

Brucella

Brucellosis in animals is generally typified by late-term abortions and inflammatory lesions in the male reproductive tract. *Brucella* is the cause of brucellosis, a true zoonotic disease (i.e. human-to-human transmission has not been identified). It is occupational disease of cattle rancher, dairy farmers, veterinarians, abattoir workers, meat inspectors, laboratory workers, hunters and travelers. Brucellosis has many different names both involving humans (Undulant Fever, Mediterranean Fever, Malta Fever, Rock Fever of Gibraltar and Gastric Fever) as well as animals (□ Bang's Disease, Enzootic Abortion, Epizootic Abortion, Slinking of Calves, Ram Epididymitis and Contagious Abortion).

Scientific Classification

Kingdom : Bacteria

Phylum: Proteobacteria

Class: Alpha Proteobacteria

Order: Rhizobiales

Family: Brucellaceae

Genus: ***Brucella***

Species

B. abortus
B. canis
B. melitensis
B. microti
B. neotomae
B. bovis
B. suis
B. pinnipediae

Etiology

The causative agent of brucellosis was isolated by **Sir David Bruce in 1886**, from a soldier in Malta. Brucellosis is caused by members of the bacterial genus *Brucella*. There are a few different species of *Brucella*, each with slightly different host specificity. *B. melitensis* which infects goat and sheep, *B. abortus* which infects cows, *B. suis* infects pigs, *B. ovis* infects sheep and *B. neotomae*. Recently a new species was discovered in marine mammals: *B. pinnipediae*.

Habitat

Brucella species are obligate parasites and can survive in suitable environment for long period of time. They survive in contaminated soil, urine, animal faeces and manure for several weeks.

Morphology

Gram negative cocci, coccobacilli or short rods from 0.6 to 1.5 μm long by 0.5 to 0.8 μm wide. These are facultative intracellular pathogens. They occur singly, in pairs, short chains or small clusters. *In vivo*, the coccoid form is usually seen with organisms arranged in clusters within cytoplasm of infected cells. Flagella, endospore and capsules are absent although capsule like structures have been reported in preparations treated with antiserum.

Resistance physical and chemical agents

The organism is killed by pasturization, by heating at 70°C for 10 minutes and at boiling point within few seconds. In raw milk the organism is destroyed only with production of lactic acid. *Brucella* are sensitive to strong salt solution, phenol, formalin and other disinfectant. Freezing can not kill the bacteria.

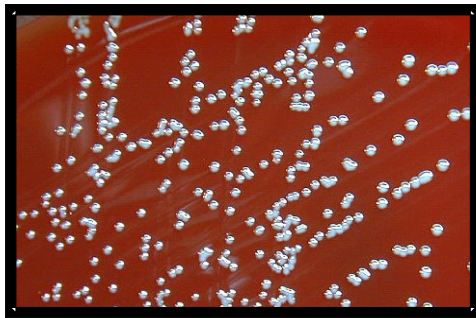
Epidemiology

Animal to animal transmission through the oral route, Contamination of the udder during milking and contact with aborted fetuses. The infected newborn lambs are also considered to be common factor for spreading disease. The venereal transmission of the disease occurs through infected male or contaminated semen. Animal to human transmission occurs by infected tissues. Contaminated materials must be handled under laminar flow hood in (biosafety 3 conditions). Transmission could be either by contaminated food, invasion by intact skin, inhalation of aerosols containing the bacteria and aerosol.

Cultural characteristics

The optimum temperature for growth is 37°C with 10% carbon dioxide tension and PH ranges from 6.8 to 7.2. Grown on high quality peptone based medium and is improved by addition of blood or serum. Serum dextrose agar is suitable solid medium. Good growth can be obtained in soya peptone broths such as trypticase soy or tryptose soya broth. Media for cultivation of *Brucella*. The available commercial basal medium used for isolation of *Brucella* organisms are *Brucella* medium

base, trypticase soy agar, Columbia agar, serum dextrose agar, albimi agar and glycerol dextrose agar. The addition of 2-5 % Bovine or Equine serum is necessary for the growth of strains such as *B. abortus* biovar 2. Appropriate antibiotics are added in order to suppress the growth of organisms other than *Brucella*. The most widely used medium is Farrell's medium, which is prepared by the addition of six antibiotics i.e., Polymyxin B sulphate, Bacitracin, Cycloheximide, Nalidixic acid, Nystatin, Vancomycin. After 48 hours of growth at 37°C on serum dextrose agar most *Brucella* strains will produce colonies 0.5 to 1.0 mm in diameter. These are raised, convex and have a circular outline. In case of smooth strain the colonies are transparent and pale yellow like "droplet of honey" appearance with slimy surface when viewed in transmitted light. Non smooth colonies tend to be more opaque with a granular surface and can vary in colour from dead white to dark brown; off white to buff are probably the most frequently seen colour.



Brucella colonies on blood agar

Biochemical Characteristics

Except *B. ovis* other species of *Brucella* ferment glucose. They do not liquefy gelatin and are negative for Methyl red and Voges-Proskauer test but catalase and oxidase positive. Some strains produce hydrogen sulphide but not indole. They also reduce nitrate to nitrite and breakdown urea.

Antigenic structure

A and M of brucellae are the two major somatic antigens that have been recognized so far. Their ratio varies in different species. The A:M ratio in *Brucella abortus* is about 20:1 and in *B. melitensis* is 1:20. Serological cross reactions of brucellae have been demonstrated with other species of bacteria like *Escherichia coli* (serotype 0116 and 0157), *Francisella tularensis*, *Vibrio cholerae*, *Salmonella* serotypes and *Yersinia enterocolitica*.

Pathogenicity

In all the susceptible host brucella organisms grow intracellularly producing a bacteremic phase followed by localization in the reticulo-endothelial system. The organism has got affinity for the gravid uterus for presence of erythritol, which favours multiplication. Sexually immature animals are often less susceptible to brucella infection. In female abortion occurs in the third trimester with retained placenta. Orchitis and epididymitis are common in male. Respiratory distress and lung infections are also observed. Brucellosis in boar is characterised by unilateral or bilateral orchitis, lameness,

posterior paralysis, spondylitis, metritis and abscesses formation.

Diagnosis

Diagnosis of *Brucella* infection is based on history of abortion and characteristic lesions in the placenta, isolation and identification of the organisms, serological and laboratory animal inoculation test. Specimens such as aborted foetus, placenta, pieces of lungs, liver, cotyledons, spleen of aborted foetus, serum, semen, milk, vaginal swabs, swabs from male reproductive organs are to be collected for isolation of bacteria in suitable media. On staining smears prepared from infected materials by *Brucella* differential staining techniques, the organism occurs characteristically in clumps intracellularly and extracellularly. Fluorescent antibody techniques can also be employed to demonstrate the organism. The serological test like rose bengal test (RBT), complement fixation test (CFT), enzyme linked immuno sorbent assay (ELISA), milk Ring Test (MRT), standard agglutination test (SAT), rapid plate agglutination test, whey agglutination, indirect immunofluorescence and complement fixation test are useful for diagnosis of *Brucella* infection. In addition Polymerized chain reaction (PCR), restriction fragment length polymorphism (RFLP) and pulse field gel electrophoresis (PFGE) have also been developed recently for detection and identification of *Brucella* species. Infected material injected subcutaneously to male guinea pig develop orchitis (Straus reaction) with production of suppurative lesions.

References

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