BRUCELLA MELITENSIS SLIDE AND TUBE TEST KIT

Code	Product Name	Pack Size
SE051A	Brucella Melitensis Slide and Tube Test Kit	1 x 5 ML

Intended Use

For qualitative determination of febrile antibodies in human serum.

Principle of the method

The Bacterial Antigens is a slide and tube agglutination test for the qualitative and semi-quantitative detection of antibodies Brucella in human serum. The reagents, standardized suspensions of killed and stained bacteria, agglutinate killed and stained bacteria agglutinate when mixed with samples containing the homologous antibody.

Clinical Significance

Febrile diseases diagnostic may be assessed either by micro organism isolation in blood, stools or urine, or by titration of specific antibodies, somatic (O) and flagella (H). The detection of these antibodies forms the basis for the long- established Widal test. This test dictates that a serum with high levels of agglutinating antibodies to O and H >1/100 is indicative of the infection with these microorganism .

Contents

Reagent 1 : Brucella Melitensis Antigen

Different correlative letters to the reference correspond to different variables of presentation.

Reagents Composition

Bacterial Antigens: Suspensions of Brucellas in glycine buffer, pH 8.2. Preservative.

Controls: Animal serum. Preservative

Calibration

There is not any International Reference for the sensitivity standardization of these reagents. For this reason, Beacon uses an internal control that contains animal serum with antibodies Brucellas titered with commercial reagents of certified quality.

Preparation And Stability

Antigen suspensions: Ready to use. It should be gently mixed before to use. Always keep vials in vertical position. If the position is changed, gently mix to dissolve aggregates that may be present.

Controls: Ready to use.

Reagents deterioration: Presence of particles and clumps. All the components of the kit are stable until the expiration date on the label when stored at +2-+8ºC. Protected from light and contaminations. Do not freeze.

Additional Equipment

Mechanical rotator adjustable to 80-100 r.p.m.- Heater at +37ºC. Vortex mixer. - Pipettes 50 µL.

BEACON

Sample

Fresh serum. Stable 8 days at +2-+8ºC. The samples with presence of fibrin should be centrifuged before testing. Do not use highly hemolized or lipemic samples.

Procedure

A. Slide Agglutination Method (Qualitative Test)

1. Bring the reagents and samples to room temperature. The sensitivity of the test may be reduced at low temperatures.
2. Place 50 µL of the sample to be tested and 1 drop of each control into separate circles on the slide test.
3. Mix the antigen vial vigorously or on a vortex mixer before using. Add 1 drop (50 µL) of antigen to each circle next to sample to be tested.
4. Mix with a Disposable Applicator Sticks and spread over the entire area enclosed by the circle.
5. Place the slide on a mechanical rotator at 80 - 100 r.p.m., for 1 minute.

B. Slide agglutination method (titration)

1. Using a micropipette, deliver 80, 40, 20, 10 and 5 µL of undiluted serum into separate circles of the slide test.
2. Place 1 drop (50 µL) of the antigen to each circle next to the sample to be tested.
3. Mix with a Disposable Applicator Sticks, and spread over the entire area enclosed by the circle.
4. Place the slide on a mechanical rotator at 80-100r.p.m., for 1 minute.

C. Tube agglutination method

1. Prepare a row of tube test for each sample as follows:

Dilutions	1/20	1/40	1/80	1/160	1/320	1/640	---
Sample(µL)	100	---	---	---	---	---	---
NaCL 9 g/L (mL)	1.9	1	1	1	1	1	---
	1 mL →	1 mL →	1 mL →	1 mL →	1 mL →	1 mL →	1 mL Discard

2. Prepare 2 tubes for Positive and Negative control: 0.1 ml Control + 0.9 ml NaCl 9 g/L.
3. Add a drop (50µL) of antigen suspension to each tube.
4. Mix thoroughly and incubate tube test at+37ºC for 24h.

Reading And Interpretation

Slide Agglutination Method

Examine macroscopically the presence or absence of clumps within 1 minute after removing the slide from the rotator comparing test results with control serums.

The reactions obtained in the slide titration method, are roughly equivalent to those which would occur in tube

With serum dilutions of 1/20, 1/40, 1/80, 1/160 and 1/320 respectively. If a reaction is found it is advisable to confirm the reaction and establish the titer by a tube test.

Tube Agglutination Test

Examine macroscopically the pattern of agglutination and compare the results with those given by all control tubes. Positive control should give partial or complete agglutination. Negative Control should not give visible clumping. Partial or complete agglutination with variable degree of clearing of the supernatant fluid is recorded as a positive. The serum titer is defined as the highest dilution showing a positive result.

Quality Control

Positive and Negative controls are recommended to monitor the performance of the procedure, as well as a comparative pattern for a better result interpretation. All result different from the negative control result, will be considered as a positive.

Reference Ranges

Brucellas: Titers ≥ 1/80 indicate infection.

The level of "normal" agglutinins to these organisms varies in different countries and different communities.

It is recommended that each laboratory establish its own reference range.

Performance Characteristics

All the performance characteristics of the Bacterial Antigens may be found in the corresponding Technical Report and They are available on request.

Interferences

Bilirubin (20 mg/dL), hemoglobin (10 g/L), lipids (10 g/L) and rheumatoid factors (300 IU/mL), do not interfere.

Limitations Of Procedure

False negative results can be obtained in early disease, immune-unresponsiveness, prozone (Brucelosis), and antibiotic treatment. (somatic).

Serological cross-reactions with Brucella have been reported in cases of infection or vaccination with some strains of Vibrio cholerae, Pasteurella, Proteus OX19 and Y. enterocolitica (serotype 9).

Notes

1. When testing for Brucella antibodies it is recommended to reduce sample volume to 20 µL in order to avoid prozone.
2. In some geographical areas with a high prevalence of febrile antibodies, it is recommended to dilute the sample ¼ in NaCl 9 g/L before to perform the assay.
3. A single positive result has less significance than the demonstration of a rising or falling antibodies titer as evidence of infection. A clinical diagnosis should not be made on findings of a single test result, but should integrate both clinical and laboratory data.

Bibliography

1. Edward J Young. Clinical Infectious Diseases 1995; 21: 283-290.
2. Coulter JBS. Current Pediatrics 1996; 6: 25-29.
3. David A et al. Currebt Opinion in Infectious Diseases 1994; 7: 616-623.
4. David Retal Current Opinion in Infectious Diseases 1993; 6: 54-62.
5. Bradley D Jones. Annu RevImmunol 1996;14:533 - 61.

Symbols Used On Labels

REF	Catalogue Number		Manufacturer
	See Instruction for Use	LOT	Lot Number
CONT	Content		Storage Temperature
	Expiry Date	IVD	In Vitro Diagnostics

BEA/24/BRM/SE/IFU VER-02
21/09/2025