BRUCELLOSIS

a. Clinical and laboratory features
   a. Size: 0.5 – 0.7 µm in diameter and 0.6 – 1.5 µm in length. 1

   b. Modes of transmission
      i. Ingestion, particularly eating dairy products prepared with unpasteurized milk (soft cheese, yogurt, ice cream) 2 or under cooked meat such as liver. 3
      ii. Aerosolization, particularly among laboratory workers (10% - 100% infection rate). 4
      iii. Contamination of conjunctiva or traumatized skin by infected animal products. 5

   c. Virulence
      i. Intracellular survival of at least 15% - 30% of ingested organisms. 1
      ii. After entering the human body and being taken up by local tissue lymphocytes, Brucellae are transferred through regional lymph nodes into the circulation and are subsequently seeded throughout the body, with tropism for the reticuloendothelial system. 1

   d. Spectrum of Disease
      i. Systemic disease that can involve any organ or system of the body. 1
      ii. The infective dose of Brucella, especially that of B. melitensis, is very low (10 organisms). 1
      iii. The incubation period is usually between seven days and three months, although as long as 10 months have been reported. 6
      iv. Human brucellosis is known for presenting with protean manifestations. 7
      v. The symptoms and clinical signs most commonly reported are fever, fatigue, malaise, chills, sweats, headaches, myalgia, arthralgia and weight loss. 1,7-11
      vi. Some other reported cases have presented with joint pain 7,8, low back ache 7,8, involuntary movements of limbs 8, burning feet 8 or ischemic heart attacks 12.
      vii. “Typically, no or few objective signs are apparent that specifically point to brucellosis. Enlargement of the liver, spleen and/or lymph nodes may occur as may other signs referable to almost any other organ system. These febrile patients may be referred to as patients with pyrexia of unknown origin or the symptoms and signs are confused with those of other diseases such as enteric fever, malaria, rheumatic fever, tuberculosis, cholecystitis, thrombophlebitis, fungal infection, autoimmune disease and tumors.” 1,7-9,13
      viii. Human brucellosis is known for complications. Complications can be very diverse depending on the specific site of infection. 1 Osteoarticular, genitourinary, gastrointestinal, nervous, cardiovascular, skin and mucous membranes and respiratory complications are observed. Bone and joint involvement is the most frequent complication of brucellosis and occurs in up to 40% of cases in some series. 1 Three distinct forms exist; peripheral arthritis, sacroiliitis and spondylitis. Peripheral arthritis is the most common and is non-erosive, since it usually involves the knees, hips, ankles and wrists in the context of acute infection. 7,8,14,15
Epididymoorchitis is the most frequent genitourinary complication in men and may be confused with testicular cancer or tuberculosis. Brucellosis during pregnancy poses a substantial risk of spontaneous abortion or intrauterine transmission of infection to the infant. "As the largest organ of the reticuloendothelial system, the liver is probably involved in the majority of cases of brucellosis even though liver function tests are normal or values are usually only mildly abnormal." Invasion of central nervous system (CNS) occurs in about 5-7% of the cases of Brucella melitensis infection. Meningitis, encephalitis, meningoencephalitis, meningovascular disease, brain abscesses and demyelinating syndromes have all been reported. Brucellae are rarely isolated from cerebrospinal fluid (CSF), but antibodies to Brucella species are present in the serum and CSF in the majority of cases. Brucella endocarditis occurs in less than 2% of cases but accounts for the majority of Brucella-related deaths. Early recognition, adequate antibiotic treatment and the absence of signs of heart failure can guide the practitioner toward prolonged, conservative treatment. Complications involving the skin, although rare, are reported in the literature. A multinational review of cases by Pappas et al and a recent publication by Mantur and coworkers indicate that the pulmonary involvement is not rare.

e. Stages of the disease (CDC technical information, http://www.cdc.gov/ncidod/dbmd/diseaseinfo/brucellosis_t.htm)
   - Acute (< 2 months)
   - Subacute/undulant (2-12 months)
   - Chronic (> 1 year)

Bacterial Load

Mouse

Study 1. Kahl-McDonagh and coworkers studied the kinetics of clearance of Brucella abortus 2308 and Brucella melitensis from BALB/c mice. Four or five female BALB/c mice were infected with aerosolized 2308 in a Madison aerosol chamber using three different chamber doses, 5 x 10^7 CFU/ml (A), 5 x 10^8 CFU/ml (B), or 5 x 10^9 CFU/ml (C). The initial lung colonization was evaluated immediately after challenge to determine the quantity of Brucella inhaled for each chamber dose. Mice were euthanized at 1, 2, 4, 6, or 8 weeks postchallenge to determine the numbers of Brucella persisting in the lungs, livers, and spleens. The recovery of organisms is plotted as the total CFU/organ (means ± standard errors). The lower limit of detection was ≥5 CFU.

Study 2: Smither and coworkers evaluated the kinetics of infection in BALB/c mice following exposure to aerosolized B. suis 1330. Groups of mice were exposed to retained doses of 10^2, 10^3, or 10^4 CFU of B. suis 1330. Half of the mice in each group were immunized s.c. with the B.melitensis Rev.1 live vaccine 30 days prior to challenge with B. suis. The remaining mice remained nonimmunized and served as control. At 14, 21, and 28 days postchallenge, all mice
were culled, organs were homogenized, and the \textit{B. suis} 1330 loads in spleens, lungs, and livers were determined.\textsuperscript{24}

Based on the results shown in Figures 1 and 2 from Kahl-McDonagh et al\textsuperscript{23} and Figure 2 of Smither et al.\textsuperscript{24}, the following average pathogen loads were recorded for the mouse lung, liver and spleen during the acute phase of the disease:

- lung: 5.0 log\textsubscript{10} CFU/g (lung of a mouse $\sim$1 g)
- liver: 3.2 log\textsubscript{10} CFU/g (mouse liver $\sim$2 g)
- spleen: 5.5 log\textsubscript{10} CFU/g (mouse spleen $\sim$0.125g)

**Blood/plasma concentrations**

It is difficult to detect and determine the concentration of \textit{Brucella} in blood. There were numerous attempts to culture bacteria from the blood of \textit{B. melitensis} and \textit{B. suis} infected mice sampled at multiple times post aerosol infection (1 day to 4 weeks after) and after different challenge levels. The single success was attained using blood of \textit{B. suis} infected mice exposed to a high aerosol dose (10\textsuperscript{4} CFU) at 2 weeks post-infection. One of the reasons of failure is that \textit{Brucella} is hard to culture, requiring quite a lot of sub-culturing and lengthy incubation to see it (2-3 weeks are possible not long enough). The main reason, though, might be that \textit{Brucella} is so transient in the blood that by taking a single sample one could quite easily miss it. In larger animals, a surge of bacteremia is likely to occur around an abortion event. Obviously the mouse is not the best model to work with in this case. In humans it would be the same: there might be a high bacterial load at one point but not continuously, so hitting the right time is difficult (Dr. Sophie Smither, Defence Science and Technology Laboratory, UK, personal communications).

**References**