LBXSY1=1 or LBXSY1=3) and (0<=LBDSY3<8) and (LBDSY4=2)

(9) Laboratory Quality Control and Monitoring

The NHANES quality control and quality assurance protocols (QA/QC) meet the 1988 Clinical Laboratory Improvement Act mandates. Detailed quality control and quality assurance instructions are discussed in the NHANES Laboratory/Medical Technologists Procedures Manual (LPM). Read the LABDOC file for detailed QA/QC protocols.

(10) Data Processing and Editing

Serum specimens were processed, stored and shipped to Division of AIDS, STD, and TB Laboratory Research, National Center for Infectious Diseases, National Centers for Disease Control and Prevention, Atlanta, Georgia. Detailed specimen collection and processing instructions are discussed in the NHANES Laboratory/Medical Technologists Procedures Manual (LPM). Read the LABDOC file for detailed data processing and editing protocols. The analytical methods are described in the Description of the Laboratory Methodology section.

(11) Data Access:

All data are publicly available.

(12) Analytic Notes for Data Users:

12.1 The analysis of NHANES 2001-2002 laboratory data must be conducted with the key survey design and basic demographic variables. The NHANES 2001-2002 Household Questionnaire Data Files contain demographic data, health indicators, and other related information collected during household interviews. They also contain all survey design variables and sample weights for these age groups. The phlebotomy file includes auxiliary information such as the conditions precluding venipuncture. The household questionnaire and phlebotomy files may be linked to the laboratory data file using the unique survey participant identifier SEQN.

(13) References

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