



PRINCIPLE

The Immunospecific Antibodies present in serum of infected host reacts with O⁺ & H⁺ antigen present in the killed bacterial suspension of Salmonella & results in agglutination or clumps on the slide

CLINICAL SIGNIFICANCE

The disease like Enteric Fever or Typhoid fever shows symptoms generally by very high consistent fever, loss of appetite, transitory Bacteremia, round or oval shaped ulcers with smooth peritoneal surface of Peyer's patches and solitary lymphoid follicles of ileum etc. The disease is caused by organism Salmonella typhosa. This micro-organism produces 2 kinds of antigen "O" antigens on the cell wall & "H" antigen on its flagella. In response to stimulation, host immune system produces antibodies to counteract the effects of corresponding antigen. One more species of salmonella S. Paratyphoid A or paratyphoid B cause paratyphoid fever that is characterized by milder cause of disease. Also this organism produces somatic O⁺ & flagellar antigen which is termed as A(H) & B(H) respectively. Similar antigenic properties also observed in Salmonella spp that causes food poisonings & fetal infection.

SAMPLE COLLECTION & STORAGE

- Fresh clear, serum sample preferred.
- Sample should be stored at 2-8°C away from direct light.

PRECAUTIONS

- * Widal reagent kit is for In vitro diagnostic use only.
- * All the reagents should be brought to room temperature before use.

- * Shake Antigen vial well before use & also include Positive & Negative control sera for greater proficiency in interpretation of result.
- * Patient history should be taken into account before giving the final result.
- * Empty the dropper after use in order to avoid the possibilities of false positive results.
- * In a non inoculated person, the titer as high as 1: 80 between 7th or 10th day of fever is of diagnostic value & the same titre increases gradually during subsequent period.
- * In an inoculated person the question of anamnestic response should always be born in mind and 'H' in subsequent period.

KIT CONTENTS & STORAGE	4x5ML	2+2X5ML
S.typhi "O" Antigen	1 Bottle	2 Bottles
S.typhi H" Antigen	1 Bottle	2 Bottles
S.Paratyphi A(H)' Antigen	1 Bottle
S.Paratyphi B(H)' Antigen	1 Bottle
Widal Positive Control	1 Vial	1 Vial
Accessories	Glass Slide	Glass Slide

All reagents are to be stored at 2.8 °C and stable till expiry date mentioned.

PROCEDURE

A) Rapid Slide Test (Screening Test)

- Clean the glass slide provided in the kit
- Put one drop of Positive control in the first circle
- Place one drop of patient's serum in each of the remaining circles
- Then add one drop of O, H, A(H), B(H) antigen to each patient's sample circle and any
- Antigen to the positive control circle
- Spread the contents to till the whole circle area.

RESULTS

If agglutination occurs in Case of H or AH or BH and granular agglutination in case of O indicates POSITIVE reaction and proceed for quantitative testing. Rock the slide for 1 minute & observe for agglutination.

B) Quantitative Slide Test

Clean the glass slide provided in the kit and proceed as follows:

Circle	Serum vol	Appropriate Antigen drop		Corresponding Titre
1	0.005ml	1drop	Mix well & rotate for 1 minute & observe for agglutination	1:320
2	0.01ml	1drop		1:160
3	0.02ml	1drop		1:80
4	0.04ml	1drop		1:40
5	0.08ml	1drop		1:20

RESULTS

A positive reaction indicated by diagnostic titre of 1 :80

The titre of the total antibody is the minimum amount of serum which shows clear agglutination.

Repeat the above procedure with all the other antigens which showed agglutination in Rapid Screening test

LIMITATIONS

As with all diagnostic tests , the final diagnosis should be based on correlation of test result with other clinical symptoms & findings

Bibliography :

1. Kilen. G. (1976: Mannual of Clinical Immunology ASM, 264
2. Rantz, L.d. Di. caprio, J.M. Randall, E., (1952); Am. J.Med.sci24