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suffering from contagious abortion.<sup>6</sup> In 1917, A. C. Evans recognized that Bang's organism was identical to that described by Bruce as the causative agent of human brucellosis. The organism infects mainly cattle, sheep, goats, and other ruminants, in which it causes abortion, fetal death, and genital infection.<sup>7,8</sup> Humans, who are usually infected incidentally by contact with infected animals or ingestion of dairy foods, may develop numerous symptoms in addition to the usual ones of fever, malaise, and muscle pain. Disease frequently becomes chronic and may relapse, even with treatment.

The ease of transmission by aerosol suggests that *B*□ organisms might be a candidate for use as a biological warfare agent. Indeed, the United States began development of *B*□ as a biological weapon in 1942. The agent was formulated to maintain long-term viability, placed into bombs, and tested in field trials during 1944–1945 using animal targets. By 1967, the United States terminated its offensive program for development and deployment of *B*□ as a biological weapon. Although the munitions developed were never used in combat, the studies reinforced the concern that *B*□ organisms might be used against U.S. troops as a biological warfare agent.<sup>9</sup>

Brucellae are small, nonmotile, nonsporulating, nontoxigenic, nonfermenting, aerobic, Gram-negative coccobacilli that may, based on DNA homology, represent a single species.<sup>10</sup> Conventionally, however, they are classified into six species, each comprising several biovars. Each species has a characteristic, but not an absolute, predilection to infect certain animal species (Table 25-1). Only *B*□, *B*□, *B*□, and *B*□ cause disease in man. Infection of humans with *B*□ and *B*□ has not been described.

Brucellae grow best on trypticase, soy-based, or other enriched media with a typical doubling time of 2 hours. Most biovars of *B*□, *D* 9 97DJF 2D7 . E , 9(JF 2D). J

thionine or basic fuchsin dyes; agglutination by antisera directed against certain lipopolysaccharide epitopes; and by susceptibility to lysis by bacte-

riophage. Recently, analysis of fragment lengths of deoxyribonucleic acid (DNA) cut by various restriction enzymes has also been used to differentiate brucellae groupings.<sup>10</sup>

The lipopolysaccharide (LPS) component of the outer cell membranes of brucellae is quite different—both structurally and functionally—from that of other Gram-negative organisms.<sup>11,12</sup> The lipid A portion of a B $\square$  organism LPS contains fatty acids 16 carbons long, and lacks the 14-carbon myristic acid typical of lipid A of Enterobacteriaceae. This unique structural feature may underlie

the remarkably reduced pyrogenicity (less than 1/100th) of B $\square$  LPS, compared with the pyrogenicity of *E. coli* LPS.<sup>13</sup> In addition, the O-polysaccharide portion of LPS from smooth organisms contains an unusual sugar, 4,6-dideoxy-4-formamido- $\alpha$ -D-mannopyranoside, which is expressed either as a homopolymer of  $\alpha$ -1,2-linked sugars (A type), or as 3  $\alpha$ -1,2 and 2  $\alpha$ -1,3-linked sugars (M type). These variations in O-polysaccharide linkages lead to specific, taxonomically useful differences in immunoreactivity between A and M sugar types.<sup>14</sup>

Animals may transmit B $\square$  organisms during septic abortion, at the time of slaughter, and in their milk. Brucellosis is rarely, if ever, transmitted from person to person. The incidence of human disease is thus closely tied to the prevalence of infection in sheep, goats, and cattle, and to practices that allow exposure of humans to potentially infected animals or their products. In the United States, where most states are free of infected animals and where dairy products are routinely pasteurized, illness occurs primarily in individuals such as veterinarians, shepherds, cattlemen, and slaughterhouse workers who have occupational exposure to infected animals. In many other countries, humans more commonly acquire infection by ingestion of unpasteurized dairy products, especially cheese.

Less obvious exposures can also lead to infection. In Kuwait, for example, disease with a relatively

A C B □ □

in lymph nodes, liver, spleen, mammary gland, joints, kidneys, and bone marrow.

In macrophages, brucellae may inhibit fusion of phagosomes and lysosomes, and replicate in the phagosome.<sup>23</sup> If unchecked by macrophage microbicidal mechanisms, the bacteria destroy their host cells and infect additional cells. Brucellae can also replicate extracellularly in host tissues. Histopathologically, the host cellular response may range from abscess formation to lymphocytic infiltration to granuloma formation with caseous necrosis.

Studies in experimental models have provided important insights into host defenses that eventually control infection with *B*□ organisms. Se-



A C B □ □

ganism does not have O-polysaccharide on its surface. Immunoenzymatic assays (eg, enzyme-linked immunosorbent assays [ELISAs]) have been developed for use with *B. melitensis*, but are not well standardized. ELISAs developed for other brucellae similarly suffer from lack of standardization.

In addition to serologic testing, diagnosis should

axial skeleton are favored targets; arthritis appears in approximately one third of patients. Fatalities occur rarely, usually in association with central nervous system or endocardial infection.

Serologic diagnosis uses an agglutination test that detects antibodies to lipopolysaccharide. This test, however, is not useful to diagnose infection caused by *B. melitensis*, a naturally O-polysaccharide deficient strain. Infection can be most reliably con-

firmed by culture of blood, bone marrow, or other infected body fluids, but the sensitivity of culture varies widely.

Nearly all patients respond to a 6-week course of oral therapy with a combination of rifampin and doxycycline; fewer than 10% of patients relapse. Six weeks of doxycycline with addition of streptomycin for the first 3 weeks is also effective therapy. No vaccine is available for humans.

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