

INTENDED USE

The Biogenix Syphilis Ab Rapid Test is a lateral flow chromatographic immunoassay for the qualitative detection of antibodies (IgG, IgM and IgA) to *Treponema pallidum* (Tp) in human serum or plasma. It is intended to be used by healthcare professionals as a screening test and as an aid in the diagnosis of infection with Tp. The test is not automated and does not require any additional instrument. Any reactive specimen with the Biogenix Syphilis Ab Rapid Test must be confirmed with alternative testing method(s) and clinical findings.

SUMMARY AND EXPLANATION OF THE TEST

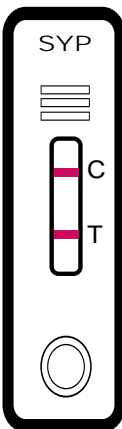
Tp, a spirochete bacterium, is the causative agent of the venereal disease syphilis. Although syphilis rates are declining in the United States after an epidemic outbreak between 1986 and 1990, the incidence of syphilis in Europe has increased since 1992, especially in the countries of the Russian Federation where peaks of 263 cases per 100,000 have been reported. In 1995, WHO (World Health Organization) reported 12 million new cases of syphilis. At present, the rate of positive syphilis serological tests among HIV-infected individuals continues to rise.

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

The Biogenix Syphilis Ab Rapid Test was developed to detect antibodies (IgG, IgM and IgA) to recombinant antigens of Tp in serum or plasma within 15 minutes. The test can be performed by minimally trained personnel and without cumbersome laboratory equipment.

TEST PRINCIPLE

The Biogenix Syphilis Ab 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, and 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
- Dropper
- One package insert (instruction for use)

MATERIALS REQUIRED BUT NOT PROVIDED

- Clock or Timer
- Disposable gloves

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 the test device if pouch is not intact.
- Do not use expired devices or components.
- Bring all reagents to room temperature (15-30°C) before use.
- Do not use the components of different lots and of any other type of test kit as a substitute for the components in this kit.
- Do not use hemolyzed blood 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 bio-hazardous waste.
- Handle the negative and positive controls in the same manner as the patient specimens.
- The test result should be read 15 minutes after a specimen is applied to the sample well of the device. Reading the test 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.
- Clean up spills thoroughly using appropriate disinfectant.

REAGENT PREPARATION AND STORAGE INSTRUCTIONS

All reagents are ready to use as supplied. Store test kit at 2-30°C. If stored at 2-8°C, ensure that all reagents are brought to room temperature before opening. The unopened test device is stable through the expiration date printed on the label, when stored at recommended temperature. Do not freeze the kit or expose the kit to temperatures above 30°C. The test device is sensitive to humidity and heat. Perform the test immediately after removing the test device from the foil pouch.

SPECIMEN COLLECTION AND HANDLING

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

Plasma

Step 1: Collect blood specimen into a lavender, blue or green top collection tube (containing EDTA, citrate or heparin, respectively in Vacutainer) by venipuncture.

Step 2: Separate the plasma by centrifugation.

Step 3: Carefully withdraw the plasma into a new pre-labeled tube.

Serum

Step 1: Collect blood specimen into a red top collection tube (containing no anticoagulants in Vacutainer®) by venipuncture.

Step 2: Allow the blood to clot.

Step 3: Separate the serum by centrifugation.

Step 4: 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. The 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.

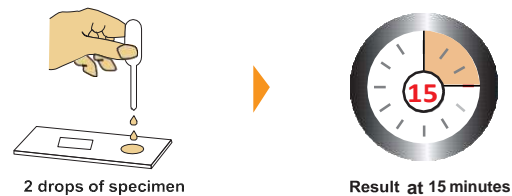
ASSAY PROCEDURE

Step 1: Bring the specimen and test components to room temperature if refrigerated or frozen. Mix the specimen well prior to assay once thawed.

Step 2: When ready to test, open the pouch at the notch and remove the device. Place the test device on a clean, flat surface.

Step 3: Be sure to label the device with the specimen ID number.

Step 4: Fill the specimen transfer device with the specimen. Holding the specimen transfer device vertically, dispense 2 drops (about 50-60µL) of specimen into the sample well, making sure there are no air bubbles.



Note: Add 1 drop of Saline or Phosphate-Saline buffer (common buffers used in clinics not provided in the kit) to the sample well if flow migration is not observed in the result window within 30 seconds, which could occur with highly viscous specimens.

Step 5: Set up timer.

Step 6: Results can be read at 15 minutes. Positive results may be visible in as soon as 1 minute.

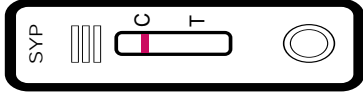
Do not read result after 20 minutes. To avoid confusion, discard the test device after interpreting the result.

QUALITY CONTROL

- Internal Control:** This test contains a built-in control feature, the C line. The C line develops after adding specimen. 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 kits is used.
 - A new shipment of test 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.

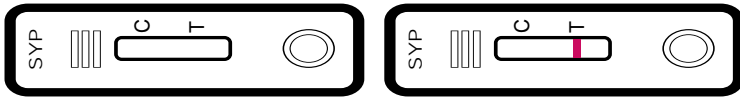


- POSITIVE RESULT:** If both the C and T lines are developed, the test indicates the presence of anti-*Tp* antibodies in the specimen. The result is positive or reactive.



Specimens with positive results should be confirmed with alternative testing method(s) and clinical findings before a diagnostic decision 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 883 samples from susceptible subjects were tested with the Biogenix Syphilis Ab Rapid Test in comparison with a commercial Syphilis Ab ELISA test. Comparison for all subjects is shown in the following table.

ELISA	Biogenix Syphilis Ab Rapid Test		Total
	Positive	Negative	
Positive	68	0	68
Negative	2	813	815
Total	70	813	883

Relative Sensitivity:100%, Relative Specificity:99.75%,Overall Agreement:99.77%

2. Cross-Reactivity

The negative specimen was spiked with serum specimens of infectious diseases and then tested according to the standard procedure. The results showed that the Biogenix syphilis Ab Rapid Test had no cross-reaction with the following tested serum specimens of infectious disease.

Specimen	Sample Size	Syphilis Ab Reactivity
Dengue Positive Serum	10	Negative
HAV Positive Serum	10	Negative
HCV Positive Serum	10	Negative
HIV Positive Serum	10	Negative
HBsAg Positive Serum	10	Negative
ANAPositive Serum	5	Negative
RF Positive Serum(≤2,500 IU/ml)	5	Negative

3. Interference

Common substances (such as pain and fever medication and blood components) may affect the performance of the Biogenix Syphilis Ab Rapid Test. This was studied by spiking these substances into four levels of Syphilis Ab standard controls.

The results are presented in the following table and demonstrate that at the concentrations tested, the substances studied do not affect the performance of the Syphilis Ab Rapid Test.

Note: - : Negative; + : Weak Positive; +++ : Strong Positive

Potential Interfering Substances Spiked	Syphilis Ab Reactivity		
	Negative	Weak Positive	Strong Positive
Control	-	+	+++
Bilirubin 15 mg/dL	-	+	+++
Creatinine 5 mg/dL	-	+	+++
Glucose 120 mg/dL	-	+	+++
Albumin 5g/L	-	+	+++
Salicylic Acid 4.34 mmol/L	-	+	+++
EDTA 3.4 µmol/L	-	+	+++
Urea 40 mg/dL	-	+	+++

LIMITATIONS OF TEST

- 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 if the antibodies that are detected are not present during the stage of disease in which a sample is collected.
- False negative results may arise because of hook effect due to very high titer of antibodies in sample. Repeat the test by using different dilution of same sample.
- Infection may progress rapidly. If the symptoms persist and the result from the Biogenix Syphilis Ab Rapid Test is non-reactive, it is recommended to test with an alternative method or to re-sample the patient a few weeks later.
- Some specimens containing unusually high titers of heterophile antibodies or rheumatoid factor may affect expected results.
- A positive result indicates a past or present infection. The results obtained with this test should only be interpreted in conjunction with other diagnostic procedures and clinical findings.

WASTE MANAGEMENT & DISPOSABLE

The contents of RDTs can be divided into :

Infectious waste:

- sharps (lancets, needles, scalpel blades)
- blood collection devices (tubes, straws, and loops); gloves; swabs; and cotton
- used cassettes. **Non-infectious waste (Recyclable):**
- packaging materials, desiccant, buffer, and unused or unusable RDTs.

****You must collect and dispose each type of waste in separate containers as per your waste management policies.**

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Mfg. By: BIOGENIX INC. PVT. LTD.

Factory: B - 19/A, S.I.L Ancillary Estate, Amausi Industrial Area, Nadarganj, Kanpur Road,Lucknow - 226008 (U.P.), India

Email: biogenix2007@yahoo.com, info@biogenixinc.com

Website: www.biogenixinc.com

Customer care no:+919140971443

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