Typhoid Ag Rapid Test Device
Package Insert

Cat: ITY-602 **Specimens:** Feces
Version: 02 **Effective Date:** 2018-10

For professional *in vitro* diagnostic use only.

INTENDED USE

The Typhoid Ag Rapid Test Device (Feces) is a lateral flow immunoassay for the simultaneous detection and differentiation of *Salmonella Typhoid* in feces.

SUMMARY AND EXPLANATION OF THE TEST

Typhoid fever is caused by S. typhi, a Gram-negative bacterium. World-wide an estimated 17 million cases and 600,000 associated deaths occur annually[1]. Patients who are infected with HIV are at significantly increased risk of clinical infection with Typhoid[2]. Evidence of H. pylori infection also presents an increase risk of acquiring typhoid fever. 1-5% of patients become chronic carrier harboring S. typhi or S. Para typhi in the gallbladder.

The clinical diagnosis of typhoid fever depends on the isolation of Typhoid from blood, bone marrow, feces or a specific anatomic lesion. In the facilities that can not afford to perform this complicated and time-consuming procedure, Filix-Widal test is used to facilitate the diagnosis. However, many limitations lead to difficulties in the interpretation of the Widal test[3,4].

In contrast, the Typhoid Ag Rapid Test Device (Feces) is a simple and rapid laboratory test. The test simultaneously detects S. typhoid antigen thus to aid in the determination to the S. typhiod.

TEST PRINCIPLE

The Typhoid Ag Rapid Test Device (Feces) is a lateral flow chromatographic immunoassay. The test cassette consists of: 1) a burgundy colored conjugate pad containing S. typhoid antibody conjugated with colloid gold, 2) a nitrocellulose membrane strip containing two test bands (S. typhoid bands) and a control band (C band). The S. typhoid band is pre-coated with monoclonal anti- S. typhoid for the detection of S. typhoid Ag, and the C band is pre-coated with goat anti mouse IgG.

When an adequate volume of test specimen is dispensed into the sample well of the cassette, the test specimen migrates by capillary action across the test cassette. S. typhoid Ag if present in the patient specimen will bind to the S. typhoid Ab conjugates. The immunocomplex is then captured on the membrane by the pre-coated S. typhoid antibody, forming a burgundy colored S. typhoid band, indicating a S. typhoid positive test result.

Absence of any test bands suggests a negative result. The test contains an internal control (C band) which should exhibit a burgundy colored band of the immunocomplex of goat anti Mouse IgG/Mouse IgG-gold conjugate regardless of the color development on any of the test bands. Otherwise, the test result is invalid and the specimen must be retested with another device.

MATERIALS

Materials Provided

- Test devices
- Droppers
- Single Buffer
- Package insert

Materials Required But Not Provided

- Specimen collection containers
- Feces paper
- Centrifuge
- Timer

WARNINGS AND PRECAUTUIONS

- For in-vitro diagnostic use only
- For professional use only
- Use the test device only once
- Do not eat, drink or smoke in the area where the specimens or kits are handled.
- Do not use test if pouch is damaged.
- Do not use test kit after expiration date
- Do not mix Sample Collection Tubes from different lots.
- Do not open the Test Cassette foil pouch until you are ready to perform the test.
- Do not spill solution into the reaction zone
- Do not touch the reaction zone of the device to avoid contamination
- Avoid cross-contamination of samples by using a new specimen collection container and specimen collection tube for each sample.
- All patient samples should be treated as if capable of transmitting disease. Observe established precautions against microbiological hazards throughout testing and follow standard procedures for proper disposal of specimens.
- Do not use more than the required amount of liquid.
- Bring all reagents to room temperature (15-30°C) before use.
- Wear protective clothing such as laboratory coats, disposable gloves and eye protection when specimens are being tested.
- Evaluate the test result after 10 minutes and not beyond 20 minutes.
- Store and transport the test device always at 2-30°C (36°-86°F)

REAGENT PREPARATION AND STORAGE INSTRUCTIONS

- The kit should be stored at 2-30°C until the expiry date printed on the sealed pouch.
- The test must remain in the sealed pouch until use.
- **Do not freeze.**
- Cares should be taken to protect components in this kit from contamination. Do not use if there is evidence of microbial contamination or precipitation. Biological contamination of dispensing equipment, containers or reagents can lead to false results.

SPECIMEN COLLECTION AND HANDLING

- Patients should not collect samples during their menstrual period, if they have bleeding hemorrhoids, blood in the urine, or if they have strained during bowel movement.
- Collect a random sample of feces in a clean dry container or receptacle.
- Unscrew and remove the collection tube applicator stick. Be careful not to spill or spatter solution from container.
- Collect random sample by using the applicator stick. Take sample from various surfaces of the feces specimen from 3 points.
- Re-insert the applicator stick into the tube and screw the cap tightly. Be careful not to break the tip of the Sample Collection Tube.
- The diluted sample must be tested in 30 minutes, otherwise the results would be not correct.

ASSAY PROCEDURE

Bring tests, specimens, buffer and/or controls to room temperature (15-30°C) before use.

1. Specimen collection and pre-treatment:
   1) Unscrew and remove the dilution tube applicator. Be careful not to spill or spatter solution from the tube. Collect specimens by inserting the applicator stick into at least 3 different sites of the feces.
   2) Place the applicator back into the tube and screw the cap tightly. Be careful not to break the tip of the dilution tube.
   3) Shake the specimen collection tube vigorously to mix the specimen and the extraction buffer. Specimens prepared in the specimen collection tube may be stored for 6 months at -20°C if not tested within 1 hour after preparation.
2. Testing
   1) Remove the test from its sealed pouch, and place it on a clean, level surface. Label the test with patient or control identification. To obtain a best result, the assay should be performed within one hour.
   2) Using a piece of tissue paper, remove the tip of the dilution tube. Hold the tube vertically and dispense **3 drops of solution** into the specimen well (S) of the test device.

      Avoid trapping air bubbles in the specimen well (S), and do not drop any solution in observation window.

      As the test begins to work, you will see color move across the membrane.
3. Wait for the colored band(s) to appear. The result should be **read at 10 minutes**. Do not interpret the result after 20 minutes.

INTERPRETATION OF ASSAY RESULT

POSITIVE RESULT:

The colored line in the control line region (C) appears and a colored line appears in test line region(T). The result is positive for S. typhoid Ag infection.

NEGATIVE RESULT:

One colored band appears in the control band region (C). No band appears in the test band region (T)

INVALID RESULT:

Control line (C) falls to appear. Insufficient buffer volume or incorrect procedural techniques are the most likely reasons for control line failure. Review the procedure and repeat the procedure with a new test device. If the problem persists, discontinue using the test kit immediately and contact your local distributor.

NOTE:

1. The intensity of the color in test region (T) may vary depending on the concentration of aimed substances present in the specimen. Therefore, any shade of color in the test region should be considered positive. Besides, the substances level cannot be determined by this qualitative test.
2. Insufficient specimen volume, incorrect operation procedure, or performing expired tests are the most likely reasons for control band failure.

QUALITY CONTROL

1. **Internal Control:** This test contains a built-in control feature, the C band. The C line develops after adding specimen and sample diluent. Otherwise, review the whole procedure and repeat test with a new device.
2. **External Control:** Good Laboratory Practice recommends using the external controls, positive and negative (provided upon request), to assure the proper performing of the assay, in particularly, under the following circumstances:
   a. New operator uses the kit, prior to performing testing of specimens.
   b. A new lot of test kit is used.
   c. A new shipment of kits is used.
   d. The temperature used during storage of the kit fall outside of 2°C -30°C.
   e. The temperature of the test area falls outside of 15°C -30°C.

PERFORMANCE CHARACTERISTICS

Clinical Performance For S. typhoid Test

A total of 115 samples from susceptible subjects were tested by the Typhoid Ag Rapid Test Rapid Test and by another commercial S. typhi test. Comparison for all subjects is showed in the following table.

	Typhoid Ag Rapid Test		
S. typhoid Commercial test	**Positive**	**Negative**	**Total**
Positive	21	1	22
Negative	0	93	93
Total	21	94	115

Relative Sensitivity: 100%, Relative Specificity: 98.9%, Overall Agreement: 99.1%

LIMITATIONS OF TEST

1. The Assay Procedure and the Test Result Interpretation must be followed closely when testing the presence of antigens to S. typhoid in feces from individual subjects. Failure to follow the procedure may give inaccurate results.
2. The S. typhoid Ag Rapid Test device (Feces) is limited to the qualitative detection of antigens to *S. typhoid* in human feces. The intensity of the test band does not have linear correlation with the antigen titer in the specimen.
3. A negative result for an individual subject indicates absence of detectable *S. typhoid* Ag. However, a negative test result does not preclude the possibility of exposure to *S. typhiod*.
4. A negative result can occur if the quantity of *S. typhoid* Ag present in the specimen is below the detection limit of the assay, or the antigens that are detected are not present during the stage of disease in which a sample is collected.
5. If the symptom persists, while the result from S. typhoid Ag Rapid Test device (Feces) is negative or non-reactive result, it is recommended to re-sample the patient few days late or test with an alternative test method, such as bacterial culture method.
6. Some specimens containing unusually high titer of heterophile antigens or rheumatoid factor may affect expected results.

REFERENCES

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