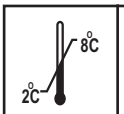
BS ISO-15223-1:2012(E) MEDICAL DEVICES SYMBOL

	Temperature Limitation	 2001-06	Date of Manufacture
IVD	In vitro Diagnostic Device	LOT	Batch Code
	Company name & address		Refer Operating Instructions
	Use by	 Company	Company name
EC REP	Authorised Representative in European Community		Do Not Reuse
	Sufficient for		KEEP AWAY FROM SUNLIGHT
	KEEP DRY	 NON STERILE	NON-STERILE
CONTROL -	NEGATIVE CONTROL	CONTROL +	POSITIVE CONTROL

Manufactured in India by :

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R- 2, 05/2013

TYPHO (WIDAL) Slide Test Kit

BHAT Bio Scan®

For Professional Use

Rapid slide test for qualitative and semiquantitative, in vitro determination of specific antibodies present in serum against Salmonella typhi 'O','H' Salmonella paratyphiA(H) and B(H) antigens.

IVD

Read Pack Insert before use provided along with the kit

REF **TYP**

INTENDED USE :

Typho (Widal) test is a Rapid slide test for qualitative and semiquantitative, in vitro determination of specific antibodies present in serum against Salmonella typhi 'O' & 'H' and Salmonella paratyphi A(H) & B(H) antigens.

INTRODUCTION :

Salmonella is a member of the gram negative bacilli Enterobacteriaceae. S.typhi is more likely than other species to systematically invade the body, enter the blood circulations, and cause the serious febrile disease,typhoid fever. Typhoid or enteric fever is a clinical syndrome characterized by fever, headache, prostration, cough, splenomegaly and leucopenia. With out antimicrobial theraphy, the illness is prolonged and associated with serious complications. The incubation period from the time of exposure until the time of development of signs and symptoms is 7 to 14 days(the range is from 3 to 60 days). Relapse occurs in 5% to 10% of untreated patients. Symptoms in relapse are usually milder than those of the initial illness and begin about 2 weeks after discontinuation of antimicrobial theraphy.

The carrier state is asymptomatic. The chronic intestinal carrier state, defined as documented fetal excretion of S.typhi for a minimum of one year, is observed in 1% to 3% of typhoid fever patients. The gall bladder is the site of persistent intestinal infection.

PRINCIPLE :

Killed Salmonella suspensions displaying the somatic 'O' and flagellar 'H' antigens agglutinate with the antibodies present in the serum of patients exposed to these organisms.

STORAGE AND STABILITY :

Storage :

Store the reagents at 2 to 8°C. Do not use the reagent beyond the expiry date mentioned on it. Before performing the test bring all the reagents to Room temperature. Replace the reagents to 2 to8°C soon after performing the test. DO NOT FREEZE THE REAGENTS.

Stability :

1. The unopened kit is stable for 24 months from the date of manufacturing as indicated on the package.
2. The opened kit is stable for 3 months from the date of opening.
3. Repeated freeze thaw of reagents from 2-8°C to Room temperature several times will reduce the stability of the kit.

PACK SIZE : 4x5ml / 4x10ml

CONTENTS OF THE KIT :

Sl.No.	Contents	Pack size	
		4x5ml	4x10ml
1.	Salmonella typhi 'O' Antigen	5ml	10ml
2.	Salmonella typhi 'H' Antigen	5ml	10ml
3.	Salmonella typhi A (H) Antigen	5ml	10ml
4.	Salmonella typhi B (H)Antigen	5ml	10ml
5.	Widal Positive Control	0.5ml	0.5ml
6.	Glass slide	1No.	1No.
7.	Product pack insert	1	1

SPECIMEN :

Use fresh serum for testing. The specimen should be Non-hemolysed and free from contamination. The specimen may be stored at 2 to8°C for upto 8 days and at –20°C for upto 4 weeks.(Use EDTA,Heparin or Oxalate as anticoagulant).

SPECIMEN COLLECTION AND HANDLING :

Collect blood in a clean sterilized vial and allow it to clot. Separate the serum by centrifugation at 10000 rpm for 10 minutes at room temperature. It is recommended that fresh samples should be used. If serum is not used for testing immediately it should be stored at 2-8°C or Frozen at –20°C. Bring specimen to room temperature,and specimen should be mixed properly and centrifuged before use. Do not heat or repeated freeze thaw the specimens.

PRECAUTIONS :

1. For invitro diagnostic use only.
2. Bring all reagents and specimens to Room temperature, prior to testing.
3. Avoid using lipemic, haemolysed or contaminated specimen.
4. All human serum and plasma samples should be considered potentially infectious. It is recommended that all specimens of human origin should be handled as recommended for any potentially infectious human serum or blood specimen in the center for disease control/National Institute of Health Manual"Biosafety in Microbiological and Biomedical Laboratories",1984.
5. Never pipette by mouth.
6. Do not smoke, eat or drink in areas in which specimens or kit reagents are handled.
7. Wear disposable gloves while handling specimens and kit reagents. Afterwards wash hands carefully.
8. Avoid splashing or forming aerosols.
9. Discard all materials and specimens as if capable of transmitting infection. The preferred method of disposal is autoclaving for a minimum of one hour at 121°C. Liquid waste containing acid may be mixed with sodium hypochlorite in volumes such that the final mixture contains 50-500mg/l available chlorine. Allow 30 minutes for decontamination to be completed.

NOTE :

1. Liquid waste containing acid must be neutralized with a proportional amount of base prior to the addition of sodium hypochlorite.
2. Spills should be wiped thoroughly using either an iodophor disinfectant or sodium hypochlorite solution. Materials used to wipe up spills should be added to biohazardous waste matter for proper disposal.
3. Deterioration is indicated by a significant decrease by weak agglutination.
4. Do not use reagents after the expiration date printed on the label.
5. When removing reagents from the bottles, use aseptic technique to avoid contamination.
6. Mix the reagent bottle gently before use.
7. Do not use contaminated serum sample for testing.
8. Ensure reused glass slide is disinfected, washed thoroughly and rinsed free of detergents.

TEST PROCEDURE :

A. Qualitative Analysis

1. Bring the reagents and specimens to room temperature.
2. Mix the antigen suspension thoroughly prior to use.
3. Drop/pipette onto separate cells of the slide.
4. Serum sample - 1 drop

Positive control serum - 1 drop

Antigen suspension O, H, A (H), B (H) - 1 drop each on respective cells

Antigen suspension (O) on control cell'O' - 1 drop

Antigen suspension (H) on control cell'H' - 1 drop

Mix the contents of each cell, spreading the reagent mixture over the entire area of the cell. Tilt the slide back and forth to ensure thorough mixing. Observe results at the end of 1 minute under high intensity of light.

B. Semi quantitative Analysis :

To a clear glass slide add as follows.

Cells	Serum Volume	Antigen Drop	Corresponding titre
1	0.08 ml	1 Drop	1 : 20
2	0.04 ml	1 Drop	1 : 40
3	0.02 ml	1 Drop	1 : 80
4	0.01 ml	1 Drop	1 : 160
5	0.005 ml	1 Drop	1 : 320

INTERPRETATION OF RESULTS :

A. Qualitative Analysis :

Read the results under high intensity of light. Regardless of the degree of reactivity and test results showing slight but definite agglutination is reported as reactive or positive. Complete absence of agglutination and a clear suspension indicates negative result.

B. Semi quantitative Analysis :

Agglutination may be observed in normal serum upto a titre of 1:60. A titre of 1:80 (Slide) or more is considered significant and a rise in titre after a few days will confirm the diagnosis. Individuals who have been previously immunized or inoculated with TAB vaccine or have a history of enteric illness. To confirm the infection a rise in titre after a few days should be checked. A moderate rise in titre of all three (H) agglutinations simultaneously against all 'H' suggestive of TAB vaccination.

TROUBLE SHOOTING :

FALSE POSITIVE	
Cause	**Remedy**
1. Contaminated serum or reagents	Make sure that there is no contamination of serum or reagent. Use clean slide for testing. Do not read the result after 4 minutes.
2. Drying in slide test	Do not read the result after 4 minutes
3. Improper mixing of Antigen suspensions	Mix the Antigen suspencious thoroughly before use.

FALSE NEGATIVE	
Cause	**Remedy**
1. Contamination of serum or reagents	Make sure that there is no contamination of Serum or reagent Use clean slide for testing.
2. Drying in slide test	Do not read the result after 4 minutes.
3. Improper Mixing of Antigen suspensions	Mix the Antigen suspensions thoroughly before use.

WEAKLY / DELAYED REACTION	
Cause	**Remedy**
1. Prolonged storage of serum specimen	Store the serum sample at 2-8°C for upto 8 days and at 20°C for upto 4 weeks.
2. Expired reagent	Check the expiry date on the reagent
3. Improper storage conditions of the kit	The kit has to be stored at 2-8°C.
4. Improper Mixing of Antigen suspensions	Mix the Antigen suspensions thoroughly before use.

PERFORMANCE CHARACTERISTICS :

Accuracy : Bhat Bio-Scan® Typho (Widal) Test meets the requirements when tested against DCI approved kits.

SPECIFICITY :

No. of Negative samples tested	No. of Negative by Bhat Bio-Scan® Typho (Widal) Test	Specificity (%)
40	40	100

SENSITIVITY :

No. of Positive samples tested	No. of Positive by Bhat Bio-Scan®Typho (Widal) Test	Sensitivity (%)
35	35	100

DIAGNOSTIC VALUE :

Salmonella are motile organisms with 'O' (Somatic) & 'H' (Flagellar) antigens. 'O' antigens of many species have many antigenic components in common, S. paratyphi A&B are commonly encountered in our country, and hence are employed in the tests. Agglutination appears at the beginning of the 2nd week, reach a maximum during the 3rd week and may persists for weeks or months after convalesence. Cross agglutinations with the 'O' suspensions of S.typhi and S.paratyphi'A' & S.Paratyphi 'B' often take place due to possession of some common epitopes of the somatic antigens. The titre over 1:120 with either 'O' or 'H' suspension is practically considered significant. In the case of S.paratyphi 'A' & S.paratyphi 'B' infections, the 'H' agglutinations are more marked and high titres are usually recorded.

LIMITATIONS OF THE TEST :

1. It is recommended that with every set of tests, positive control should be included. The result of positive control should be read before reading the test result. The satisfactory result of positive control indicates that the reagents are working well.
2. Factors other than reagents, which might affect the performance of the test, include cleanliness of glassware, meticulous follow up of the procedure.
3. Positive result only indicates salmonella infection. A rise in titer after a week should be checked to confirm the infection.
4. Agglutination with O Antigen with titer less than 1:80 is not significant.

REFERENCES :

1. Cruickshank. R. (1965), Medical Microbiology, 11th Edit, P 907.
2. Felix, A. (1942), Brit.Med.J., 11.,597.
3. Protell, RL.,etal., 1971, Lancet, 11,330.