

Rose Bengal Test

Asst.Lec.safa saad

Asst.Lec.sally saadi

The Rose Bengal test (RBT)

- The Rose Bengal test (RBT) is a simple, rapid slide-type agglutination assay performed with a stained *B. abortus* suspension at pH 3.6-3.7 and plain serum.
- It is often used as a screening test in human brucellosis and would be optimal for small laboratories with limited means.
- False-negative reactions occur especially in the early stages of acute infection.

Brucellosis in humans

- Brucellosis in humans is usually associated with the consumption of unpasteurized milk and soft cheeses made from the milk of infected animals, primarily goats, infected with *Brucella melitensis* and with occupational exposure of laboratory workers, veterinarians, and slaughterhouse workers.

Introduction:

- Brucellosis is a worldwide zoonosis caused by the bacterial genus *Brucella*.
- These organisms localize in the reproductive organs of host animals, causing abortions and sterility.
- They are shed in large numbers in the animal's urine, milk, and placental fluid.

Agglutination test used to diagnose brucellosis in humans and animals

Procedure:

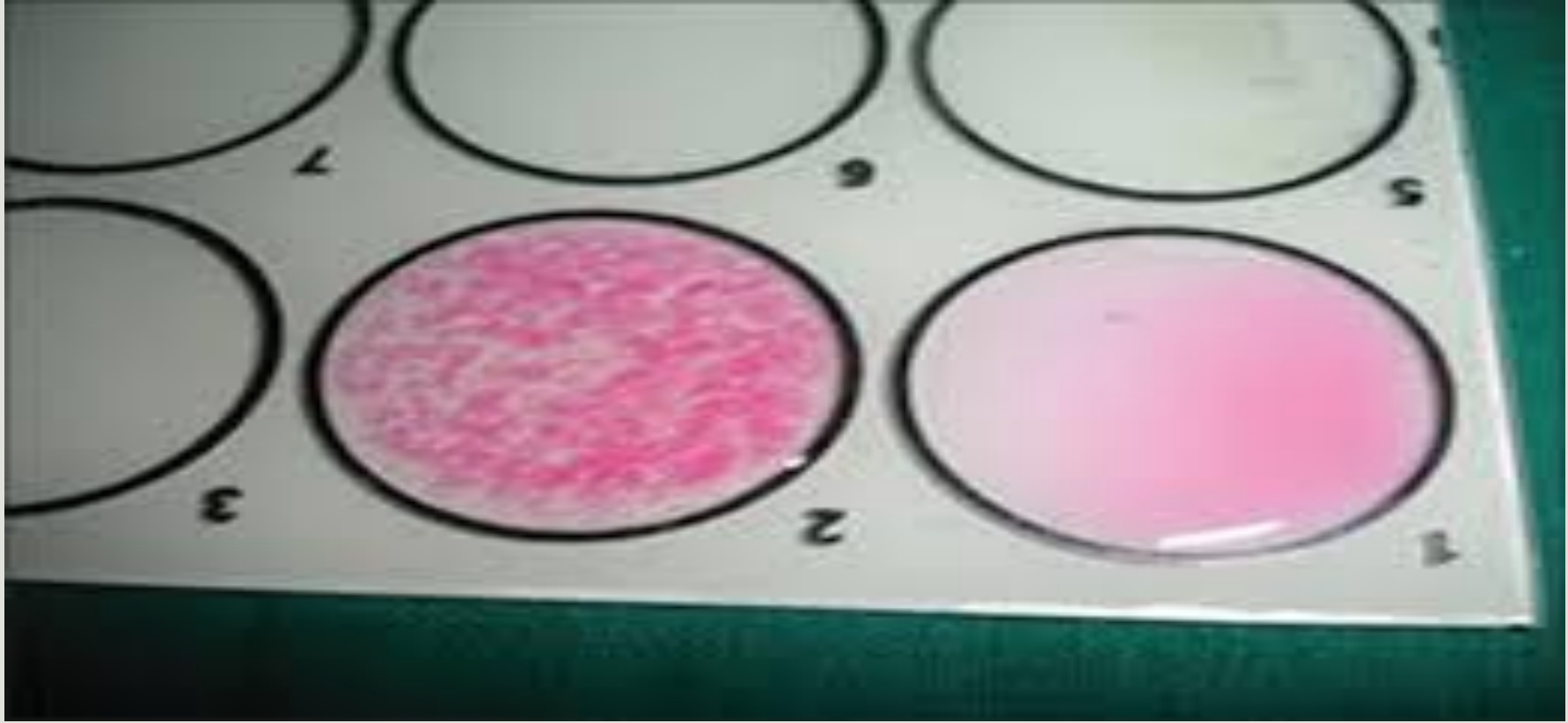
- Allow the reagents and sample to reach room temperature.
- Place 50 μ L of the sample and one drop of each positive and negative control into separate circles on the slide test.
- Shake the Rose Bengal reagent gently before using and add a drop of this reagent next to the sample to be tested.
- Mix both drops with a stick, spreading them over the entire surface of the circle. Use different stirrers for each sample.
- Rotate the slide with a mechanical rotator at 80-100 rpm for 2 minutes, and read the results (this is the optimum time limited).

Principle of test

- Ab content of patient's serum is measured by adding a constant amount of Brucella abortus colored Ag to serially diluted serum.
- High Ab titers above 80 units are considered clinically significant.
- Could be **Quantitative** or **Qualitative** test

Reading the result & Interpretation:

- No agglutination= absence of specific antibodies
- Agglutination (even slight) = presence of specific antibodies
 - If agglutination appear after 15 seconds = (1:640)
 - If agglutination appear after 30 seconds = (1:320)
 - If agglutination appear after 1 min. = (1:160)
 - If agglutination appear after 1.30 min. = (1:80)
- Patient history should be taken into account before giving the result.
- This test is a screening test only for the detection of Brucella agglutinins. If result is positive it must be confirmed by other serological tests for Brucellosis.



Laboratory diagnosis of Brucellosis:

No.	Methods	Time Consuming
<u>Serological level</u>		
1	Rose Bengal test by Rapid Slide agglutination (screening) test	2 min
2	Rose Bengal test by Tube Agglutination test	2-4 hours
3	Brucella IgG/IgM by Immunochromatographic assay	5 minutes
4	2 Mercaptoethanol Test (ME)	15 minutes
5	ELISA (IgG/ IgM)	45 minutes – 2 hours
6	ECL (IgG/ IgM)	45 minutes – 2 hours
<u>Molecular level</u>		
7	PCR (Polymerase Chain Reaction)	7-10 days

Thanks

