

Agglutination Reaction

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Objectives



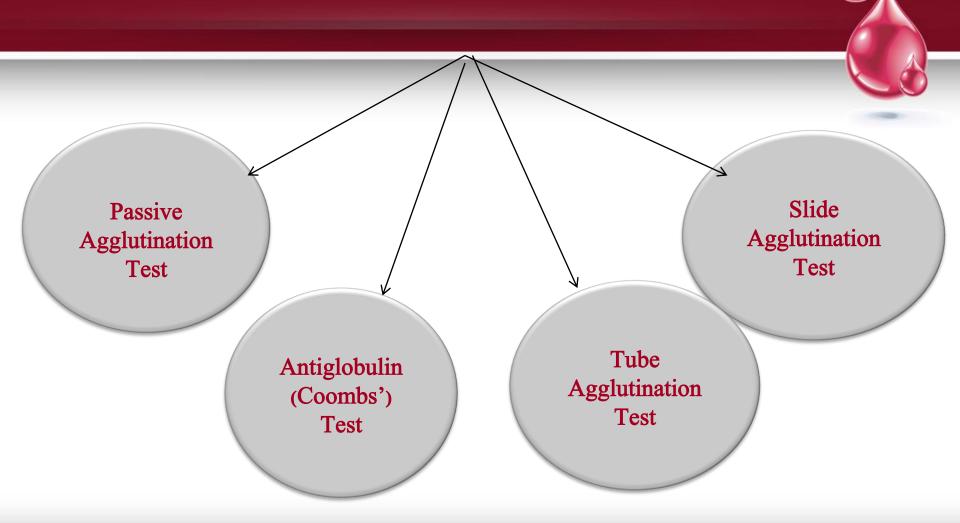
• Discuss principle and applications of agglutination reactions and their uses.

Introduction



Antibodies are mixed with their corresponding antigens on the surface of large, easily sedimented particles such as animal cells, erythrocytes, or bacteria, the antibodies cross-link the particles, forming visible clumps. This reaction is termed agglutination.

Agglutination Reaction



1. Slide Agglutination



- A drop of sterile saline is placed on one of the divisions of the slide or plate and is emulsified in it bacterial culture.
- The diagnostic serum is taken and is then mixed the latter.
- The mixing is completed by tilting the slide to and from.

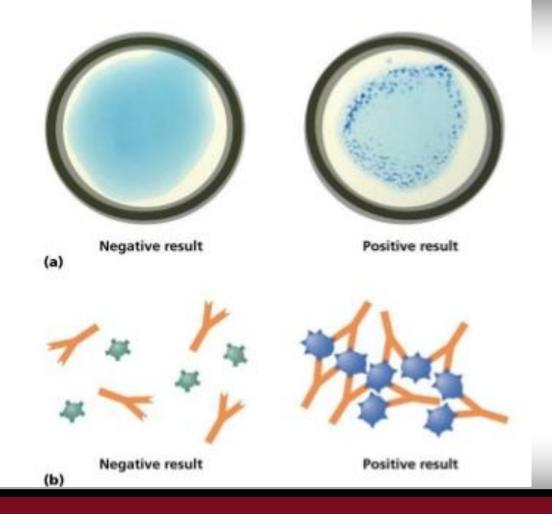
1. Slide Agglutination



- Distinct clumping within 60 seconds is a positive result.
- On another area of the slide or plate, in parallel with the test.
- The control test is performed, adding only saline, instead of serum, to the bacterial suspension. If clumping takes place, the bacterial suspension is autoagglutinable and the reaction with the serum must be disregarded.

1. Slide Agglutination





1. Uses of Slide Agglutination



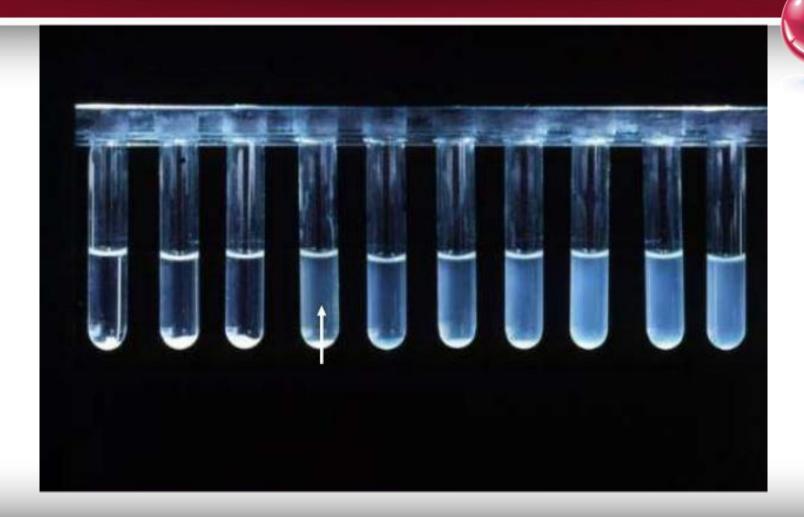
- 1. For the identification of unknown bacterial cultures.
- 2. Very rapid: It requires only small quantities of culture and serum.
- 3. Also the method for blood grouping and cross-matching.

2. Tube Agglutination



- Serum from a patient thought to be infected with a given bacterium is serially diluted in a series of tubes to which the bacteria is added.
- The last tube showing visible agglutination will reflect the serum antibody titer of the patient.
- The reciprocal of the greatest serum dilution that elicits a positive agglutination is known as the agglutinin titer.

2. Tube Agglutination



2. Uses of Tube Agglutination



- Tube agglutination is routinely employed for the serological diagnosis of typhoid, brucellosis and typhus fever.
- 1. Widal test: Used for the diagnosis of enteric fever.
- 2. Tube agglutination test for brucellosis.
- 3. Weil-Felix reaction: Weil-Felix reaction for serodiagnosis of typhus fevers is heterophil agglutination test.

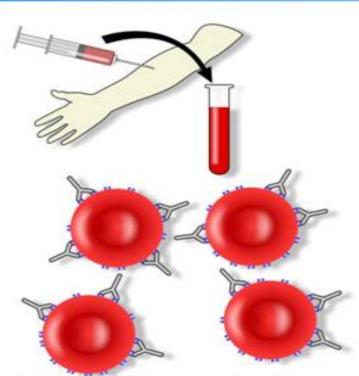
3. Antiglobulin (Coombs') Test

The antiglobulin test for Rh antibodies was originally devised by British immunologist, RRA coombs.

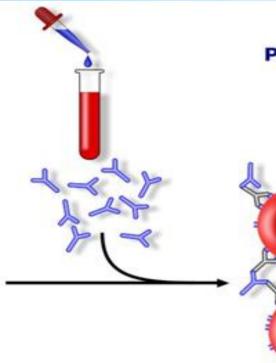
3. Antiglobulin (Coombs') Test



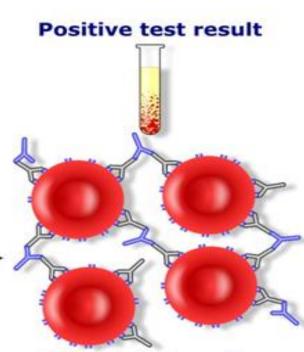
Direct Coombs test / Direct antiglobulin test



Blood sample from a patient with immune mediated haemolytic anaemia: antibodies are shown attached to antigens on the RBC surface.



The patient's washed RBCs are incubated with antihuman antibodies (Coombs reagent).

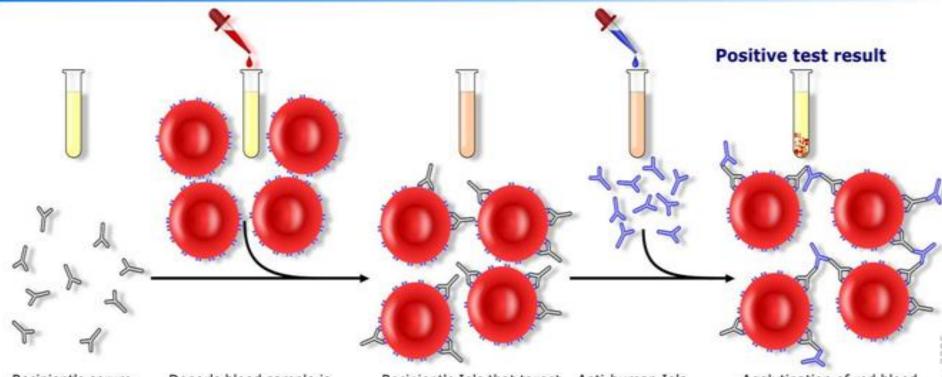


RBCs agglutinate: antihuman antibodies form links between RBCs by binding to the human antibodies on the RBCs.

3. Antiglobulin (Coombs') Test



Indirect Coombs test / Indirect antiglobulin test



Recipient's serum is obtained, containing antibodies (Ig's). Donor's blood sample is added to the tube with serum. Recipient's Ig's that target the donor's red blood cells form antibody-antigen complexes. Anti-human Ig's (Coombs antibodies) are added to the solution. Agglutination of red blood cells occurs, because human Ig's are attached to red blood cells.

3. Uses of Antiglobulin (Coombs') Test



Used for demonstrating any type of incomplete or nonagglutinating antibody, as e.g. in brucellosis.

4. Passive Agglutination Test



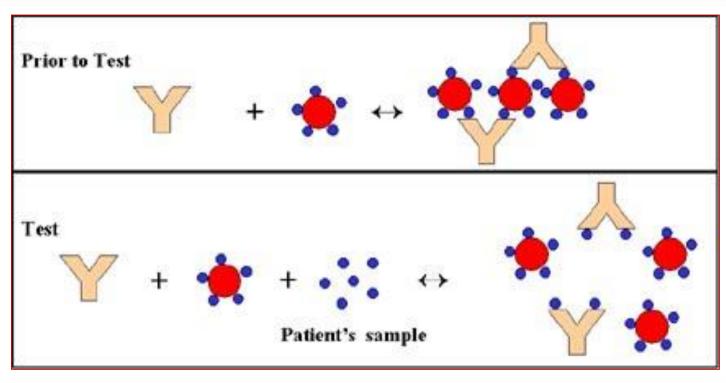
 A precipitation reaction can be converted into agglutination reaction by coating soluble antigen on to the surface of carrier particles. The commonly used carrier particles are red cells, latex particles or bentonite. Such test is more convenient and more sensitive for detection of antibodies. Such tests are known as passive agglutination tests.

4. Passive Agglutination Test

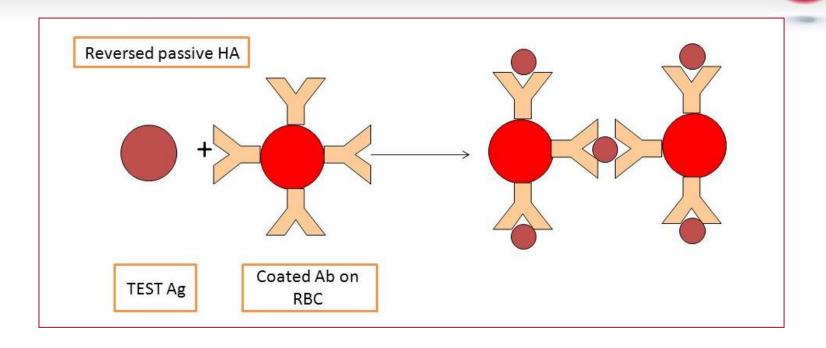
• Reversed passive agglutination: When instead of antigen, the antibody is adsorbed to carrier particles in tests for estimation of antigens, the technique is known as reversed passive agglutination.

4. Passive Agglutination Test





4. Reversed Passive Agglutination Test



4. Uses of Passive Agglutination Test



- Co-agglutination can be used for detecting the presence of antigens in serum, urine and CSF.
- Several commercial suppliers have prepared co-agglutination reagents for identification of antigens of various streptococcal groups, including Lancefield groups A, B, C, D, F, G, and N; *Streptococcus pneumoniae; Neisseria Meningitidis; N. Gonorrhoeae; and Haemophilus Influenzae* types A to F grown in culture.



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