**LESSON 9.  
Microbiology diagnosis of diseases, caused by *Spirochetes* and *Rickettsiae***

**LESSON PLAN:**

• Pathogenic spirochetes. General characteristics, classification.

• Treponemas. The causative agent of syphilis, morpho-biological characteristics, antigen structure, pathogenicity factors, pathogenesis. The causative agents of syphilis-like diseases (frambesis, pinta). Microbiological diagnostics: microscopic and serological methods (research of treponemal and non-treponemal antibodies (RPR, DIFR, TPHA, etc.) and their diagnostic significance. Principles of treatment of syphilis.

• Borrelia. The causative agents, morpho-biological characteristics, pathogenicity factors, pathogenesis. Microbiological diagnostics. The causative agent of Lyme disease, the pathogenesis of the disease. Microbiological diagnostics.

• Leptospirosis causative agent, morpho-biological characteristics, classification. Source of infection, ways of infection, pathogenesis, microbiological diagnosis, principles of specific treatment and prevention.

• Pathogenic rickettsiae, morpho-biological characteristics. Classification of rickettsioses.

• Causes of scabies (Rickettsia prowazekii, Rickettsia typhi), pathogenicity factors, pathogenesis and microbiological diagnosis. Principles of specific treatment and prevention.

• Pathogenesis and microbiological diagnosis of diseases caused by spotted fever group rickettsiae (spotted fever of steep rocks - R. rickettsii, Marseille fever - R. conorii, flower-like rickettsiosis - R. akari, North Asian tick-borne rickettsiosis - R. sibirica).

• Orientia tsutsugamushi – Bush fever, morpho-biological characteristics, pathogenesis and microbiological diagnosis.

• Genus Ehrlichia (monocytic ehrlichiosis – E. sennetsu, E. chaffeesis, granulocytic ehrlichiosis – E. ewingii, Anaplasma phagocytophilum), morpho-biological characteristics, pathogenesis and microbiological diagnosis of the diseases they cause

• The causative agent of Q-fever (Coxiella burnetii), morpho-biological characteristics, pathogenesis and microbiological diagnosis.

The bacteria in the order Spirochaetales have been grouped together on the basis of their common morphologic properties. These spirochetes are thin, helical (0.1 to 0.5 × 5 to 20 μm), gram-negative bacteria. The order Spirochaetales is subdivided into 4 families and 14 genera, of which 3 genera (*Treponema* and *Borrelia* in the family Spirochaetaceae, and *Leptospira* in the family Leptospiraceae) are responsible for human disease.

***TREPONEMA PALLIDUM***

**Trigger Words**

Thin spirochete, sexually transmitted disease, congenital infections, painless ulcer (chancre)

**Biology and Virulence**

ᑏᑏ Coiled spirochete (0.1 to 0.2 × 6 to 20 μm) too thin to be seen with Gram or Giemsa stains; observed by darkfield microscopy

ᑏᑏ Outer membrane proteins promote adherence to host cells

ᑏᑏ Hyaluronidase facilitates perivascular infiltration

ᑏᑏ Coating of fibronectin protects against phagocytosis

ᑏᑏ Tissue destruction primarily results from host’s immune response to infection

**Epidemiology**

ᑏᑏ Humans are the only natural host

ᑏᑏ Syphilis transmitted by sexual contact or congenitally

ᑏᑏ Syphilis occurs worldwide, with no seasonal incidence

**Diseases**

ᑏᑏ Syphilis presents as primary disease (painless ulcer [chancre] at site of infection, with regional lymphadenopathy and bacteremia), secondary syphilis (flulike syndrome with generalized mucocutaneous rash and bacteremia), and late-stage disease (diffuse chronic inflammation and destruction of any organ or tissue); congenital (latent multiorgan malformations, fetal death)

**Diagnosis**

ᑏᑏDarkfield or direct fluorescent antibody microscopy is useful if mucosal ulcers are observed in primary or secondary stages of syphilis

ᑏᑏ Serology is very sensitive in secondary and late stages of syphilis

**Treatment, Prevention, and Control**

ᑏᑏ Penicillin is the drug of choice; doxycycline is administered if patient is allergic to penicillin

ᑏᑏ Safe sex practices should be emphasized, and sexual partners of infected patients should be treated

ᑏᑏ No vaccine is available

***BORRELIA***

**Trigger Words**

Large spirochetes, erythema migrans, Lyme disease, relapsing fever, hard and soft ticks, body louse

**Biology and Virulence**

ᑏᑏ Borreliae are large (0.2 to 0.5 × 8 to 30 μm) and can be seen when stained with aniline dyes (e.g., Giemsa, Wright stains)

ᑏᑏ Immune reactivity against Lyme disease agents may be responsible for clinical disease

***EPIDEMIOLOGY***

**Lyme Disease**

ᑏᑏ *B. burgdorferi* causes disease in the United States and Europe; *B. garinii* and *B. afzelii* cause disease in Europe and Asia

ᑏᑏ Transmitted by hard ticks from mice to humans; reservoirs include mice, deer, and ticks; vectors include *Ixodes scapularis* in eastern and midwestern United States, *I. pacificus* in the western United States, *I. ricinus* in Europe, and *I. persulcatus* in Eastern Europe and Asia

ᑏᑏ Most Lyme disease cases in the United States are from two principal foci: Northeast and Mid-Atlantic states (Maine to Virginia) and the Upper Midwest (Minnesota, Wisconsin)

ᑏᑏ Individuals at risk for Lyme disease include people exposed to ticks in areas of high endemicity

ᑏᑏ Worldwide distribution

ᑏᑏ Seasonal incidence corresponds to feeding patterns of vectors; most cases of Lyme disease in the United States occur in late spring and early summer (feeding pattern of nymph stage of ticks); peak in June and July

**Epidemic Relapsing Fever**

ᑏᑏ Etiologic agent is *B. recurrentis*

ᑏᑏ Person-to-person transmission; reservoir includes humans; vector includes human body louse

ᑏᑏ Individuals at risk are people exposed to lice (epidemic disease) in crowded or unsanitary conditions

ᑏᑏ Occurs in Ethiopia, Eritrea, Somalia, and Sudan

**Endemic Relapsing Fever**

ᑏᑏ Many *Borrelia* species are responsible

ᑏᑏ Transmitted from rodents to humans; reservoirs include rodents, small mammals, and soft ticks; vector includes soft ticks

ᑏᑏ Individuals at risk are people exposed to ticks (endemic disease) in rural areas

ᑏᑏ Worldwide distribution; in the western part of the United States

**Diseases**

ᑏᑏ Borreliae are responsible for two human diseases: Lyme disease and relapsing fever (epidemic and endemic)

ᑏᑏ *Borrelia* species responsible for relapsing fever are able to undergo antigenic shift and escape immune clearance; periodic febrile and afebrile periods result from

antigenic variation

**Diagnosis**

ᑏᑏ Serology is test of choice for Lyme disease

ᑏᑏ Polymerase chain reaction tests available for Lyme disease but relatively insensitive

ᑏᑏMicroscopy is the test of choice for diagnosis of relapsing fever

**Treatment, Prevention, and Control**

ᑏᑏ For early localized or disseminated Lyme disease, treatment is with amoxicillin, tetracycline, cefuroxime; late manifestations are treated with intravenous penicillin or ceftriaxone

ᑏᑏ For relapsing fever, treatment is with tetracycline or erythromycin

ᑏᑏ Improved sanitary conditions to decrease risk of epidemic relapsing fever

ᑏᑏ Reduced exposure to hard ticks (Lyme disease) and soft ticks (relapsing fever) through use of insecticides, application of insect repellents to clothing, and wearing protective clothing that reduces exposure of skin to insects

***LEPTOSPIRA***

**Trigger Words**

Thin, spirochetes, flulike disease, aseptic meningitis, Weil disease, zoonotic, contaminated water exposure

**Biology and Virulence**

ᑏᑏ Thin, coiled spirochetes (0.1 × 6 to 20 μm) that grow slowly in specialized cultures

ᑏᑏ Able to directly invade and replicate in tissues, inducing an inflammatory response

ᑏᑏ Immune complex produces renal disease (glomerulonephritis)

ᑏᑏ Most disease is a mild virus-like syndrome

ᑏᑏ Systemic leptospirosis presents most commonly as aseptic meningitis

ᑏᑏ Over whelming disease (Weil disease) is characterized by vascular collapse, thrombocytopenia, hemorrhage, and hepatic and renal dysfunction

**Epidemiology**

ᑏᑏ US reservoirs: rodents (particularly rats), dogs, farm animals, and wild animals

ᑏᑏ Humans: accidental end-stage host

ᑏᑏ Organism can penetrate the skin through minor breaks in the epidermis

ᑏᑏ People are infected with leptospires through exposure to water contaminated with urine from an infected animal or handling of tissues from an infected animal

ᑏᑏ People at risk are those exposed to urine-contaminated streams, rivers, and standing water; occupational exposure to infected animals for farmers, meat handlers, and veterinarians

ᑏᑏ Infection is rare in the United States but has worldwide distribution

ᑏᑏ Disease is more common during warm months (recreational exposure)

**Diagnosis**

ᑏᑏMicroscopy not useful because too few organisms are generally present in fluids or tissues

ᑏᑏCulture blood or cerebrospinal fluid in the first 7 to 10 days of illness; urine after the first week

ᑏᑏ Serology using the microscopic agglutination test is relatively sensitive and specific but not widely available in resource-limited countries; enzymelinked immunosorbent assay tests are less accurate but can be used to screen patients

**Treatment, Prevention, and Control**

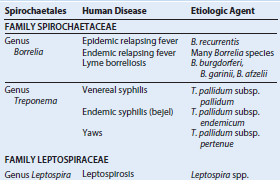
ᑏᑏ Treatment with penicillin or doxycycline

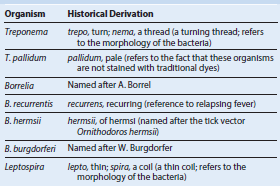
ᑏᑏ Doxycycline but not penicillin is used for prophylaxis

ᑏᑏ Herds and domestic pets should be vaccinated

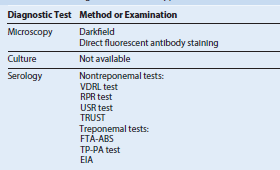
ᑏᑏ Rats should be controlled

**Medically Important Genera in the Order Spirochaetales**





Diagnostic Tests for Syphilis



**Conditions Associated with False-Positive Serologic Test Results for Syphilis**

**Nontreponemal Tests**

Viral infection

Rheumatoid arthritis

Systemic lupus erythematosus

Acute or chronic illness

Pregnancy

Recent immunization

Drug addiction

Leprosy

Malaria

Multiple blood transfusions

**Treponemal Tests**

Pyoderma

Rheumatoid arthritis

Systemic lupus erythematosus

Psoriasis

Crural ulceration

Skin neoplasm

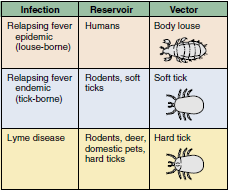
Drug addiction

Mycoses

Lyme disease

Acne vulgaris

**Epidemiology of *Borrelia* infections.**



**Definition of Lyme Disease**

**Clinical Case Definition**

Either of the Following:

Erythema migrans (≈5 cm in diameter)

At least one late manifestation (i.e., musculoskeletal, nervous system, or cardiovascular involvement) and laboratory confirmation of infection

**Laboratory Criteria for Diagnosis**

At Least One of the Following:

Isolation of *Borrelia burgdorferi*

Demonstration of diagnostic levels of IgM or IgG antibodies to the spirochetes

Significant increase in antibody

**Bacteria and Diseases Associated with Cross-Reactions in Serologic Tests for Lyme Borreliosis**

*Treponema pallidum*

Oral spirochetes

Other *Borrelia* species

Juvenile rheumatoid arthritis

Rheumatoid arthritis

Systemic lupus erythematosus

Infectious mononucleosis

Subacute bacterial endocarditis

***RICKETTSIA RICKETTSII***

**Trigger Words**

Intracellular bacteria, Rocky Mountain spotted fever, vasculitis, tick, micro immunofluorescence test

**Biology and Virulence**

ᑏᑏ Small intracellular bacteria

ᑏᑏ Stain poorly with Gram stain; best with Giemsa or Gimenez stains

ᑏᑏ Replication occurs in cytoplasm and nucleus of endothelial cells, with resulting vasculitis

ᑏᑏ Intracellular growth protects the bacteria from immune clearance

**Epidemiology**

ᑏᑏ *R. rickettsii* is the most common rickettsial pathogen in the United States

ᑏᑏ Hard ticks (e.g., dog tick, wood tick) are the primary reservoirs and vectors

ᑏᑏ Transmission requires prolonged contact

ᑏᑏ Distribution in Western Hemisphere; in United States, the majority of infections are reported in five states: North Carolina, Oklahoma, Arkansas, Tennessee, and Missouri

ᑏᑏ Disease is most common from April through September

**Diseases**

ᑏᑏ Rocky Mountain spotted fever characterized by high fever, severe headache, myalgias, and rash; complications common in untreated patients or where diagnosis is delayed

**Diagnosis**

ᑏᑏ Serology (e.g., microimmunofluorescence test) is used most commonly for diagnosis

**Treatment, Prevention, and Control**

ᑏᑏ Doxycycline is the drug of choice

ᑏᑏ People should avoid tick-infested areas, wear protective clothing, and use effective insecticides

ᑏᑏ People should remove attached ticks immediately

ᑏᑏ No vaccine is currently available

***RICKETTSIA PROWAZEKII***

**Trigger Words**

Intracellular bacteria, louse-borne typhus, Brill-Zinsser disease, vasculitis, human reservoir, micro immunofluorescence test

**Biology and Virulence**

ᑏᑏ Small intracellular bacteria

ᑏᑏ Stain poorly with Gram stain; best with Giemsa or Gimenez stains

ᑏᑏ Replicate in cytoplasm of endothelial cells, with resulting vasculitis

ᑏᑏ Intracellular growth protects the bacteria from immune clearance

**Epidemiology**

ᑏᑏ Humans are the primary reservoir, with person-to-person transmission by louse vector

ᑏᑏ It is believed that sporadic disease is spread from squirrels to humans via squirrel fleas

ᑏᑏ Recrudescent disease can develop years after initial infection

ᑏᑏ People at greatest risk are those living in crowded, unsanitary conditions

ᑏᑏ Disease is worldwide, with most infections in Central and South America and Africa

ᑏᑏ Sporadic disease is seen in the eastern United States

**Diseases**

ᑏᑏ Epidemic typhus (louse-borne typhus) characterized by high fever, severe headache, and myalgias

ᑏᑏ Recrudescent typhus (Brill-Zinsser disease) is a milder form of the disease

**Diagnosis**

ᑏᑏ The microimmunofluorescence test is the test of choice

**Treatment, Prevention, and Control**

ᑏᑏ Doxycycline is the drug of choice

ᑏᑏ Controlled through improvements in living conditions and reduction of the lice population through use of insecticides

ᑏᑏ Inactivated vaccine is available for high risk populations

***EHRLICHIA AND ANAPLASMA***

**Trigger Words**

ᑏᑏ Intracellular bacteria, monocytic and granulocytic disease, ticks

**Biology and Virulence**

ᑏᑏ Small intracellular bacteria that stain poorly with Gram stain; best with Giemsa or Gimenez stains

ᑏᑏ Replicates in phagosome of infected cells

ᑏᑏ Intracellular growth protects bacteria from immune clearance

ᑏᑏ Able to prevent fusion of phagosome with lysosome of monocytes or granulocytes

ᑏᑏ Initiates inflammatory response that contributes to pathology

**Epidemiology**

ᑏᑏ Depending on the species of *Ehrlichia,* important reservoirs are white-tailed deer, white-footed mouse, chipmunks, voles, and canines

ᑏᑏ Ticks are important vectors, but transovarian transmission in inefficient

ᑏᑏ Disease in United States is most common in the southeastern, Mid-Atlantic, midwestern, and south central states

ᑏᑏ People at greatest risk are those exposed to ticks in the endemic areas

ᑏᑏ Disease is most common from April to October

**Diseases**

ᑏᑏ Diseases are human monocytic ehrlichiosis and human anaplasmosis (formerly called *human granulocytic ehrlichiosis*)

**Diagnosis**

ᑏᑏMicroscopy of limited value

ᑏᑏ Serology and nucleic acid amplification tests are methods of choice

**Treatment, Prevention, and Control**

ᑏᑏ Doxycycline is the drug of choice; rifampin is an acceptable alternative

ᑏᑏ Prevention involves avoidance of tick infested areas, use of protective clothing and insect repellents, and prompt removal of embedded ticks

ᑏᑏ Vaccines are not available

***COXIELLA BURNETII***

**Trigger Words**

Intracellular bacteria, flulike illness, subacute endocarditis, inhalation exposure, phase I and II antigens

**Biology and Virulence**

ᑏᑏ Small intracellular bacteria that stain poorly with Gram stain; best with Giemsa or Gimenez stains

ᑏᑏ Replicate in phagosomes of infected cells

ᑏᑏ Exists in two forms: small cell variant infectious, extremely stable to environmental factors; large cell variant is the metabolically active form

ᑏᑏ Phase transition occurs during infection: phase I with intact LPS, phase II with truncated LPS (O-antigen sugars missing)

ᑏᑏ Intracellular growth protects the bacteria from immune clearance

ᑏᑏ Able to replicate in acidic environment of phagosomes

ᑏᑏ Extracellular form extremely stable; can survive in nature for a prolonged period

**Epidemiology**

ᑏᑏ Many reservoirs, including mammals, birds, and ticks

ᑏᑏ Most human infections associated with contact with infected cattle, sheep, goats, dogs, and cats

ᑏᑏ Most disease acquired through inhalation; possible disease from consumption of contaminated milk; ticks are not an important vector for human disease

ᑏᑏ Worldwide distribution

ᑏᑏ No seasonal incidence

**Diseases**

ᑏᑏ Most infections are asymptomatic; most common acute presentation is nonspecific influenza-like syndrome; less than 5% develop significant acute disease (pneumonia, hepatitis, pericarditis, fever)

ᑏᑏ Endocarditis most common form of chronic disease

**Diagnosis**

ᑏᑏDetection of antibody response to phase Iand phase II antigens is test of choice

**Treatment, Prevention, and Control**

ᑏᑏ Doxycycline is the drug of choice for acute infections; hydroxychloroquine combined with doxycycline is used to treat chronic infections

ᑏᑏ Phase I antigen vaccines are protective and safe if administered in a single dose before the animal or human has been exposed to *Coxiella;* not available in the United States for animals or humans

**Organism Historical Derivation**

*Rickettsia rickettsii -*Named after Howard Ricketts, who implicated the wood tick as the vector of Rocky Mountain spotted fever

*R. akari akari, -* mite; the vector of rickettsia pox

*R. prowazekii -* Named after Stanislav von Prowazek, an early investigator of typhus who was a victim of this disease

*R. typhi - typhi,* typhus or fever

*Orientia tsutsugamushi - Orientia,* Orient; *tsutsugamushi,* “mite disease,” the popular name of this disease in the Orient

*Ehrlichia -* Named after the German microbiologist Paul Ehrlich

*E. chaffeensis -* First isolated in an Army reservist at Fort Chaffee, Arkansas

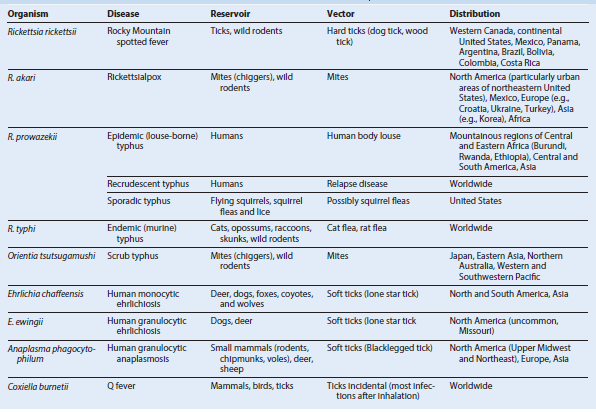
*E. ewingii -* Named after the American microbiologist William Ewing

*Anaplasma- an,* without; *plasma,* anything formed (a thing without form; referring to the intracytoplasmic inclusions)

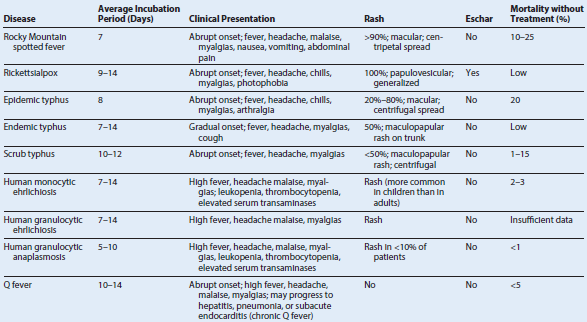
*A. phagocytophilum - phago,* to eat; *kytos,* a vessel or enclosure; *philein,* to love (found in phagocytes)

*Coxiella burnetiid -* Named after Herald Cox and F.M. Burnet who isolated the bacterium from ticks in Montana and patients in Australia, respectively

**Epidemiology of Infections Caused by *Rickettsia* and Related Bacteria**

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**Human Diseases Caused by Rickettsia and Related Bacteria**

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