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Brucellosis

Last Modified: May 2008

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Overview

- Organism
- History
- Epidemiology
- Transmission
- Disease in Humans
- Disease in Animals
- Prevention and Control
- Actions to Take

In today's presentation we will cover information regarding the organism that causes brucellosis and its epidemiology. We will also talk about the history of the disease, how it is transmitted, species that it affects (including humans, if applicable), and clinical and necropsy signs observed. Finally, we will address prevention and control measures for brucellosis, as well as actions to take if brucellosis is suspected.

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The Organism

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Brucella spp.

- Gram negative, coccobacilli bacteria
- Facultative, intracellular organism
- Environmental persistence
  - Temperature, pH, humidity
  - Frozen and aborted materials
- Multiple species

This organism is an aerobic, small, Gram-negative coccobacillus or short rod that can persist in the environment invariably depending on temperature, pH, and humidity. *Brucella* spp. can persist indefinitely if frozen or protected in aborted fetuses or placentas. It is a facultative, intracellular pathogen and thus requires prolonged treatment with clinically effective antibiotics.

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Species	Biovar/Serovar	Natural Host	Human Pathogen
B. abortus	1-6, 9	cattle	yes
B.melitensis	1-3	goats, sheep	yes
B. suis	1, 3	swine	yes
	2	hares	yes
	4	reindeer, caribou	yes
	5	rodents	yes
B. canis	none	dogs, other canids	yes
B. ovis	none	sheep	no
B. neotomae	none	Desert wood rat	no
B. maris (?)		marine mammals	?

Six named species occur in animals. One or more unnamed species of *Brucella* have been found in marine mammals. Formal names proposed for marine mammal isolates are *B. maris* for all strains, or *B. pinnipediae* for strains from pinnipeds (seals, sea lions, and walruses) and *B. cetaceae* for isolates from cetaceans (whales, porpoises, and dolphins). Some species of *Brucella* contain biovars. Five biovars have been reported for *B. suis*, three for *B. melitensis*, and up to nine for *B. abortus*. Each *Brucella* species is associated most often with certain hosts. *B. abortus* usually causes brucellosis in cattle, bison, and buffalo. *B. melitensis* is the most important species in sheep and goats, but *B. ovis* can also cause infertility in rams. *B. canis* causes disease almost exclusively in dogs. *B. neotomae* is found in rodents, but has not been linked to disease. *B. suis* contains more diverse isolates than other *Brucella* species, and these isolates have broader host specificity. *B. suis* biovars 1, 2, and 3 are maintained in pigs; European hares are also a reservoir for biovar 2. Biovar 4 mainly affects reindeer and caribou, and is not normally found in pigs. This biovar was formerly known as *B. rangiferi*. Biovar 5 occurs in rodents. Marine mammal *Brucella (Brucella maris?)* has recently been discovered and while it has not been fully characterized, it is thought to infect humans. Order of pathogenicity to humans: *B. melitensis*, *B. suis* (biovars 1,3, and 4), *B. abortus*, and rarely *B. suis* biovar 2, *B. canis,* and marine mammal *Brucella*.

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The Many Names of Brucellosis

Human Disease	Animal Disease
• Malta Fever	• Bang's Disease
• Undulant Fever	• Enzootic Abortion
• Mediterranean Fever	• Epizootic Abortion
• Rock Fever of Gibraltar	• Slinking of Calves
• Gastric Fever	• Ram Epididymitis
	• Contagious Abortion

Due to its illustrious history, brucellosis has many different names involving both humans and animals. Undulant Fever for humans and Bang's Disease for animals are the two most widely recognized common names.

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History

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History of Malta Fever

- 450 BC: Described by Hippocrates
- 1905: Introduction into the U.S.
- 1914: *B. suis* Indiana, United States
- 1953: *B. ovis* New Zealand, Australia
- 1966: *B. canis* in dogs, caribou, and reindeer

In his book Epidemics, Hippocrates first described a condition of reccurring fever and death with a duration of 4 months in 450 B.C. Undulant fever did not enter into the United States until 1905 through the shipping of 65 Maltese goats on the *S.S. Joshua Nicholson. B. suis* was isolated in 1914 by Traum in the U.S. from aborting swine in Indiana. *B. ovis* was isolated in 1953 from sheep with ram epididymitis in New Zealand and Australia. *B. canis* was discovered in 1966 from dogs, caribou, and reindeer.

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Sir William Burnett (1779-1861)

- Physician General to the Navy
- Differentiated the various fevers affecting soldiers

The island of Malta was given to the Knights of the Order of St. John in 1530, and contagious fevers were noted from that time well into the 19th century. During the 17th and 18th centuries there were numerous reports of undulant fevers from all over the Mediterranean and most were given local names (Mediterranean fever, Rock fever of Gibraltar, Cyprus fever, Danube fever). Sir William Burnett was a physician to the British Navy in 1810 and was the first person to differentiate between the various fevers affecting seamen in the Mediterranean. It is thought that Malta became such an important center for the study of undulant fever because many British troops were sent there to recuperate following the Crimean War (1853-1856), along with skillful medical doctors utilizing clinical thermometers to monitor the disease progression.

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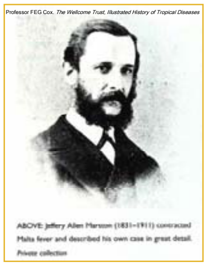

Jeffery Allen Marston

- Contracted Malta fever
- Described his own case in great detail

J.A. Marston was an army surgeon (British) who, after contracting the Malta fever, wrote the first detailed account of the disease (his own illness). He was afflicted with an irregular fever for 30 to 90 days, gastrointestinal symptoms, and muscle and joint pains.

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Professor FEG Cox. The Wellcome Trust, Illustrated History of Tropical Diseases

Sir David Bruce (1855-1931)

- British Army physician and microbiologist
- Discovered *Micrococcus melitensis*

The microorganism responsible for Malta fever was discovered by a British Army physician, Sir David Bruce, on July 9, 1887, which he called *Micrococcus melitensis*. It was isolated from the spleen of a British soldier who had died of the disease. He also identified that the organism grew best at higher temperatures and speculated that this accounted for the increased frequency of cases in hot summer months. He later established goats as the main reservoir for infection by identifying the organism in their blood, urine, and milk. This discovery helped explain the epidemiology of the disease. For example, officers were three times more likely to become ill because they drank more milk than private soldiers, and large numbers of cases were found in hospitals where milk was widely distributed.

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Bernhard Bang (1848-1932)

- Danish physician and veterinarian
- Discovered *Bacterium abortus* could infect cattle, horses, sheep, and goats

A Danish physician and veterinarian, Bernhard Bang discovered *Bacterium abortus* in 1897 while investigating contagious abortion that had been affecting cattle in Denmark for over a century. He also discovered the organism affected horses, sheep, and goats. Thus the disease became known as "Bang's disease".

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History

- Alice Evans, American bacteriologist
  - Credited with linking the organisms
  - Similar morphology and pathology between:
    - Bang's *Bacterium abortus*
    - Bruce's *Micrococcus melitensis*
- Nomenclature today credited to Sir David Bruce
  - *Brucella abortus* and *Brucella melitensis*

The connection between animals and humans was discovered by Alice Evans, an American bacteriologist in the 1920s. The morphology and pathology of the organism was very similar between Bang's *Bacterium abortus* and Bruce's *Micrococcus melitensis*. The name of Sir David Bruce has been carried on in today's nomenclature of the organisms.

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Transmission

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Transmission to Humans

- Conjunctiva or broken skin contacting infected tissues
  - Blood, urine, vaginal discharges, aborted fetuses, placentas
- Ingestion
  - Raw milk & unpasteurized dairy products
  - Rarely through undercooked meat

Brucella abortus and *B. suis* infections often are the result of occupational exposure through contact with infected tissues, blood, urine, vaginal discharge, aborted fetuses, and placentas. Typically, absorption occurs when the bacteria are exposed to areas of abraded or broken skin or the conjunctival sac of the eye. People are often infected with *B. suis* during the slaughter of swine or assisting in farrowing of infected sows. *Brucella melitensis* infection is primarily foodborne and results from consuming infected unpasteurized milk or dairy products.

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Transmission to Humans

- Inhalation of infectious aerosols
  - Pens, stables, slaughter houses
- Inoculation with vaccines
  - *B. abortus* strain 19, RB-51
  - *B. melitensis* Rev-1
  - Conjunctival splashes, injection
- Person-to-person transmission is very rare
- Incubation varies
  - 5-21 days to three months

Inhalation of infectious aerosols can also occur while cleaning out an infected animal's pen, or in a slaughter house. The current vaccines available are strain 19 and RB-51 for *B. abortus* immunization and Rev-1 for *B. melitensis.* Self inoculation could occur by a needle stick of a vaccine when handling animals, or when infected tissue or body fluids splash onto the conjunctiva of the eye. Conjunctival splashes are more likely to get a larger dose than an injection of a vaccine. Either route of exposure requires antibiotic therapy at treatment levels to prevent disease onset. Person to person transmission is very rare but has been reported as the result of blood transfusions, bone marrow transplants, and sexual contact between lab workers and their spouse. Rare congenital infections seem to result from transplacental transmission or the ingestion of breast milk. Congenital infections might also occur if the infant is exposed to organisms in the mother's blood, urine, or feces during delivery.

Incubation period in humans is from 5 to 21 days up to three months. This often adds to the difficulty of diagnosis due to the latency of clinical signs.

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Transmission in Animals

- Ingestion of infected tissues or body fluids
- Contact with infected tissues or body fluids
  - Mucous membranes, injections
- Venereal
  - Swine, sheep, goats, dogs
  - Fomites

B. abortus, *B. melitensis*, *B. suis*, and *B. canis* are usually transmitted between animals by contact with the placenta, fetus, fetal fluids, and vaginal discharges from an infected animal. Most or all *Brucella* species are also found in semen. Males can shed these organisms for long periods or lifelong. The importance of venereal transmission varies with the species. It is the primary route of transmission for *B. ovis*. *B. suis* and *B. canis* are also spread frequently by this route. *B. abortus* and *B. melitensis* can be found in semen, but venereal transmission of these organisms is uncommon. Some *Brucella* species have also been detected in other secretions and excretions including urine, feces, hygroma fluids, saliva, and nasal and ocular secretions. In most cases, these sources seem to be relatively unimportant in transmission; however, some could help account for direct non-venereal transmission of *B. ovis* between rams. *Brucella* can be spread on fomites including feed and water. In conditions of high humidity, low temperatures, and no sunlight, these organisms can remain viable for several months in water, aborted fetuses, manure, wool, hay, equipment, and clothes. *Brucella* can withstand drying, particularly when organic material is present, and can survive in dust and soil. Survival is longer when the temperature is low, particularly when it is below freezing.

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Epidemiology

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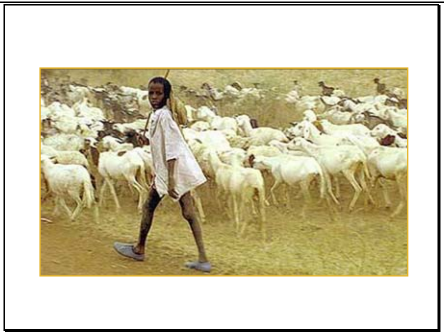

Pictured is a sheep herder in Africa. Sheep and other livestock are a major source for human infection. Placenta and aborted fetus are the most important sources of infection for other animals.

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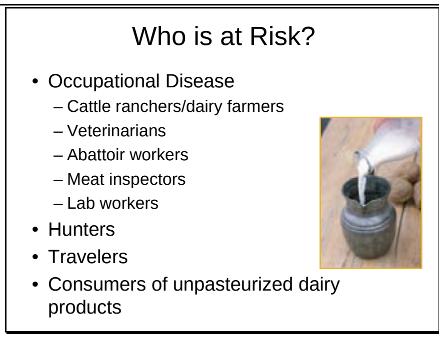

Brucellosis is predominantly an occupational disease of those working with infected animals or their tissues, but can also infect consumers of unpasteurized dairy products, and hunters who unknowingly handle infected animals. Illness in people can be very protracted and painful, and can result in an inability to work and loss of income; a socioeconomic disease. Travelers to areas with enzootic disease who consume local delicacies, such as goat, sheep, or camel milks or cheeses, may become infected.

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B. melitensis is particularly common in the Mediterranean. It also occurs in the Middle East, Central Asia, around the Arabian Gulf, and in some countries of Central America. This organism has been reported from Africa and India, but it does not seem to be endemic in northern Europe, North America (except Mexico), Southeast Asia, Australia, or New Zealand. There have been annual incidence reports of up to 78 cases per 100,000 people in the Mediterranean and Middle East. Greater than 550 cases have been reported from endemic areas that have no mandatory animal control measures. In some countries where animals are controlled, such as Southern Europe, incidence reports of 77 cases per 100,000 people are reported annually. A seroprevalence rate of 20% was identified on the Arabic Peninsula, with greater than 2% having active brucellosis. In the Middle East, *B. melitensis* may exceed the prevalence of *B. abortus* in cattle in some areas as it has emerged on some intensive dairy farm operations.

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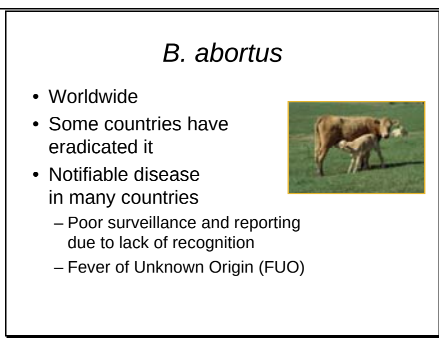

B. abortus is found worldwide in cattle-raising regions, except in Japan, Canada, some European countries, Australia, New Zealand, and Israel, where it has been eradicated. Eradication from domesticated herds is nearly complete in the U.S. *B. abortus* persists in wildlife hosts in some regions, including the Greater Yellowstone Area of North America. The actual incidence of infection may be higher by 10- 25% because cases often go unrecognized and get classified as fevers of unknown origin (FUO). It is a notifiable disease.

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B. suis

- Biovars 1 and 3
  - Worldwide problems where swine are raised
- Free
  - United Kingdom, Canada
- Eradicated
  - Holland, Denmark
- Low Incidence
  - Middle East, North Africa

In the past, *B. suis* was found worldwide in swine-raising regions. This organism has been eradicated from domesticated pigs in the U.S., Canada, many European countries, and other nations. However, it persists in wild and/or feral swine populations in some areas, including the U.S., Europe, and Queensland, Australia. Sporadic outbreaks are reported in domesticated herds or humans due to transmission from this source. *B. suis* continues to occur in domesticated herds in some countries of South and Central America (including Mexico) and Asia. *B. suis* biovars 1 and 3 are found worldwide, but other biovars have a limited geographic distribution. Biovar 2 occurs in wild boar in much of Europe. Biovar 4 (rangiferine brucellosis) is limited to the Arctic regions of North America and Russia, including Siberia, Canada, and Alaska. Biovar 5 (murine brucellosis) is found in the former USSR. Due to religious reasons, there are very low rates of incidence in the Middle East, North Africa, and India.

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B. suis

- Low Levels
  - United States and Australia
  - Persistent problem in feral swine
- Biovar 1
  - Established in cattle in Brazil and Columbia
- Biovar 2
  - Enzootic in wild hares in Europe

Australia and the United States have reduced the prevalence to a very low level, but it remains a persistent problem in feral swine in both countries, and eradication in this population appears unlikely. *B. suis* biovar 1 has also become established in cattle in Brazil and Columbia. *B. suis* biovar 2 is primarily found in Europe, and is enzootic in wild hares, posing a problem when swine have direct contact with this infected population.

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B. ovis

- Most sheep-raising regions
  - Australia
  - New Zealand
  - North America
  - South America
  - South Africa
  - Many European countries

B. ovis probably occurs in most sheep-raising regions of the world. It has been reported from Australia, New Zealand, North and South America, South Africa, and many countries in Europe.

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B. canis

- Poorly understood
- 1-19% prevalence in United States
- Rarely causes disease in humans

B. canis probably occurs throughout most of the world; however, New Zealand and Australia appear to be free of this organism. *B. canis* serosurveys have found prevalence rates in the U.S. from 1-19%, and up to 28% in Mexico. Cats are susceptible as well. *B. canis* rarely causes disease in humans.

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Brucella in Marine Mammals

- Culture-positive or seropositive animals
  - North Atlantic Ocean
  - Mediterranean Sea
  - Arctic, including Barents Sea
  - Atlantic and Pacific coasts of North America
  - Coasts of Peru, Australia, New Zealand, Hawaii, Solomon Islands, Antarctic

Brucella species also seem to be widespread in marine mammal populations. Culture-positive or seropositive animals have been found in the North Atlantic Ocean, the Mediterranean Sea, and the Arctic, including the Barents Sea. Infected or exposed animals have also been found along the Atlantic and Pacific coasts of North America; the coasts of Peru, Australia, New Zealand, and Hawaii; and in the Solomon Islands and the Antarctic.
As of July 2007, only four human infections with marine mammal *Brucella* have been reported. (Photo of a Weddell seal with brucellosis; taken from http://news.bbc.co.uk/1/hi/sci/tech/610444.stm)

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Brucellosis in U.S.: 1975-2006

Because the U.S. control and eradication program for brucellosis has nearly eliminated *B. abortus* infections among U.S. herds, the risk of a human acquiring the infection, either from occupational exposure to domestic livestock or from consuming contaminated food products, is small. A summary of cases is published each year in the Morbidity and Mortality Weekly Report from the CDC. Today in the United States, most cases come from consuming contaminated imported unpasteurized milk products. There were 125 human cases reported to the CDC in 2002. (Graph from the Summary of Notifiable Diseases 2006, CDC website.)

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Brucellosis

- United States
  - Approximately 100 cases per year
  - Less than 0.5 cases/100,000 people
  - Mostly California, Florida, Texas, Virginia
  - Many cases associated with consumption of foreign cheeses

Incidence in the United States is less 0.5 cases per 100,000 people. Most cases are reported from California, Florida, Texas, and Virginia. There have been about 100 cases reported each year for the last 10 years .

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Disease in Humans

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Human Disease

- Can affect any organ or organ system
- All patients have a cyclical fever
- Variability in clinical signs
  - Headache, weakness, arthralgia, depression, weight loss, fatigue, liver dysfunction

Brucellosis can involve any organ or organ system, and have a very insidious onset with varying clinical signs. The one common sign in all patients is an intermittent/irregular fever of variable duration, thus the term *undulant fever*. The acute form (<8 weeks from illness onset) is characterized by symptomatic, nonspecific, or flu-like symptoms, including fever, malaise, anorexia, headache, myalgia, and back pain. Drenching sweats can occur, particularly at night. Splenomegaly, hepatomegaly, coughing, and pleuritic chest pain are sometimes seen. Gastrointestinal signs, including anorexia, nausea, vomiting, diarrhea, and constipation, occur frequently in adults but less often in children. In many patients, the symptoms last for two to four weeks and are followed by spontaneous recovery. Others develop an intermittent fever and other persistent symptoms that typically wax and wane at 2- 14 day intervals. Most people with this undulant form recover completely in three to 12 months. A few patients become chronically ill. Relapses can occur months after the initial symptoms, even in successfully treated cases.

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Human Disease

- 20-60% of cases
  - Osteoarticular complications
    - Arthritis, spondylitis, osteomyelitis
- Hepatomegaly may occur
- Gastrointestinal complications
- 2-20% of cases
  - Genitourinary involvement
    - Orchitis and epididymitis most common

Complications are seen occasionally, particularly in the undulant and chronic forms. The most frequent complication of brucellosis appears to be involvement of the bones and joints. The sacroiliac joint is often affected in countries where *B. melitensis* predominates. Arthritis of the hips, knees, and ankles also occurs with spondylitis being the primary complaint. Complications involving the bones and joints are reported in 20-60% of patients with clinical brucellosis. The liver is generally involved in most cases of brucellosis as it is an organ involved in the reticulo-endothelial system (RES), much like the skeletal system. It is involved in trying to remove the infection from the body and as a result hepatomegaly may occur. It is possible to isolate organisms from the liver without signs of hepatic dysfunction or signs of liver disease. When foodborne exposure occurs, the gastrointestinal complications are no worse than the systemic signs of any other route of infection, including nausea, vomiting, anorexia, weight loss, and abdominal discomfort. Genitourinary complications can occur with the testicles being most frequently involved. Evidence of orchitis, or epididymo-orchitis can occur in association with systemic infection. Renal involvement is rare. To date there is no convincing evidence that pregnant women with brucellosis have more abortions than do those with other bacteremic infections if the disease is identified and treated promptly. While venereal transmission has not been proven, brucella organisms have been identified in banked human sperm.

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Human Disease

- Neurological
  - Depression, mental fatigue
- Cardiovascular
  - Endocarditis resulting in death
- Chronic brucellosis is hard to define
  - Length, type and response to treatment variable
  - Localized infection
- Blood donations of infected persons should not be accepted

Neurological complications, such as depression and mental fatigue, are common complaints (up to 5% of cases), but invasion of the CNS occurs in <2% of cases. Cerebral spinal fluid often contains antibodies to *Brucella*, but it is rare to isolate the organism. Cardiovascular complications are rare, but endocarditis accounts for the majority of deaths from this disease. Often antibiotics and replacement of the affected valves are essential in the course of treatment depending on the timing of the diagnosis from initial exposure. To date, there is no uniform definition of chronic brucellosis due to the variability of organ systems affected and response to, length of, or type of, treatment. A suppurative, localized infection is often refractory to antibiotic treatment and may require surgical intervention before a cure can be achieved.

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Human Disease

- Congenitally infected infants
  - Low birth weight
  - Failure to thrive
  - Jaundice
  - Hepatomegaly
  - Splenomegaly
  - Respiratory difficulty
  - General signs of sepsis (fever, vomiting)
  - Asymptomatic

The symptoms of congenital brucellosis are variable. Some congenitally infected infants are delivered prematurely, while others are born at full term. Common symptoms include low birth weight, fever, failure to thrive, jaundice, hepatomegaly, and splenomegaly. Some newborns with congenital brucellosis have respiratory difficulty or severe respiratory distress, hypotension, vomiting, and other signs of sepsis. Other infants may be asymptomatic or have only mild symptoms at birth. Whether brucellosis can lead to spontaneous abortion in humans is controversial.

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Diagnosis in Humans

- Isolation of organism
  - Blood, bone marrow, other tissues
- Serum agglutination test
  - Four-fold or greater rise in titer
  - Samples 2 weeks apart
- Immunofluorescence
  - Organism in clinical specimens
- PCR

Microscopic examination of stained smears can be useful for a presumptive diagnosis, particularly if the direct examination is supported by other tests. They are not truly acid-fast; however, they are resistant to decolorization by weak acids, and stain red against a blue background with the Stamp's modification of the Ziehl-Neelsen method. Other organisms, such as *Coxiella burnetii*, can resemble *Brucella*. In humans, the definitive diagnosis is by culture or serology. *Brucella* species can sometimes be isolated from the blood early in the infection; bone marrow is often positive at this stage. Occasionally, bacteria can be recovered from the cerebrospinal fluid, urine, or tissues. Most *Brucella* species form colonies within a few days, but isolates from seals grow slowly and may take 7-10 days to become visible on selective media. *Brucella* isolates can be identified to the species and biovar level by phage typing and cultural, biochemical, and serological characteristics. Most human infections are diagnosed by serology. Tests used include serum agglutination, a modified Coombs' (antiglobulin) technique, ELISAs, and immunoblotting (Western blotting). Serologic diagnosis is complicated by previous exposures and other factors; a definitive diagnosis usually requires a fourfold rise in titer. Immunostaining can sometimes demonstrate the presence of *Brucella* spp. in a clinical specimen. PCR techniques can also be used for diagnosis. PCR has begun to gain popularity in the diagnosis of brucellosis due to the high specificity and sensitivity of the test, as well as the quick turn around of results. Chronic brucellosis can be extremely difficult to diagnose if the serologic results are equivocal and the organism cannot be cultured.

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Treatment of Choice

- Combination therapy has the best efficacy
  - Doxycycline for six weeks in combination with streptomycin for 2-3 weeks or rifampin for 6 weeks
- CNS cases treat 6-9 months
  - Same for endocarditis cases plus surgical replacement of valves

Prolonged treatment with clinically effective antibiotics are necessary to penetrate these facultative, intracellular pathogens. Combination therapy has shown the best efficacy for treatment in adults and examples of regimens follow. Doxycycline orally for six weeks, in addition to streptomycin for two to three weeks has been efficacious. Doxycycline in combination with rifampin for six weeks has also been used with success. CNS cases involve extensive long term treatment. Patients developing endocarditis also require long term treatment along with replacement of damaged valves.

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Prognosis

- May last days, months, or years
- Recovery is common
- Disability is often pronounced
- About 5% of treated cases relapse
  - Failure to complete the treatment regimen
  - Sequestered infection requiring surgical drainage
- Case-fatality rate: <2% (untreated)
  - Endocarditis caused by *B. melitensis*

Brucellosis is rarely fatal if treated; in untreated persons, estimates of the case fatality rate vary from less than 2% to 5%. Deaths are usually caused by endocarditis or meningitis. Although recovery is common, disability is often pronounced depending on the localization of infection and response to treatment. Approximately 5% of treated cases will relapse weeks to months after therapy has ended due to the failure to complete the treatment regimen or infection that requires surgical drainage. Antibiotic resistant strains of *Brucella* have been reported, but the clinical importance of that fact is not well understood.

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Animals and Brucellosis

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Clinical Signs: Cattle & Bison

- Third trimester abortions with *B. abortus*
- Retained placenta
  - Once expelled will have a leathery appearance
- Endometritis
- Birth of dead or weak calves
  - Respiratory distress and lung infections
- Low milk yield

Brucellosis causes abortions in the third trimester of pregnancy when unvaccinated cattle are exposed to the infectious organism. It is stated that greater than 80% of cattle will abort if exposed during this critical time of gestation. The organisms enter through the mucous membranes and can cause inflammation of the placenta. Abortion can occur within 2 weeks or up to 5 months following infection. The overall appearance of the placenta is a leathery look. The intercotyledonary area is thickened and has a wet appearance. The fetus may look normal if aborted acutely after infection, or autolytic if not expelled for a period of time. The pregnancy may end with a stillborn or weak calf. Often, retained placentas and decreased milk yield follow. Once a cow has aborted from infection, subsequent gestations are normal, after a period of temporary sterility. Only 5% have residual sterility. Most cows will shed the organisms in the milk and uterine discharges for life following infection. Infections in nonpregnant females are usually asymptomatic.

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Clinical Signs: Sheep & Goats

- *B. melitensis* causes late term abortions
  - Retained placenta
  - Birth of dead or weak lambs/kids
- Goats - articular and periarticular hygroma localizations
- *B. ovis* causes abortions, fertility problems
  - Orchitis, epididymitis
  - Abnormal breeding soundness exam
  - Organisms present in semen

The primary causal agent of abortion in sheep and goats is *B. melitensis*, which has similar signs to *B. abortus* in cattle. Abortion generally occurs late term or results in stillbirths or weak lambs/kids. The organisms enter through the mucous membranes and can cause inflammation and retention of the placenta. Sheep and goats usually abort only once, but reinvasion of the uterus and shedding of organisms can occur during subsequent pregnancies. Acute orchitis and epididymitis can occur in males, and may result in infertility. Arthritis is seen occasionally in both sexes. Many non-pregnant sheep and goats remain asymptomatic. *B. ovis* affects sheep but not goats. It can cause abortions, placentitis, and neonatal death. The most important clinical signs are epididymitis and orchitis resulting in fertility problems. It is venereally transmitted, and shedding of the organism can be greater than four years in rams. Semen quality deteriorates rapidly and often inflammatory cells are present. Epididymal enlargement can occur unilaterally or bilaterally and the tunics become thickened and develop adhesions. Fibrous atrophy of the testes is permanent. If no outward clinical signs are palpable, semen must then be repeatedly cultured to catch intermittent shedders. Abortions, placentitis and perinatal mortality can be seen in ewes but are uncommon. Systemic signs are rare.

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Clinical Signs: Swine

- *B. suis*
- Prolonged bacteremia
- Abortion, early or late gestation
- Fertility problems
  - Sows temporary
  - Boars, unilateral or bilateral orchitis
- Lameness, posterior paralysis, spondylitis, metritis, abscesses

Pigs are primarily affected with *B. suis* which causes a long lasting bacteremia. Localization of the infection can vary, and thus, so do the clinical signs. Abortion can occur up to 80% of the time; when abortions occur early in gestation, infected animals often go undetected and are rebred. Temporary or permanent sterility is common and is sometimes the only sign. Boars can have unilateral or bilateral orchitis affecting their fertility. Other signs include lameness, posterior paralysis, spondylitis, metritis, and abscess formation in various locations of the body.

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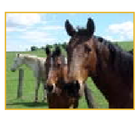
Clinical Signs: Horses

- *B. abortus* most common
  - Susceptible to *B. suis*
- Fistulous Withers or Poll Evil
  - Inflammation of the supraspinous bursa
  - Exudative process
    - Fills with clear viscous liquid
    - Can eventually rupture



Horses are susceptible to *B. abortus* or *B. suis* from infectious or traumatic origin. Clinically, these animals have an inflammation in the supraspinous bursa or supra-atlantal bursa; this is referred to as Fistulous Withers or Poll Evil, respectively. The bursal sac becomes distended by a clear, viscous, straw-colored exudate and develops a thickened wall. It can rupture, leading to secondary inflammation. In chronic cases, nearby ligaments and the dorsal vertebral spines may become necrotic. *Brucella*-associated abortions are rare in horses.

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Clinical Signs: Dogs

- Susceptible to
  - *B. melitensis, B. abortus,* and *B. suis*
- *B. canis* causes abortions
  - Last trimester of pregnancy
  - Prolonged vaginal discharge
  - Bacteremia
  - Failure to conceive, stillbirths, prostatitis, epididymitis

Dogs are susceptible to infections with *B. melitensis*, *B. abortus*, and *B. suis*, but the major cause of abortion in this species is *B. canis*. Generally, dogs will abort in the last trimester of pregnancy (seventh to ninth week of gestation) and have prolonged vaginal discharge. Bacteremia often occurs up to eighteen months post-exposure. Other clinical signs include stillbirths, failure to conceive/early embryonic death, lymphadenitis, epididymitis, periorchitis, and prostatitis.

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Clinical Signs: Wildlife

- Elk
  - Abortions, no retained placenta
- Moose
  - Debilitated, death
- Predators not clinical, but are vectors
  - Coyotes, crows, vultures, bears
    - Aid in disease spread by carrying infected tissues away from abortion site

Elk generally lose their first pregnancy after becoming infected with *B. abortus*, but do not have problems with retained placenta or infertility as cattle do. Elk infection has been documented in Alaska, Canada, and mainland United States. It is thought that moose are very susceptible to infection with *Brucella abortus*, unlike other wildlife. Seropositive free-ranging moose have not been found in North America, and it is often thought this is due to the rapid death that follows once they become infected. In caribou and reindeer, *B. suis* biovar 4 can cause abortion and retained placenta. Metritis and mastitis can also occur. Males may develop orchitis. Lameness can occur in both sexes from arthritis, bursitis, tenosynovitis, and/or hygromas. Subcutaneous abscesses are also seen. In hares, *B. suis* biovar 2 infection is characterized by nodules in the internal organs, particularly the reproductive organs, as well as the subcutaneous tissues and muscles. The nodules can become purulent. The animal's body condition may be minimally affected. *B. ovis* can also cause poor semen quality in red deer stags, but abortions have not been reported in hinds (females).

Predators act as vectors to spread the disease, but are resistant to infection. They often come upon an abortion site and drag infected tissues and fluids to other areas, contaminating a larger area for susceptible species to become infected.

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Diagnosis in Animals

- Isolation of organism
  - Blood, semen, other tissues
- Serology
  - Brucellosis card test, ELISA
- Brucella milk ring test
- Demonstration by fluorescent antibody of organism in clinical specimen
  - Placenta, fetus

Brucellosis can be diagnosed by culture, serology, or other tests. Microscopic examination of smears stained with the Stamp's modification of the Ziehl-Neelsen method can be useful for a presumptive diagnosis. Organisms may be found in abortion products, vaginal discharges, milk, semen, or various tissues. This test is not definitive. Other organisms, such as *Chlamydophila abortus* and *Coxiella burnetii,* can resemble *Brucella*. Direct examination may not detect the small numbers of organisms present in milk and dairy products. Blood cultures are often used to detect *B. canis* in dogs. In this species, bacteremia (which may be intermittent) can persist for up to five years and possibly longer. *B. canis* requires a specific test as it does not have a smooth lipopolysaccharide cell wall. Genetic techniques can also be used for biotyping. The vaccine strains (*B. abortus* strains S19 and RB51, and *B. melitensis* Rev 1) can be distinguished from field strains by their growth characteristics and sensitivity to antibiotics and other additives. Brucellosis is often diagnosed by serology. Serological tests are not completely specific and cannot always distinguish reactions due to *B. melitensis* from cross-reactions to other bacteria, particularly *Yersinia enterocolitica* O:9. Serology is commonly used at slaughter plants and when marketing cattle. The Brucella milk ring test is used on pooled milk samples and fluorescent antibody of the organism in the placenta and fetus for abortion cases.

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Treatment of Animals

- Combination antibiotic therapy has the best efficacy
- Surgical drainage plus antibiotics
- Often expensive
- High rate of failure
- Indemnity program from government

There is no practical treatment for infected cattle or pigs, but long-term antibiotic treatment is sometimes successful in infected dogs. Prolonged treatment with clinically effective antibiotics are necessary to penetrate these facultative, intracellular pathogens. Combination therapy has shown the best efficacy for treatment, but due to the expense incurred and the high rate of failure, it often is not practical. Some dogs relapse after treatment. Surgical drainage, if appropriate, along with antibiotics may be of some use. Fertility may remain low even if the organism is eliminated. With the indemnity program, owners often opt for depopulation instead of treatment.

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Prognosis

- Disease may last days, months, or years
- Eradication program in the United States often leads to slaughter of certain species
  - Cattle, bison, horses, sheep, goats, swine

The disease may last days to years depending on the species and type of infection. In the United States, animals are often serologically tested and carriers of brucellosis are eliminated. So prognosis for certain animal species, such as cattle, horses, bison, swine, sheep, and goats, is not good and may result in termination of life.

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Yellowstone National Park

B. abortus was first detected in bison in 1917 in Yellowstone National Park.

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Bison in Yellowstone

- Goal = Brucellosis free by 2010
- Can leave the park to winter feed in Wyoming
- Up to 50% sero-positive
- Congregate at calving

Up until 1998, the Park Service was limiting the number of bison that were allowed to leave the park by hazing them back in or shooting them. Their goal was to make the Park brucellosis free by 2010. Today, bison and elk are allowed to leave the park in winter to eat at feeding grounds in Wyoming. Here researchers have been able to test the animals, and it was discovered that up to 50% of bison test seropositive for *B. abortus*. This indicates exposure, not necessarily infection. This has allowed *B. abortus* to become established in the elk population. Research is ongoing to look at the efficacy of immunizing bison and elk orally with the RB51 vaccine. Bison seem more likely to abort or shed organisms than cattle after vaccination with same dose. Bison tend to congregate more at calving which aids in the spread of disease to their herdmates through infected tissues and fluids. Brucellosis is not a major factor in herd survival for elk or bison – winter is.

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Elk in Yellowstone

- Exposed to *B. abortus* via winter feeding grounds
- Isolate themselves at calving
  - Clean the area
  - Remain separate from herd for a few days
- Less disease transmission between herdmates

Elk are thought to be exposed to infected bison via the winter feeding grounds in western Wyoming where approximately 37% of them tested seropositive. In contrast, only 1-2% tested seropositive in non-feeding grounds. In the wild, elk tend to isolate themselves around the time of calving and separate from the herd for a few days after giving birth. Being meticulous creatures, elk consume the placental tissues and fluids to try and avoid attracting predators to the area. These factors lead to less disease transmission between herdmates.

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Prevention and Control

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Prevention and Control

- Education about risk of transmission
  - Farmer, veterinarian, abattoir worker, butcher, consumer, hunter, public
- Wear proper attire if dealing with infected animals/ tissues
  - Gloves, masks, goggles
- Avoid consumption of raw dairy products

Educate those at greatest risk about the routes of transmission of brucellosis. Properly protecting yourself, if you are an “at risk” individual, by wearing gloves, masks, goggles, and coveralls to prevent exposure to tissues and body secretions of infected animals can help. Pasteurization or boiling milk and avoidance of eating unpasteurized dairy products will help decrease human exposure to brucellosis.

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Prevention and Control

- Immunize in areas of high prevalence
  - Young goats and sheep with Rev-1
  - Calves with RB51
  - No human vaccine
- Eradicate reservoir
  - Identify, segregate, and/or cull infected animals

Vaccinating calves at 4 to 12 months of age with RB51 for *B. abortus* and goats and sheep with Rev-1 for *B. melitensis* has helped eliminate infection in these animals, thus decreasing possible exposure to humans. At this time, RB51 is being tested for efficacy in bison. More research needs to be done before it is approved, but preliminary results look promising. Elimination of livestock reservoirs is the most effective means to reduce cases of disease in people, so strict adherence to federal laws for identifying, segregating and/or culling infected animals is essential to success.

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Prevention and Control

- *B. suis, B. ovis,* and *B. canis*
  - Venereal transmission
  - Separate females at birthing to reduce transmission on the farm or in kennel

When it comes to the venereal transmitted organisms, *B. suis*, *B. ovis*, and *B. canis*, it is important to separate females during birthing to reduce exposure and transmission to susceptible animals on the farm or in the kennels.

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RB51

- Approved for use February 1996 for calves
- Able to differentiate “wild type” exposure from immunization
  - Lacks LPS-O antigen that causes antibody response on serologic or milk tests
- Infectious to humans
  - Serologically negative upon testing post-exposure
  - CDC registry of human exposures
  - 32 documented exposures as of 1998

RB51 was approved for use by APHIS in February 1996, and for use in the eradication program in March. It was the first new vaccine for brucellosis in 50 years, and it is given only to calves 4-12 months old. This attenuated strain is less virulent in cattle so they will shed fewer organisms if they become infected. The biggest advantage with RB51 is the ability to protect via vaccination, but detect those infected with the wild type virus. This vaccine lacks the surface antigen, LPS-O, that induces an antibody response and therefore is able to be used as a surveillance tool in serodiagnostic tests, such as tube agglutination tests, the “card test”, milk ring test, and complement fixation. Vaccination of cattle with RB51 does not induce antibody responses that cause positive reactions on diagnostic tests now used. This becomes a problem when humans are accidentally inoculated through a needle stick. The advantage of testing seronegative in cattle after immunization makes diagnosis in humans post-exposure difficult because they too test seronegative. RB51 is considered infectious for humans and only federally accredited veterinarians should administer the vaccine. The CDC keeps a registry of all human exposures, and they can be notified at (404) 639-3158. As of 1998 there have been 32 documented exposures. Of those, three reported inflammation at the inoculation site, and one had intermittent fever, chills, headache, and myalgia.

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U.S. Eradication Program

- U.S. Department of Agriculture
  - 1934: Cooperative State-Federal Brucellosis Eradication Program
    - Removal of diseased cattle due to drought
- 1951: APHIS became involved
- 1957: 124,000 positive herds
- Approach
  - Test, slaughter, trace back, investigate, and vaccinate

The United States Department of Agriculture started the Cooperative State-Federal Brucellosis Eradication Program in 1934. Increasing public health concern and drought conditions made it necessary to reduce cattle herds, so the diseased were the first eliminated. The Program also implemented testing, quarantine, and elimination standards that are still followed today. In 1951, the National Brucellosis Program was initiated by the Animal and Plant Health Inspection Service and made it mandatory that all states comply. In 1957, there were more than 124,000 cattle herds known to be infected. The approach was to test herds and remove positives, depopulate if necessary, vaccinate new animals, and trace back reactors through the market identification program to the herds of origin. They have successfully reduced the number of herds from 124,000 in 1957 to 2 as of December 2003.

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U.S. Eradication Program

- Target date for eradication was December 31, 1998
- Surveillance
  - Brucellosis ring test
    - Pooled milk
  - Market Cattle Identification
    - Blood test, individual
- Indemnity for whole herd depopulation
  - $250 nonregistered cattle/bison
  - $750 or 95% of value minus salvage value for registered cattle

The goal was to eradicate brucellosis by December 31, 1998. At that time there were four cattle herds and one bison herd affected, but all had at least one negative test. There are two primary surveillance procedures to locate infection without having to test each animal in every herd. Milk from dairy herds is checked two to four times a year by testing a small sample obtained from creameries or farm milk tanks for evidence of brucellosis, also known as the brucellosis ring test. Bison herds and cattle herds that do not produce milk for sale are routinely checked for brucellosis by blood-testing animals sold from these herds at livestock markets or at slaughter. The blood agglutination test is used to pinpoint infection within a herd. Should a herd test positive, it must then be depopulated. Financial compensation to the producer varies by offering a fixed rate, which is $250 per animal for cattle or bison that are not registered, or $750 per head for registered cattle, minus their salvage value. The Appraisal option has been introduced based on fair market value for registered cattle, and producers are then offered 95% of that value, again, minus the salvage value.

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U.S. Eradication Program

- Fiscal Year 2001
  - 4.7 million calves vaccinated
  - 9.9 million cattle tested under the Market Cattle Identification program
  - 3 brucellosis herds depopulated
    - Indemnity paid = $211,153
    - An additional $47,700 for purchase of animals or diagnostic purposes

The number of cattle tested for brucellosis in Fiscal Year 2001 under the Market Cattle Identification test was 9.9 million, 4.2% more than fiscal year 2000. There were 4.7 million calves vaccinated for brucellosis in fiscal year 2001 compared to 4.4 million in fiscal year 2000. Only 3 brucellosis affected herds were depopulated in the United States in fiscal year 2001, at a cost of $211,153 in indemnity. An additional $47,700 was spent to purchase animals that were traced out of affected herds or for diagnostic purposes.

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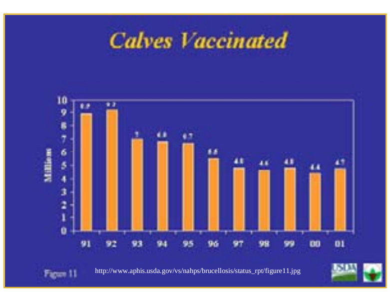

This is a graph depicting the number of calves vaccinated in millions from 1991 to 2001.

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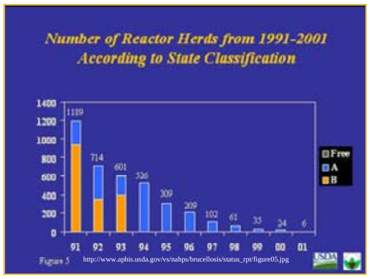

This graph depicts the number of reactor herds based on state classification from 1991 through 2001.

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Brucellosis Classes

- Free
  - Feb 1, 2008 – U.S. class-free in cattle
- A: No more than 0.25% infection rate and cattle must be tested before export
- B: Infection rate of no more than 1.5% and must be tested before interstate movement

The United States does not have any B or C class states.

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***B. abortus* Exposure**

- 1997: Kansas State University
  - 14 month old heifer admitted to hospital with calving complications
    - Vaccinated with RB51 at 8 months
    - 10 times the dose for known pregnant cattle
  - 9 humans exposed
  - Treated with doxycycline
    - No clinical signs

In May 1997, a fourteen month old heifer was admitted to the veterinary school in Manhattan, Kansas for complications involving parturition. She had been vaccinated at eight months of age with RB51, but it was unknown at that time that she was pregnant. This dose, while adequate for the heife,r was ten times the dosage recommended for pregnant or adult animals. After failure of a vaginal delivery, a cesarean section was performed, later followed by necropsy on the stillborn calf. Due to complications, the delivering heifer was also euthanized. In all, nine people became exposed to *Brucella abortus*, 1 farmer, 4 veterinary students, and 4 veterinary clinicians. Within one week after exposure, eight of the exposed started on a prophylactic regimen of doxycycline, and three also received rifampin. All were followed for six months, during which time no one showed clinical signs.

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***Brucella* as a Biological Weapon**

- Aerosolized *B. melitensis*
  - City of 100,000 people
  - Inhale 1,000 cells (2% decay per min)
  - Case-fatality rate of 0.5%
  - 50% hospitalized for 7 days
    - Outpatients required 14 visits
    - 5% relapsed
- Results
  - 82,500 cases requiring extended therapy
  - 413 deaths
  - $477.7 million economic impact

A bioterrorism scenario has been evaluated using an aerosolized *Brucella melitensis* agent spread along a line with the prevailing winds with optimal meteorologic conditions. It assumed that the infectious dose to infect 50 (ID_{50}) percent of the population would require inhalation of 1,000 vegetative cells. The decay of the organism is estimated to be at 2% per minute without affecting viability and virulence. The scenario also assumed, depending on the persons closeness to the point of origin, one would inhale one to ten ID_{50}. The case fatality rate was estimated to be 0.5% with 50% of the people being hospitalized and staying on average, seven days. If not hospitalized, they often made fourteen outpatient visits and received oral doxycycline for 42 days, and parenteral gentamicin for 7 days. Relapses occurred at 5% and required 14 outpatient visits in one year. In looking at the economic impact of such a threat, one must consider the cost of premature human death, and all the costs related to hospitalization and outpatient visits. The minimum cost of exposure would be around $477.7 million per 100,000 persons exposed.

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Internet Resources

- USDA APHIS VS Brucellosis Disease Information
  - www.aphis.usda.gov/animal_health/animal_diseases/brucellosis/
- WHO Fact Sheet Brucellosis
  - www.who.int/mediacentre/factsheets/en/
- Center for Food Security and Public Health
  - www.cfsph.iastate.edu
- BruNet Publication
  - www.moag.gov.il/brunet/public.htm

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Acknowledgments

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