

For rapid qualitative detection of both *Salmonella typhi* antibodies, IgG and IgM in human plasma and serum.  
Only for *In Vitro* diagnostic use.

#### CLINICAL SIGNIFICANCE

Typhoid fever and paratyphoid fever are bacterial infections caused by *Salmonella Typhi* and paratyphoid A, B, C respectively, which is transmitted through the ingestion of tainted food and water. World-wide an estimated 17 million cases and 600,000 associated deaths occur annually. Patients who are infected with HIV are at significantly increased risk of clinical infection. 1- 5% of patients become chronic carriers harboring *S. typhi* in the gallbladder. The clinical diagnosis of infections depends on isolation of *S. typhi* and paratyphi from blood, bone marrow or a specific anatomic lesion. In facilities that can not afford to perform this complicated and time-consuming procedure, Widal test is used to facilitate diagnosis. However, many limitations lead to difficulties in the interpretation of the Widal test. In contrast, the Typhoid IgG/IgM2.0 Rapid Test is a simple, fast laboratory test that simultaneously detects and differentiates IgG and IgM antibodies to *S. typhi* and paratyphi antigen5 thus aiding in the determination of current or previous exposure to *S. typhi* and paratyphi. IgM positive or IgM /IgG both positive suggest current infection, while IgG positive suggests late stage of infection, or previous infection, or latent infection.

#### PRINCIPLE

The Typhoid IgG/IgM Rapid Test is a lateral flow chromatographic immunoassay. The test cassette consists of: 1) a burgundy colored conjugate pad containing recombinant H antigen and O antigen conjugated with colloidal gold (HO conjugates) and rabbit IgG-gold conjugates, 2) a nitrocellulose membrane strip containing two test bands (G and M bands) and a control band (C band). The M band is pre-coated with monoclonal anti-human

P IgM for the detection of IgM anti-*S. typhi* and paratyphi. G band is pre-coated with reagents for the detection of IgG anti-*S. typhi* and paratyphi, and the C band is pre-coated with goat anti rabbit 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. IgM antibodies if present in the patient specimen will bind to the HO conjugate.

The immunocomplex is then captured on the membrane by the pre-coated anti-human IgM antibody, forming a burgundy colored M band, indicating a *S. typhi* or paratyphi IgM positive test result. IgG antibodies if present in the patient specimen will bind to the HO conjugates. The immunocomplex is then captured by the pre-coated reagents on the membrane, forming a burgundy colored G band, indicating a *S. typhi* or paratyphi IgG positive test result. Absence of any test bands (M and G) suggests a negative result. The test contains an internal control (C band) which should exhibit a burgundy colored band of the immunocomplex goat anti rabbit IgG/rabbit 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.

#### KIT COMPONENTS

Test Device, Assay Buffer, Sample Dropper, one desiccant and product insert.

#### PRECAUTIONS

For *In Vitro* Diagnostic Use

1. This package insert must be read completely before performing the test. Failure to follow the insert gives inaccurate test results.
2. Do not open the sealed pouch, unless ready to conduct the assay.
3. Do not use expired devices.
4. Bring all reagents to room temperature (15°C-30°C) before use.
5. Do not use the components in any other type of test kit as a substitute for the components in this kit.
6. Wear protective clothing and disposable gloves while handling the kit reagents and clinical specimens. Wash hands thoroughly after performing the test.
7. Users of this test should follow the US CDC Universal Precautions for prevention of transmission of HIV, HBV and other blood-borne pathogens.
8. Do not smoke, drink, or eat in areas where specimens or kit reagents are being handled.
9. Dispose of all specimens and materials used to perform the test as biohazardous waste.
10. Handle the Negative and Positive Control in the same manner as patient specimens.
11. The testing results should be read within 15 minutes after a specimen is applied to the sample well or sample pad of the device. Reading the test after 15 minutes may give erroneous results.

#### STORAGE & STABILITY

Store as packaged in the sealed pouch at 2-40 °C. The test device is stable through the expiration date printed on the sealed pouch. The test device must remain in the sealed pouch until use. Do not freeze.

#### SPECIMEN COLLECTION & PRESERVATION

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

##### Plasma

1. Collect blood specimen into a lavender, blue or green top collection tube (containing EDTA, citrate or heparin, respectively in Vacutainer®) by vein puncture.
2. Separate the plasma by centrifugation.
3. Carefully withdraw the plasma into a new pre-labeled tube.
4. Use fresh whole blood only.

##### Serum

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

Test specimens as soon as possible after collecting. Store specimens at 2°C- 8°C if not tested immediately. Store specimens at 2°C-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 on result interpretation.

## CLEAR TYPHI (TYPHOID IgG/IgM RAPID TEST CARD) PLASMA/SERUM

#### ASSAY PROCEDURE

1. Bring the specimen and test components to room temperature if refrigerated or frozen. Mix the specimen well prior to assay once thawed.
2. When ready to test, open the pouch at the notch and remove device. Place the test device on a clean, flat surface.
3. Be sure to label the device with specimen's ID number.
4. Dispense 2 drop (about 35-40µl) of specimen into the sample well making sure that there are no air bubbles. Then add 1 drop (about 30-35 µl) of Assay Buffer immediately.
5. Results can be read in 15 minutes.

Don't read result after 15 minutes. To avoid confusion, discard the test device after interpreting the result.

#### INTERPRETATION OF RESULTS

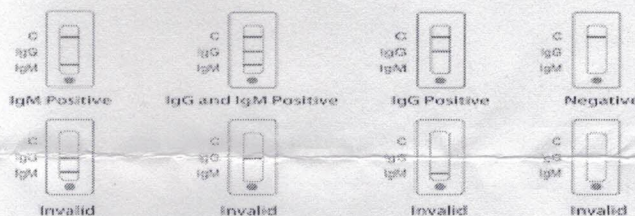
**NEGATIVE OR NON-REACTIVE RESULT:** If only the C band is present, the absence of any burgundy color in the both test bands (M and G) indicates that no anti-*S. typhi* or paratyphi antibody is detected in the specimen. The result is negative or non-reactive.

#### POSITIVE OR REACTIVE RESULT:

1. **Typhoid IgM Positive Reaction :** In addition to the presence of C band, if only M band is developed, the test indicates for the presence of anti- *S. typhi* or paratyphi IgM in the specimen. The result is IgM positive or reactive.
2. **Typhoid IgG Positive Reaction :** In addition to the presence of C band, if only G band is developed, the test indicates for the presence of anti- *S. typhi* or paratyphi IgG in the specimen. The result is IgG positive or reactive.
3. **Typhoid IgG & IgM Positive Reaction :** In addition to the presence of C band, both M and G band are developed, the test indicates for the presence of anti-*S. typhi* or paratyphi IgG and IgM in the specimen. The result is both IgG and IgM positive or reactive.

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

4. **INVALID:** If no C band is developed, the assay is invalid regardless of any burgundy color in the test bands as indicated below. Repeat the assay with a new device.



#### 3. Performance Characteristics

The kit has been evaluated with the known panel of fresh *S. typhi* IgM & IgG antibody positive and negative samples and results are compared with licensed commercially available test. Following is the in-house evaluation:

Sample type	No. of samples tested	Result on ELISA kit	CLEAR TYPHI CARD
Negative for <i>S. typhi</i>	200	200	199
<i>S. typhi</i> IgM Positive	50	50	48
<i>S. typhi</i> IgG Positive	50	50	49

Typhi IgM antibodies : Sensitivity : 96.0% Specificity : 99.5%  
Typhi IgG antibodies : Sensitivity : 98.0% Specificity : 99.5%

#### LIMITATIONS

1. The Assay Procedure and the Test Result Interpretation must be followed closely when testing the presence of antibodies to *S. typhi* or paratyphi in serum and plasma from individual subjects. Failure to follow the procedure may give inaccurate results.
2. The Typhoid IgG/IgM Rapid Test is limited to the qualitative detection of antibodies to *S. typhi* or paratyphi in human serum and plasma. The intensity of the test band does not have linear correlation with the antibody titer in the specimen.
3. A negative result for an individual subject indicates absence of detectable anti-*S. typhi* or paratyphi antibodies. However, a negative test result does not preclude the possibility of exposure to *S. typhi* or paratyphi.
4. A negative result can occur if the quantity of anti-*S. typhi* or paratyphi antibodies present in the specimen is below the detection limit of the assay, or the antibodies 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 Typhoid IgG/IgM Rapid Test is negative or non-reactive result, it is recommended to re-sample the patient few days later or test with an alternative test method, such as bacterial culture method.
6. Some specimens containing unusually high titer of heterophile antibodies or rheumatoid factor may affect expected results.
7. The results obtained with this test should only be interpreted in conjunction with other diagnostic procedures and clinical findings.

#### BIBLIOGRAPHY

1. Ivanoff BN, Levine MM, Lambert PH. Vaccination against typhoid fever: present status. Bulletin of the World Health Organization 1994; 72: 957- 71.
2. Gotuzzo E, Frisancho O, Sanchez J, Liendo G, Carillo C, Black RE, Morris JG. Association between the acquired immunodeficiency syndrome and infection with *Salmonella typhi* or *Salmonella paratyphi* in an endemic typhoid area. Archives of Internal Medicine 1991; 151: 381-2.