- **Section VIII-D: Rickettsial Agents**

Agent: Coxiella burnetii

Coxiella burnetii is the etiologic agent of Q fever. *C. burnetii* is a bacterial obligate intracellular pathogen that undergoes its developmental cycle within an acidic vacuolar compartment exhibiting many characteristics of a phagolysosome. The developmental cycle consists of a large (approximately 1 µm in length) cell variant that is believed to be the more metabolically active, replicative cell type and a smaller, more structurally stable cell variant that is highly infectious and quite resistant to drying and environmental conditions.[1-4] The organism undergoes a virulent (Phase I) to avirulent (Phase II) transition upon serial laboratory passage in eggs or tissue culture.

The infectious dose of virulent Phase I organisms in laboratory animals has been calculated to be as small as a single organism.[5] The estimated human infectious dose for Q fever by inhalation is approximately 10 organisms.[6] Typically, the disease manifests with flu-like symptoms including fever, headache, and myalgia but can also cause pneumonia and hepatomegaly. Infections range from sub-clinical to severe although primary infections respond readily to antibiotic treatment. Although rare, *C. burnetii* is known to cause chronic infections such as endocarditis or granulomatous hepatitis.[7]

OCCUPATIONAL INFECTIONS

Q fever is the second most commonly reported LAI in Pike's compilation. Outbreaks involving 15 or more persons were recorded in several institutions.[8,9] Infectious aerosols are the most likely route of laboratory-acquired infections. Experimentally infected animals also may serve as potential sources of infection for laboratory and animal care personnel. Exposure to naturally infected, often asymptomatic, sheep and their birth products is a documented hazard to personnel.[10,11]

NATURAL MODES OF INFECTION

Q fever (Q for query) occurs worldwide. Broad ranges of domestic and wild mammals are natural hosts for Q fever and sources of human infection. Parturient animals and their birth products are common sources of infection. The placenta of infected sheep may contain as many as 10^9 organisms per gram of tissue[12] and milk may contain 10^5 organisms per gram. The resistance of the organism to drying and its low infectious dose can lead to dispersal from contaminated sites.

LABORATORY SAFETY

The necessity of using embryonated eggs or cell culture techniques for the propagation of *C. burnetii* leads to extensive purification procedures. Exposure to infectious aerosols and parenteral inoculation cause most infections in laboratory and animal care personnel.[8,9] The agent may be present in infected arthropods and in the blood, urine, feces, milk, and

tissues of infected animals or human hosts. Exposure to naturally infected, often asymptomatic, sheep and their birth products is a documented hazard to personnel.[10,11] Recommended precautions for facilities using sheep as experimental animals are described elsewhere.[10,13]

Containment Recommendations

BSL-2 practices and facilities are recommended for nonpropagative laboratory procedures, including serological examinations and staining of impression smears. BSL-3 practices and facilities are recommended for activities involving the inoculation, incubation, and harvesting of embryonated eggs or cell cultures, the necropsy of infected animals and the manipulation of infected tissues. Experimentally infected animals should be maintained under ABSL-3 because infected rodents may shed the organisms in urine or feces.[8] A specific plaque-purified clonal isolate of an avirulent (Phase II) strain (Nine Mile) may be safely handled under BSL-2 conditions.[14]

SPECIAL ISSUES

Vaccines An investigational Phase I, Q fever vaccine (IND) is available on a limited basis from the Special Immunizations Program (301-619-4653) of the USAMRIID, Fort Detrick, Maryland, for at-risk personnel under a cooperative agreement with the individual's requesting institution. The use of this vaccine should be restricted to those who are at high risk of exposure and who have no demonstrated sensitivity to Q fever antigen. The vaccine can be reactogenic in those with prior immunity, thus requires skin testing before administration. The vaccine is only administered at USAMRIID and requires enrollment in their Q fever IND Immunization Program. For at-risk laboratory workers to participate in this program, fees are applicable. Individuals with valvular heart disease should not work with *C. burnetii* (See Section 7).

Select Agent *C. burnetii* is a Select Agent requiring registration with CDC and/or USDA for possession, use, storage and/or transfer. See Appendix F for additional information.

Transfer of Agent Importation of this agent may require CDC and/or USDA importation permits. Domestic transport of this agent may require a permit from USDA/APHIS/VS. A DoC permit may be required for the export of this agent to another country. See Appendix C for additional information.

Agents: Rickettsia prowazekii, Rickettsia typhi (R. mooseri), Orientia (Rickettsia) tsutsugamushi and Spotted Fever Group agents of human disease; Rickettsia rickettsii, Rickettsia conorii, Rickettsia akari, Rickettsia australis, Rickettsia siberica, and Rickettsia japonicum

Rickettsia prowazekii, Rickettsia typhi (R. mooseri), Orientia (Rickettsia) tsutsugamushi and the Spotted Fever Group agents of human disease (*Rickettsia rickettsii, Rickettsia*

conorii, Rickettsia akari, Rickettsia australis, Rickettsia siberica, and *Rickettsia japonicum)* are the etiologic agents of epidemic typhus, endemic (murine) typhus), scrub typhus, Rocky Mountain spotted fever, Mediterranean spotted fever, rickettsialpox, Queensland tick typhus, and North Asian spotted fever, respectively.

Rickettsia spp. are bacterial obligate intracellular pathogens that are transmitted by arthropod vectors and replicate within the cytoplasm of eukaryotic host cells. Two groups are recognized within the genus, the typhus group and the spotted fever group. The more distantly related scrub typhus group is now considered a distinct genus, *Orientia.* Rickettsiae are primarily associated with arthropod vectors in which they may exist as endosymbionts that infect mammals, including humans, through the bite of infected ticks, lice, or fleas.[15]

OCCUPATIONAL INFECTIONS

Pike reported 57 cases of laboratory-associated typhus (type not specified), 56 cases of epidemic typhus with three deaths, and 68 cases of murine typhus.[8] Three cases of murine typhus have been reported from a research facility.[16] Two were associated with handling of infectious materials on the open bench; the third case resulted from an accidental parenteral inoculation. These three cases represented an attack rate of 20% in personnel working with infectious materials. Rocky Mountain spotted fever is a documented hazard to laboratory personnel. Pike reported 63 laboratory-associated cases, 11 of which were fatal.8 Oster reported nine cases occurring over a six year period in one laboratory. All were believed to have been acquired as a result of exposure to infectious aerosols.[17]

NATURAL MODES OF INFECTION

The epidemiology of rickettsial infections reflects the prevalence of rickettsiae in the vector population and the interactions of arthropod vectors with humans. Epidemic typhus is unusual among rickettsiae in that humans are considered the primary host. Transmission is by the human body louse; thus, outbreaks are now associated with breakdowns of social conditions. Endemic typhus is maintained in rodents and transmitted to humans by fleas. The various spotted fever group rickettsiae are limited geographically, probably by the distribution of the arthropod vector, although specific spotted fever group rickettsiae are found on all continents.[15]

LABORATORY SAFETY

The necessity of using embryonated eggs or cell culture techniques for the propagation of *Rickettsia* spp. incorporates extensive purification procedures. Accidental parenteral inoculation and exposure to infectious aerosols are the most likely sources of LAI.[18] Aerosol transmission of *R. rickettsii* has been experimentally documented in nonhuman primates.[19] Five cases of rickettsialpox recorded by Pike were associated with exposure to bites of infected mites.[8] Naturally and experimentally infected mammals, their

ectoparasites, and their infected tissues are potential sources of human infection. The organisms are relatively unstable under ambient environmental conditions.

Containment Recommendations

BSL-2 practices, containment equipment, and facilities are recommended for nonpropagative laboratory procedures, including serological and fluorescent antibody procedures, and for the staining of impression smears. BSL-3 practices, containment equipment, and facilities are recommended for all other manipulations of known or potentially infectious materials, including necropsy of experimentally infected animals and trituration of their tissues, and inoculation, incubation, and harvesting of embryonated eggs or cell cultures. ABSL-2 practices, containment equipment, and facilities are recommended for the holding of experimentally infected mammals other than arthropods. BSL-3 practices, containment equipment, and facilities are recommended for animal studies with arthropods naturally or experimentally infected with rickettsial agents of human disease (See Appendix E).

Several species, including *R. montana, R. rhipicephali, R. belli,* and *R. canada,* are not known to cause human disease and may be handled under BSL-2 conditions. New species are being described frequently and should be evaluated for appropriate containment on a case-by-case basis. Because of the proven value of antibiotic therapy in the early stages of ricketsial infection, it is essential that laboratories have an effective system for reporting febrile illnesses in laboratory personnel, medical evaluation of potential cases and, when indicated, institution of appropriate antibiotic therapy.

SPECIAL ISSUES

Medical Response Under natural circumstances, the severity of disease caused by rickettsial agents varies considerably. In the laboratory, very large inocula are possible, which might produce unusual and perhaps very serious responses. Surveillance of personnel for laboratory-associated infections with rickettsial agents can dramatically reduce the risk of serious consequences of disease. Experience indicates that infections adequately treated with specific anti-rickettsial chemotherapy on the first day of disease do not generally present serious problems. However, delay in instituting appropriate chemotherapy may result in debilitating or severe acute disease ranging from increased periods of convalescence in typhus and scrub typhus to death in *R. rickettsii* infections. The key to reducing the severity of disease from laboratory-associated infections is a reliable medical response which includes: 1) round-the-clock availability of an experienced medical officer, 2) indoctrination of all personnel on the potential hazards of working with rickettsial agents and advantages of early therapy, 3) a reporting system for all recognized overt exposures and accidents, 4) the reporting of all febrile illnesses, especially those associated with headache, malaise, and prostration when no other certain cause exists, and 5) an open and non-punitive atmosphere that encourages reporting of any febrile illness.

Select Agent *R. prowazekii* and *R. rickettsii* are Select Agents requiring registration with CDC and/or USDA for possession, use, storage and/or transfer. See Appendix F for additional information.

Transfer of Agent Importation of this agent may require CDC and/or USDA importation permits. Domestic transport of this agent may require a permit from USDA/APHIS/VS.

REFERENCES

1. Babudieri B. Q fever: a zoonosis. Adv Vet Sci. 1959;5:81-182.
2. Ignatovich VF. The course of inactivation of *Rickettsia burnetii* in fluid media. Zh Mikrobiol Epidemiol Immunobiol. 1959;30:134-41.
3. Sawyer LA, Fishbein DB, McDade JE. Q fever: current concepts. Rev Infect Dis. 1987;9:935-46.
4. Heinzen RA, Hackstadt T, Samuel JE. Developmental biology of *Coxiella burnetii*. Trends Microbiol. 1999;7:149-54.
5. Ormsbee R, Peacock M, Gerloff R, et al. Limits of rickettsial infectivity. Infect Immun. 1978;19:239-45.
6. Wedum AG, Barkley WE, Hellman A. Handling of infectious agents. J Am Vet Med Assoc. 1972;161:1557-67.
7. Maurin M, Raoult D. Q fever. Clin Microbiol Rev. 1999;12:518-53.
8. Pike RM. Laboratory-associated infections: Summary and analysis of 3,921 cases. Health Lab Sci. 1976;13:105-14.
9. Johnson JE, Kadull PJ. Laboratory-acquired Q fever. A report of fifty cases. Am J Med. 1966;41:391-403.
10. Spinelli JS, Ascher MS, Brooks DL, et al. Q fever crisis in San Francisco: controlling a sheep zoonosis in a lab animal facility. Lab Anim. 1981;10:24-7.
11. Meiklejohn G, Reimer LG, Graves PS, et al. Cryptic epidemic of Q fever in a medical school. J Infect Dis. 1981;144:107-13.
12. Welsh HH, Lennette EH, Abinanti FR, et al. Q fever in California IV. Occurrence of C*oxiella burnetii* in the placenta of naturally infected sheep. Public Health Rep. 1951;66:1473-7.
13. Bernard KW, Helmick CG, Kaplan JE, et al. Q fever control measures: Recommendations for research facilities using sheep. Infect Control. 1982;3:461-5.
14. Hackstadt T. Biosafety concerns and *Coxiella burnetii*. Trends Microbiol. 1996;4:341-2.
15. Hackstadt T. The biology of rickettsiae. Infect Agents Dis. 1996;5:127-43.
16. Centers for Disease Control and Prevention. Laboratory-acquired endemic typhus. MMWR Morb Mortal Wkly Rep. 1978;27:215-6.
17. Oster CN, Burke DS, Kenyon RH, et al. Laboratory-acquired Rocky Mountain spotted fever. The hazard of aerosol transmission. N Engl J Med. 1977;297:859-63.
18. Hattwick MA, O'Brien RJ, Hanson BF. Rocky Mountain spotted fever: epidemiology of an increasing problem. Ann Intern Med. 1976;84:732-9.

19. Saslaw S, Carlisle HN. Aerosol infection of monkeys with *Rickettsia rickettsii*. Bacteriol Rev. 1966;30:636-45.