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Expt 2 → Widal Test.

Introduction

- The WIDAL (Widely Investigated Diagnostic Assay Laboratory) test is a serological technique which tests for the presence of salmonella antibodies in the patients serum.

- It is used to diagnose typhoid and para-typhoid in endemic areas.
- Salmonella is identified in food poisoning conditions, when the investigation is carried out of typhoid
- The patients serum is tested for following antigens:
  - Salmonella typhi 'O' antigen (somatic cells)
  - Salmonella typhi 'H' antigen (Flagellar cells)
  - Salmonella typhi 'AH' antigen
  - Salmonella paratyphi 'BH' antigen

Requirements

(i) Fresh serum, stored at 2-8° serum should not be heated or individual.

(ii) The complete kit containing five vials containing stained salmonella antigen

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Teacher's Signature :_____

- S. Typhi → O antigen
- S. Typhi → H antigen
- S. Paratyphi → AH antigen
- S. Paratyphi → BH antigen

(iii) Widal positive Control
(iv) Widal test
(v) Application Stick

Principle

Antibodies in serum, produced in response to exposure to salmonella organisms will agglutinate bacterial suspension which carries homologous antigens.

Sample

Fresh serum, store at 2-8° in case of any delay in testing. Serum should be clear and should not be heated or inactivated.

Procedure : Rapid Slide (Screening) Test :-

1. Clean the glass slide and wipe it free of water.
2. Place one drop of undiluted serum in each of the 1st four circles (1-4) & one drop

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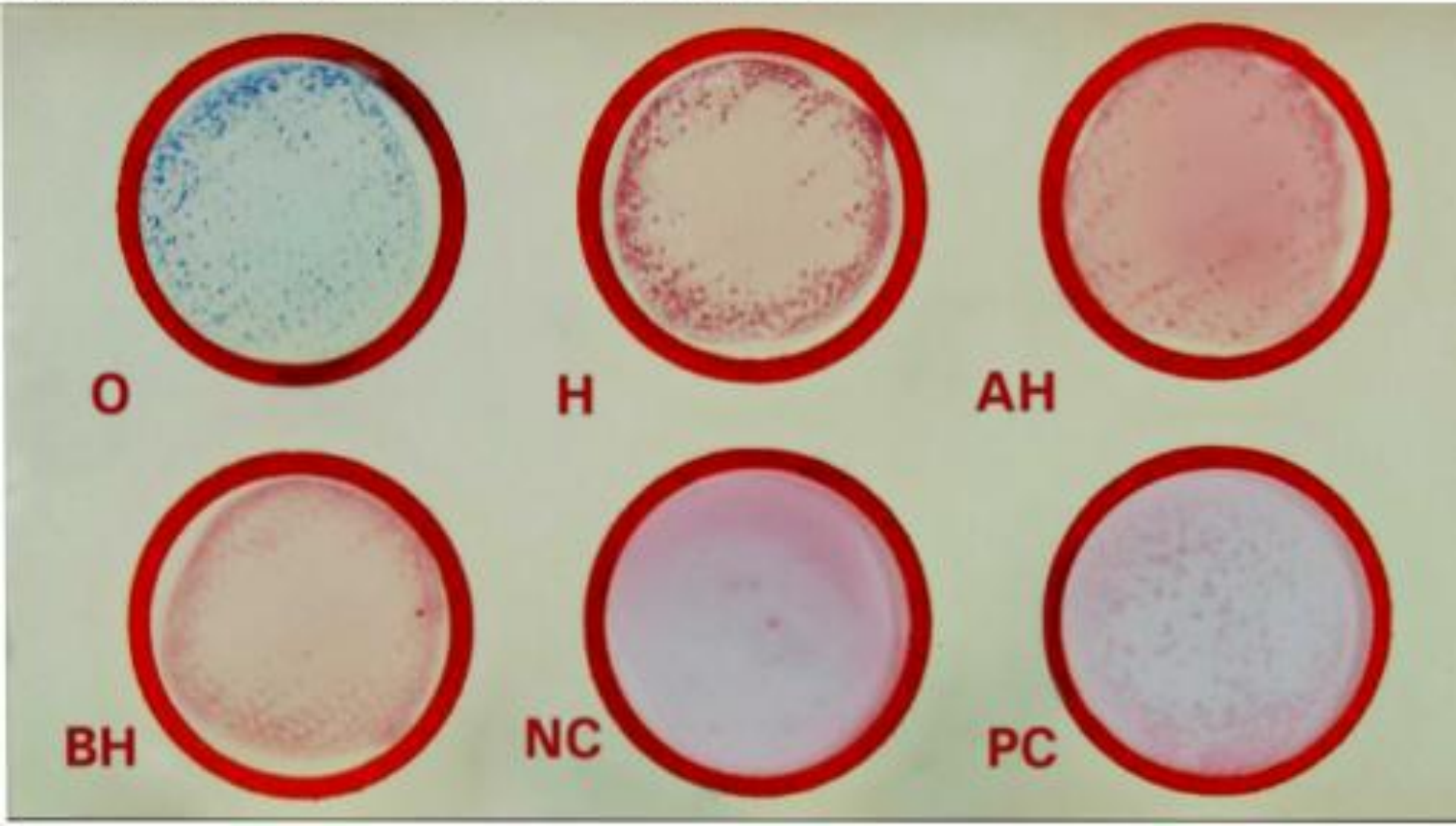

of positive control serum and negative control serum in each of the last two circles (5 & 6).

3. Place one drop of antigens O, H, AH, BH in circle 1, 2, 3 and 4 respectively and O or H antigen in 5 and 6.
4. Mix the contents of each circle with separate wooden applicator stick and spread to fill the whole area of the individual circle.
5. Rotate the slide for one minute and observe for agglutination reaction.

Result

Positive result :- Presence of agglutination rxn. which is observed b/w the antibodies of patient's serum and the antigens of salmonella organism - O, H, A-H, B-H antigen suspension.

Negative result :- Absence of agglutination reaction indicating the absence of antibodies in patients serum, thereby indicating the absence of salmonella infection in the patient.