



REF

Catalog Number R0031C

IVD

In vitro Diagnostic

INTENDED USE

The *OnSite* Syphilis Ab Combo Rapid Test is a lateral flow chromatographic immunoassay for the qualitative detection of antibodies including IgG, IgM and IgA to *Treponema pallidum* (*Tp*) in human serum, plasma or whole blood. It is intended to be used as a screening test and as an aid in the diagnosis of infection with *Tp*. Any reactive specimen with the *OnSite* Syphilis Ab Combo Rapid Test must be confirmed with alternative testing method(s) and clinical findings.

SUMMARY AND EXPLANATION OF THE TEST

Tp, a spirochete bacterium, is the causative agent of the venereal disease syphilis. Although syphilis rates are declining in the United States after an epidemic outbreak between 1986 and 1990¹, the incidence of syphilis in Europe has increased since 1992, especially in the countries of the Russian Federation where peaks of 263 cases per 100,000 have been reported². In 1995, WHO (World Health Organization) reported 12 million new cases of syphilis³. To this day, the positive rate of syphilis serological tests among HIV-infected individuals continues to rise.

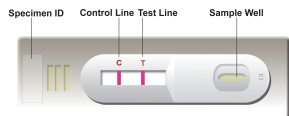
Serological detection of anti-*Tp* antibodies has long been recognized for the diagnosis of syphilis since the natural course of the infection is characterized by periods without clinical manifestations. Both IgM and IgG antibodies to *Treponema pallidum* were detected in sera from patients with primary and secondary syphilis. The IgM antibody may be detectable in the second week of infection, while IgG antibodies appear later at about 4 weeks⁴. Antibodies to *Treponema pallidum* can last for several years or even decades in the serum of a patient with untreated latent syphilis⁵.

Antigens such as Rapid Plasma Reagin (RPR) and *Tp* bacterial extracts have been used in syphilis serological tests for decades. However, RPR antigen is a nontreponemal antigen derived from bovine heart. Antibodies to RPR antigen do not develop until 1-4 weeks after the appearance of the chancre, thus this antigen lacks sensitivity to primary syphilis. The *Tp* extracts are prepared from inoculated rabbit testis and contain contaminated materials such as flagella which can lead to cross-reactions with borreliae and leptospirae in the serological test. In addition, the composition of extracts may vary from lot to lot. Recently, several highly immunogenic *Tp* specific antigens have been identified and used as an alternative to the traditional antigens with the advantages of high specificity and reproducibility⁶⁻⁹.

The *OnSite* Syphilis Ab Combo Rapid Test was developed to detect antibodies (IgM, IgG and IgA) to recombinant antigens of *Tp* in serum, plasma or whole blood. The test can be performed by minimally trained personnel and without cumbersome laboratory equipment.

TEST PRINCIPLE

The *OnSite* Syphilis Ab Combo Rapid Test is a lateral flow chromatographic immunoassay. The test cassette consists of: 1) a burgundy colored conjugate pad containing recombinant *Tp* antigens conjugated with colloidal gold (*Tp* conjugates) and a control antibody conjugated with colloidal gold, 2) a nitrocellulose membrane strip containing a test line (T line) and a control line (C line). The T line is pre-coated with non-conjugated recombinant *Tp* antigens, and the C line is pre-coated with a control line antibody.



When an adequate volume of test specimen is dispensed into the sample well of the test cassette, the specimen migrates by capillary action across the cassette. Anti-*Tp* antibody, if present in the specimen, will bind to the *Tp* conjugates. The immunocomplex is then captured on the membrane by the pre-coated *Tp* antigen forming a burgundy colored T line, indicating a *Tp* antibody positive test result. Absence of the T line suggests a negative result.

The test contains an internal control (C line) which should exhibit a burgundy colored line of the immunocomplex of the control antibodies regardless of color development on the T line. If the C line does not develop, the test result is invalid, and the specimen must be retested with another device.

REAGENTS AND MATERIALS PROVIDED

- Individually sealed foil pouches containing:
 - One cassette device
 - One desiccant
- Plastic droppers
- Sample diluent (REF SB-R0031, 5 mL/bottle)
- One package insert (instruction for use)

MATERIALS MAY BE REQUIRED AND NOT PROVIDED

- Positive Control
- Negative Control

MATERIALS REQUIRED BUT NOT PROVIDED

- Clock or Timer
- Lancing device for whole blood test

WARNINGS AND PRECAUTIONS

For in Vitro Diagnostic Use

- This package insert must be read completely before performing the test. Failure to follow the insert may lead to inaccurate test results.
- Do not open the sealed pouch unless ready to conduct the assay.
- Do not use expired devices.

- Bring all reagents to room temperature (15°C-30°C) before use.
- Do not use the components in any other type of test kit as a substitute for the components in this kit.
- Do not use hemolyzed blood specimens for testing.
- Wear protective clothing and disposable gloves while handling the kit reagents and clinical specimens. Wash hands thoroughly after performing the test.
- Users of this test should follow the US CDC Universal Precautions for prevention of transmission of HIV, HBV and other blood-borne pathogens.
- Do not smoke, drink or eat in areas where specimens or kit reagents are being handled.
- Dispose of all specimens and materials used to perform the test as biohazardous waste.
- Handle the negative and positive controls in the same manner as patient specimens.
- The test results should be read 15 minutes after a specimen is applied to the sample well or sample pad of the device. Reading the result after 20 minutes may give erroneous results.
- Do not perform the test in a room with strong air flow, i.e. an electric fan or strong air-conditioning.

REAGENT PREPARATION AND STORAGE INSTRUCTIONS

All reagents are ready to use as supplied. Store unused test device unopened at 2-30°C. If stored at 2-8°C, ensure that the test device is brought to room temperature before opening. The test device is stable through the expiration date printed on the sealed pouch. Do not freeze the kit or expose the kit to temperatures over 30°C.

SPECIMEN COLLECTION AND HANDLING

Consider any materials of human origin as infectious and handle them using standard biosafety procedures.

Plasma

- Collect blood specimen into a lavender, blue or green top collection tube (containing EDTA, citrate or heparin, respectively, in Vacutainer®) by venipuncture.
- Separate the plasma by centrifugation.
- Carefully withdraw the serum into a new pre-labeled tube.

Serum

- Collect blood specimen into a red top collection tube (containing no anticoagulants in Vacutainer®) by venipuncture.
- Allow the blood to clot.
- Separate the serum by centrifugation.
- Carefully withdraw the serum into a new pre-labeled tube.

Test specimens as soon as possible after collecting. Store specimens at 2-8°C if not tested immediately. Specimens can be stored at 2-8°C for up to 5 days. Specimens should be frozen at -20°C for longer storage.

Avoid multiple freeze-thaw cycles. Prior to testing, bring frozen specimens to room temperature slowly and mix gently. Specimens containing visible particulate matter should be clarified by centrifugation before testing. Do not use samples demonstrating gross lipemia, gross hemolysis or turbidity in order to avoid interference with result interpretation.

Blood

Drops of whole blood can be obtained by either finger tip puncture or venipuncture. Do not use any hemolyzed blood for testing.

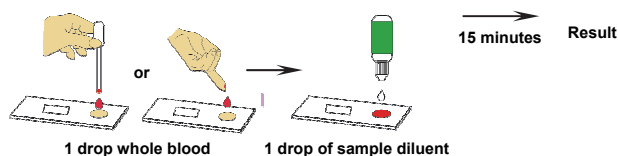
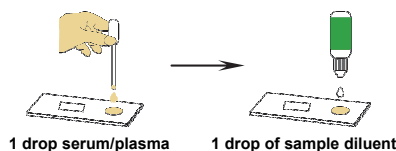
Whole blood specimens should be stored in refrigeration (2-8°C) if not tested immediately. The specimens must be tested within 24 hours of collection.

ASSAY PROCEDURE

- Bring the specimen and test components to room temperature if refrigerated or frozen. Mix the specimen well prior to assay once thawed.
- When ready to test, open the pouch at the notch and remove device. Place the test device on a clean, flat surface.
- Be sure to label the device with the specimen ID number.
- Fill the plastic dropper with the specimen.

Holding the dropper vertically, dispense 1 drop (about 30-45 µL) of serum/plasma or 1 drop of whole blood (about 40-50 µL) into the sample well making sure that there are no air bubbles.

Immediately add 1 drop (about 35-50 µL) of Sample Diluent to the sample well with bottle positioned vertically.



- Set up timer.
- Results can be read in 15 minutes. Positive results may be visible in as soon as 1 minute.

Do not read result after 20 minutes. To avoid confusion, discard the test device after interpreting the result.

QUALITY CONTROL

- Internal Control:** This test contains a built-in control feature, the C line. The C line develops after adding specimen and sample diluent. If the C line does not develop, review the entire procedure and repeat the test with a new device.
- External Control:** Good Laboratory Practice recommends using external controls, positive and negative, to ensure the proper performance of the assay, particularly under the following circumstances:
 - A new operator uses the kit prior to performing testing of specimens.
 - A new lot of test kit is used.
 - A new shipment of kits is used.
 - The temperature used during storage of the kit falls outside of 2-30°C.
 - The temperature of the test area falls outside of 15-30°C.
 - To verify a higher than expected frequency of positive or negative results.
 - To investigate the cause of repeated invalid results.

INTERPRETATION OF ASSAY RESULT

- NEGATIVE RESULT:** If only the C line is developed, the test indicates that no detectable anti-*Tp* antibody is present in the specimen. The result is negative or non-reactive.



- POSITIVE RESULT:** If both the C and T lines are developed, the test indicates the presence of anti-*Tp* antibodies in the specimen. The result is positive or reactive.



Samples with positive results should be confirmed with alternative testing method(s) and clinical findings before a positive determination is made.

- INVALID:** If no C line is developed, the assay is invalid regardless of color development on the T line as indicated below. Repeat the assay with a new device.



PERFORMANCE CHARACTERISTICS

1. Clinical Performance

A total of 1055 samples from susceptible subjects were tested with the *OnSite* Syphilis Ab Combo Rapid Test and with a TPPA (Treponema Pallidum Particle Agglutination) test. Comparison for all subjects is shown in the following table.

<i>OnSite</i> Syphilis Ab Combo Rapid Test			
TPHA	Positive	Negative	Total
Positive	318	0	318
Negative	2	735	737
Total	320	735	1055

Relative Sensitivity: 100%, Relative Specificity: 99.7%, Overall Agreement: 99.8%

2. Precision

Within run and between run precisions have been determined by testing 15 replicates with three of the samples: a negative, a weak positive and a strong positive sample. The negative, weak positive and strong positive samples were correctly identified in all of the tests performed in each run.

LIMITATIONS OF TEST

- The Assay Procedure and the Interpretation of Assay Result sections must be followed closely when testing for the presence of anti-*Tp* antibody in serum, plasma or whole blood from individual subjects. Failure to follow the procedure may lead to inaccurate results.
- The *OnSite* Syphilis Ab Combo Rapid Test is limited to the qualitative detection of anti-*Tp* antibody in human serum, plasma or whole blood. The intensity of the test line does not have a linear correlation with the antibody titer in the specimen.
- A negative result for an individual subject indicates the absence of detectable anti-*Tp* antibodies. However, a negative test result does not preclude the possibility of exposure to or infection with *Tp*.
- A negative result can occur if the quantity of the anti-*Tp* antibody present in the specimen is below the detection limits of the assay or the antibodies that are detected are not present during the stage of disease in which a sample is collected.
- Some specimens containing unusually high titers of heterophile antibodies or rheumatoid factor may affect expected results.
- The results obtained with this test should only be interpreted in conjunction with other diagnostic procedures and clinical findings.

REFERENCES

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Index of CE Symbols

Consult instructions for use	For <i>in vitro</i> diagnostic use only	Use by
Catalog #	Lot Number	Tests per kit
Store between 2-30°C	Authorized Representative	Do not reuse
Manufacturer	Date of manufacture	

CTK Biotech, Inc.
 10110 Mesa Rim Road
 San Diego, CA 92121, USA
 Tel: 858-457-8698
 Fax: 858-535-1739,
 E-mail: info@ctkbiotech.com

EC REP MDSS GmbH
 Schiffgraben 41, 30175 Hannover, Germany

PI-R0031C Rev. E
 Effective date: 2014-04-18
 English version

For Export Only. Not For Re-sale In the USA