TREPONEMA, BORRELIA AND LEPTOSPIRA

The collection of bacteria in the order Spirochaetales has been grouped together based on their common morphological properties. These "spirochetes" are **thin, helical** (0.1 to 0.5 x 5 to 20 μm), **gram-negative bacteria**. The order Spirochaetales is subdivided into two families and seven genera, of which three (*Treponema*, *Borrelia*, and *Leptospira*) are responsible for human disease.

25.1 Treponema

The two treponemal species responsible for human disease are ***T. pallidum*** (with three subspecies) and ***T. carateum***. All are morphologically identical, elicit the same serological response in humans (e.g., positive reactivity in the VDRL, FTA-ABS, MHA-TP tests), and are **susceptible to penicillin**. The organisms are distinguished by the epidemiology and clinical manifestations of their disease. ***T. pallidum* subspecies *pallidum* (referred to as *T. pallidum* in this lecture) is the etiological agent of the venereal disease syphilis; *T. pallidum* ssp. *endemicum* is responsible for bejel; *T. pallidum* ssp. *pertenue* is responsible for yaws; and *T. carateum* is responsible for pinta. Bejel, yaws, and pinta are nonvenereal diseases.**

Physiology and Structure

Syphilis is a sexually transmitted disease that has plagued humans for centuries. The spirochete responsible for this disease is a **strict human pathogen**. Natural syphilis is not found in any other species, and experimental syphilis has been established only in rabbits. *T. pallidum* is a thin**, coiled spirochete (0.1 x 5 to 15 μm)** that **cannot be grown in cell-free cultures**. Limited growth has been achieved in cultured rabbit epithelial cells, but replication is slow (doubling time, 30 hours) and can be maintained only for a few generations. The spirochetes were once considered strict anaerobes; however, it is now known that they can utilize glucose oxidatively.

The spirochetes are **too thin to be seen with light microscopy** in specimens stained with Gram or Giemsa stains. Motile forms can be visualized by **darkfield illumination or by staining with specific antitreponemal antibodies labeled with fluorescent dyes**.

Pathogenesis and Immunity

The inability to grow *T. pallidum* to high concentrations in vitro has limited detection of specific virulence factors in this organism. **The outer membrane proteins are associated with adherence to the surface of host cells**, and virulent spirochetes produce **hyaluronidase**, which may facilitate perivascular infiltration. Virulent spirochetes are also **coated with host cell fibronectin**, which can protect against phagocytosis.

Tissue destruction and lesions observed in syphilis are primarily the **consequence of the patient's immune response to infection.** The clinical course of syphilis evolves through **three phases**. The initial, or **primary**, phase is characterized by one or more **skin lesions (chancres)** at the site of spirochete penetration. Although dissemination of spirochetes in the bloodstream occurs soon after infection, the chancre represents the **primary site of initial replication**. Histological examination of the lesion reveals **endarteritis and periarteritis** (characteristic of syphilitic lesions at all stages) and infiltration of the ulcer with polymorphonuclear leukocytes and macro-phages. Ingestion of spirochetes by the phagocytic cells is seen, but the organisms frequently survive. The **secondary phase of syphilis** heralds clinical signs of disseminated disease, with prominent **skin lesions dispersed over the entire body surface**. Spontaneous remission may occur after either the primary or secondary stages, or the patient may develop **late manifestations of disease** in which virtually all tissues may be involved. Each phase represents localized multiplication of the spirochete and tissue destruction. Although replication is slow, large numbers of organisms are present in the initial chancre, as well as in the secondary lesions following dissemination of the spirochetes in the bloodstream.

Epidemiology

Syphilis is found worldwide and is **the third most common sexually transmitted disease** in the United States (after *Neisseria gonorrhoe* as and *Chlamydia infections*). Although the incidence of disease had steadily decreased since the early 1940s with the advent of penicillin therapy, this trend reversed in the mid-1980s.

Because natural syphilis is exclusive to humans and has no other known natural hosts, the most common method of spread is by direct **sexual contact**. *T. pallidum* is **extremely labile**, unable to survive exposure to drying or disinfectants. Thus inanimate objects such as toilet seats cannot contribute to the spread of syphilis. **Direct person-to-person contact is required** for transmission. This disease can also be acquired **congenitally or by transfusion with contaminated blood.** Syphilis **is not highly contagious**, with the risk of disease following a single sexual contact estimated to be 30%. However, contagiousness is influenced by the **stage of disease** in the infectious individual. As mentioned previously, the spirochetes are unable to survive on dry skin surfaces. Thus *T. pallidum* is **transferred primarily during the early stages of disease** when large numbers of organisms **are present in moist cutaneous or mucosal lesions**. **Congenital transmission** to the fetus can take place soon after the mother is infected because bacteremia characteristically occurs early during the course of the disease. A woman with untreated disease can have spontaneous bacteremia for as long as 8 years, transmitting the spirochetes to fetal tissues if she becomes pregnant during this period. After 8 years the disease can remain active, but bacteremia is not believed to occur.

With the advent of effective antimicrobial therapy, the incidence of late (tertiary) syphilis has markedly decreased. Although antibiotic therapy has decreased the length ofinfectivity in infected individuals, the incidence of primary and secondary syphilis has remained high because

of sexual practices, particularly prostitution to support drug habits. The incidence of congenital syphilis corresponds to the pattern of syphilis in women of childbearing age.

Clinical Syndromes

Primary Syphilis

The initial syphilitic chancre **develops at the site of inoculation**. The lesion starts as a **papule but then erodes to form a painless ulcer with raised borders**. In most patients painless **regional lymphadenopathy** develops 1 to 2 weeks after the appearance of the chancre, which represents a local focus for proliferation of spirochetes. Abundant spirochetes are present in the chancre and are able to disseminate throughout the patient via the lymphatics and bloodstream. The fact that **this ulcer heals spontaneously within 2 months** gives the patient a false sense of relief.

Secondary Syphilis

Clinical evidence of disseminated disease marks the second stage of syphilis. This stage is characterized by a **flu-like syndrome**, with **sore throat, headache, fever, myalgias, anorexia, generalized lymphadenopathy, and a generalized mucocutaneous rash**. The flu-like syndrome and **lymphadenopathy** generally appear first and then are followed a few days later by the disseminated skin lesions. **The rash can be quite variable (macular, papular, pustular), cover the entire skin surface** (**including palms and soles**), and can resolve slowly over a period of weeks to months. As with the primary chancre, **the rash in secondary syphilis is highly infectious**. Gradually, the rash and symptoms resolve spontaneously and the patient enters the latent or clinically inactive stage of disease.

Late Syphilis

A small proportion of patients can progress to the tertiary stage of syphilis. The diffuse, chronic inflammation characteristic of late syphilis **can cause devastating destruction of virtually any organ or tissue (e.g., arteritis, dementia, blindness). Granulomatous lesions (gummas) may be found in bone, skin, and other tissues.** The nomenclature of late syphilis reflects the organs of primary involvement (e.g., **neurosyphilis or cardiovascular syphilis**). An increased incidence of neurosyphilis despite adequate therapy for early syphilis has been documented in patients with acquired immunodeficiency syndrome (AIDS).

Congenital Syphilis

In utero infections can lead to significant fetal disease, resulting in death, **multiorgan malformations, or latent infections**. Most infected infants are born without clinical evidence of disease but then develop rhinitis followed by a widespread desquamating maculopapular rash. Late bony destruction and cardiovascular syphilis are common in untreated infants who survive the initial course of disease.

Treponema is a genus of commonly found **oral bacteria** that are closely related to **periodontitis and the etiology of implant periarthritis**. Species commonly detected in the **oral cavity are *Treponema denticola*, *Treponema scaliodontum*, *Treponema macrodentium*, *Treponema oralis*, *Treponema intermedia*, *Treponema maltophilum*, *Treponema socranskii*, and *Treponema vincentii*.**

Laboratory diagnosis

There are three important approaches.

A. Microscopy: Spirochetes are demonstrated in early lesions by **darkfield or immunofluorescence microscopy**. They are not seen on a Gram-stained smear.

B. Nonspecific Serologic Tests: These tests involve the **use of nontreponemal antigens**. Extracts of normal mammalian tissues (eg, **cardiolipin from beef heart**) react with antibodies in serum samples from patients with syphilis. These antibodies, which are a mixture of IgG and IgM, are called **"reagin"** antibodies. **Flocculation tests, eg, VDRL** (Venereal Disease Research Laboratory) **and RPR** (rapid plasma reagin) tests, detect the presence of these antibodies. These tests are **positive in most cases of primary syphilis and are almost always positive in secondary syphilis.** The titer of these nonspecific antibodies decreases with effective treatment, in contrast to the specific antibodies, which are positive for life.

False-positive reactions occur in infections such as leprosy, hepatitis B, and infectious mononucleosis and in various autoimmune diseases. Therefore, positive results have to be confirmed by specific tests. Results of nonspecific tests usually become negative after treatment and should be used to determine the response to treatment. These tests can also be **falsely negative** as a result of the prozone phenomenon. In the prozone phenomenon, the titer of antibody is too high (antibody excess) and no flocculation will occur. On dilution of the serum, however, the test becomes positive. These tests are inexpensive and easy to perform and therefore are used as a method of screening the population for infection.

The laboratory diagnosis of congenital syphilis is based on the finding that the infant has a higher titer of antibody in the VDRL test than has the mother. Furthermore, if a positive VDRL test on the infant is a false-positive result because maternal antibody has crossed the placenta, the titer will decline with time. If the infant is truly infected, the titer will remain high. However, irrespective of the VDRL results, any infant whose mother has syphilis should be treated.

C. Specific Serologic Tests: These tests involve the use of treponemal antigens and therefore are more specific than those described above. In these tests, *T. pallidum* reacts in **immunofluorescence (FTA-ABS) or hemagglutination (TPHA, MHA-TP) assays** with specific treponemal antibodies in the patient's serum. These **antibodies arise within 2-3 weeks of infection**, and so the tests are positive in most patients with primary syphilis. These tests remain positive for life after effective treatment and cannot be used to determine the response to

treatment or reinfection. They are **more expensive and more difficult to perform** than the nonspecific tests and therefore are not used as screening procedures.

Treatment, Prevention, and Control

Penicillin is the drug of choice for treating infections with *T. pallidum*. **Long-acting benzathine penicillin** is used for the early stages of syphilis, and penicillin G is recommended for congenital and late syphilis. **Tetracycline, erythromycin, and chloramphenicol** can be used as alternative antibiotics **for patients allergic to penicillin**. **Only penicillin or chloramphenicol can be used for patients with neurosyphilis.**

Because protective vaccines are not available, control of syphilis requires the practice of **safe sex techniques** and adequate contact and **treatment of sex partners of patients** who have documented infections. In recent years control of syphilis and other venereal diseases has been complicated by increases in prostitution in drug abusers.

OTHER TREPONEMES

Three other nonvenereal treponemal diseases are important: bejel, yaws, and pinta. The diseases are **primarily observed in impoverished children**.

***T.pallidum* ssp. *endemicum* is responsible for bejel, also called endemic syphilis**. Disease is **spread person-to-person by use of contaminated eating utensils**. The initial oral lesions are rarely observed, but secondary lesions include oral papules and mucosal patches. Late manifestations consist of gummas of the skin, bones, and nasopharynx. The disease is present in Africa, Asia, and Australia.

***T. pertenue* is the etiological agent of yaws, a granulomatous disease with early skin lesions** and then late destructive lesions of the skin, lymph nodes, and bones. The disease is present in primitive, tropical areas in parts of South America, Central Africa, and Southeast Asia and is spread by direct contact with infected skin lesions.

***T. carateum* is responsible for pinta, a disease primarily restricted to the skin.** After a 1- to 3-week incubation period small pruritic papules develop on the skin surface. These lesions enlarge and persist for months to years before resolution. Disseminated, recurrent, hypopigmented lesions can develop over years, resulting in scarring and disfigurement. Pinta is present in Mexico and Central and South America, and is also spread by direct contact with infected lesions.

Bejel, yaws, and pinta are **diagnosed by their clinical presentation in an endemic area**. The diagnoses of yaws or pinta are **confirmed by the detection ofspirochetes in skin lesions by darkfield microscopy** (this test cannot be used for the oral lesions in bejel). **Serologic tests for syphilis are also positive** but may develop only late in the disease course.

Penicillin, tetracycline, and chloramphenicol have been used to treat both diseases. Control of the disease is managed by treating infected individuals and eliminating person-to-person spread.

25.2 Borrelia

Members of the genus *Borrelia* are **responsible for two important human diseases:** relapsing fever and Lyme disease. **Relapsing fever** is a **febrile illness characterized by recurrent episodes of fever and septicemia, separated by afebrile periods**. Two forms of the disease are recognized. *Borrelia recurrentis* is the etiological agent of epidemic or louse-borne relapsing fever and is spread person-to-person by the human body louse (*Pediculus humanus*). Endemic relapsing fever is caused by many species of borreliae and is spread by infected ticks.

In 1977 an unusual cluster of children with arthritis in Lyme, Connecticut was described. Five years later the spirochete responsible for this disease was discovered by Burgdorfer**. Lyme disease** is a **tick-borne disease** with dermatological, rheumatological, neurological, and cardiac abnormalities. **The best clinical marker for the disease is the initial skin lesion, erythema migrans,** that occurs in 60% to 70% of infected adults and less frequently in children. Until recently all cases of Lyme disease (or Lyme borreliosis) were believed to be caused by one organism, ***B. burgdorferi***.

Physiology and Structure

Members of the genus *Borrelia* are **weakly staining, gram-negative bacilli** that resemble other spirochetes. They tend to be larger (0.2 to 0.5 x 3 to 30 µm), stain well with aniline dyes (e.g., Giemsa, Wright), and can be **easily seen in smears of peripheral blood** collected from patients with relapsing fever. From **7 to 20 periplasmic flagella** (depending on the individual species) are present between the periplasmic cylinder and the outer envelope and are responsible for the organism's **twisting motility**. Borreliae are **microaerophilic** and have **complex nutritional requirements**, making recovery in culture difficult. The species that have been successfully cultured have generation times of 18 hours or longer. Because culture is generally unsuccessful, **diagnosis of diseases caused by borreliae is by microscopy (relapsing fever) or serology (Lyme disease).**

Pathogenesis and Immunity

After exposure to infected arthropods, borreliae are able to spread in the bloodstream to multiple organs. Members of the genus do not produce recognized toxins and are rapidly removed when a specific antibody response is mounted. **The periodic febrile and afebrile cycles of relapsing fever are due to the ability of the borreliae to undergo antigenic variation**. When specific IgM antibodies are formed, agglutination with complement-mediated lysis occurs and

the borreliae are rapidly cleared from the bloodstream. However, organisms residing in internal tissues are able to alter their sero-typespecific outer envelope proteins through gene re-arrangement and emerge as antigenically novel organisms. Clinical manifestations of relapsing fever are in part a response to the **release of endotoxin by the organism.**

***B. burgdorferi* is present in low numbers in the skin tissues** when erythema migrans develops. **In the late manifestations of disease, spirochetes have also been isolated from clinical material.** Although the initial immune response to the organism is depressed, antibodies develop over a period of months to years and are responsible for complement-mediated clearance of the borreliae.

Epidemiology

The vectors for relapsing fever are **soft-shelled ticks** (*Ornithodorus* species) and the **human body louse**. **Humans are the only reservoir for *B. recurrentis*,** the etiological agent of louse-borne epidemic relapsing fever. After lice are infected, the organisms pass through the wall of the gut and multiply in hemolymph. Infected lice do not survive for more than a few months, so maintenance of disease requires crowded, unsanitary conditions (wars, natural disasters) that permit frequent contact with infected lice. **In contrast, tick-borne relapsing fever is a zoonotic disease, with rodents, small mammals, and ticks acting as the main reservoirs**. Despite the fact the borreliae produce a disseminated infection in ticks, the arthropods are able to survive and maintain an endemic reservoir of infection by transovarian transmission. Furthermore, ticks can survive for months to years between feedings.

Louse-borne relapsing fever is endemic in both tropical and temperate regions of the world, particularly Central and Eastern Africa and South America. Tick-borne disease is widespread.

Despite the relatively recent recognition of Lyme disease in the United States, retrospective studies have demonstrated that the disease was present for many years in this and other countries. Lyme disease currently has been described on six continents, in at least 20 countries, and in 46 states of the United States. The incidence of disease has risen dramatically since 1982 when 497 cases were reported to 1990 with nearly 8000 cases reported. Lyme disease is the leading vector-borne disease in the United States. Hard-shelled ticks are the major vectors of Lyme disease – *Ixodes dammini* in the Northeast and Midwest, and *I. pacificus* on the West Coast. *I. ricinus* is the major tick vector in Europe. The major reservoir hosts in the United States are the white-footed mouse and white-tail deer.

Only 30% of patients with Lyme disease recall a specific tick bite. The reason is that the nymph stages of the tick, as well as adult ticks, can transmit disease. The small nymph forms are responsible for the majority of infections. Most Lyme disease infections are reported from spring to fall, corresponding to activity of infected ticks. Person-to-person spread has not been reported.

Clinical Syndromes

Relapsing Fever

The clinical presentations of epidemic louse-borne and endemic tick-borne relapsing fever are essentially the same. After a **1-week incubation period**, the **abrupt onset** of disease is heralded with **shaking chills, fever, muscle aches, and headache. Splenomegaly and hepatomegaly** are commonly present. The symptoms correspond to the bacteremic phase of the disease and are relieved after 3 to 7 days, when the borreliae are cleared from the blood. **Bactercmia and fever return after a 1-week afebrile period**. The clinical symptoms are generally milder and shorter during this and subsequent febrile episodes. **Two or three relapses** are common, although the number of relapses can be as many as 13. The clinical course and outcome of epidemic relapsing fever tend to be more severe than with endemic disease, but this may be related to the patient's underlying poor state of health. Mortality with endemic disease is less than 5% but can be from 4% to 40% in epidemic disease.

Lyme Disease

Clinical diagnosis of Lyme disease is **complicated by the varied presentations of this disease and the lack of reliable diagnostic tests**. After an **incubation period of 3 to 30 days**, **one or more skin lesions develops at the site of the tick bite**. **The lesion (erythema migrans)** begins as a small macule or papule and then enlarges over the next few weeks, covering an area ranging from 5 to more than 50 cm in diameter. As the lesion develops, it will typically appear with a red, flat border and central clearing; however, erythema, vesicle formation, and central necrosis can also be seen. Within weeks the lesion will fade and disappear, although new, transient lesions may subsequently appear. Other early signs and symptoms of Lyme disease include **malaise, severe fatigue, headache, fever, chills, musculoskeletal pains, myalgias, and lymph-adenopathy**. These will last for an average of 4 weeks.

Late manifestations develop in almost 80% of untreated patients with Lyme disease; these occur **within a week of the onset of disease to more than 2 years later.** Two phases can be seen. The first involves **neurological symptoms** (meningitis, encephalitis, peripheral nerve neuropathy) and **cardiac dysfunction** (heart block, myo-pericarditis, congestive heart failure). These symptoms are seen in 10% to 15% of patients and can last for days to months. The second phase of late disease is characterized by **arthralgias and arthritis**. These complications can persist for months to years, during which spirochetes are only rarely visualized in the involved tissue or isolated in culture.

Treatment, Prevention, and Control

Relapsing fever has been treated most effectively with tetracycline or chloramphenicol. Tetracycline is the drug of choice except for pregnant women and young children, for whom the drug is contraindicated. The early manifestations of Lyme disease are effectively managed with **doxycydine, amoxicillin or erythromycin**. The incidence and severity of late complications are

also ameliorated with therapy. For these manifestations, **ceftriaxone, penicillin, doxycycline, or amoxicillin** have been used.

Prevention of tick-borne borrelia diseases is attempted by **avoiding ticks and their natural habitats, wearing protective clothing**, and using insect repellants. **Rodent control** for endemic relapsing fever is also important. Epidemic louse-borne disease is controlled by the use of delousing sprays and improved hygienic conditions. Vaccines are currently not available for either relapsing fever or Lyme disease.

25.3 Leptospira

Physiology and Structure

The genus *Leptospira* consists of two species: ***Leptospira interrogans*** (subdivided further into 19 serogroups and 172 serotypes) and ***Leptospira biflexa*** (38 serogroups and 65 serotypes). The species names are derived from the fact that *Leptospira* are **thin, coiled bacilli (0.1 x 6 to 20 µm) with a hook at one or both ends ("interrogans" meaning shaped like a question mark; "biflexa" for twice bent). *L. interrogans* is pathogenic for many wild and domestic animals, as well as humans. *L. biflexa* is a free-living saprophyte** found in moist environmental sites and is not associated with disease. About 22 serotypes of *L. interrogans* are responsible for human disease, with serotypes icterohaemorrhagiae, canicola, pomona, and autumnalis the most common.

The pathogenic leptospires are **obligatively aerobic and motile** by means of two periplasmic flagella, each anchored at opposite ends of the bacterium. **The leptospires can be grown on specially formulated media** enriched with rabbit serum or bovine serum albumin.

Pathogenesis and Immunity

L. interrogans can cause **subclinical infection, a mild flu-like febrile illness, or severe systemic disease (Weil's disease), with renal and hepatic failure**, extensive vasculitis, myocarditis, and death. The severity of disease is influenced by the number of infecting organisms, the host's immunological defenses, and the virulence of the infecting strain.

Because leptospires are thin and highly motile, they **can penetrate intact mucous membranes or skin surfaces through small cuts or abrasions**. They then are able **to spread in the bloodstream into all tissues, including the central nervous system**. Multiplication of *L. interrogans* proceeds rapidly and damages the endothelium of small blood vessels, which is responsible for the major clinical manifestations of disease (e.g., meningitis, hepatic and renal dysfunction, and hemorrhage). Organisms can be demonstrated in blood and cerebrospinal fluid early in the course of disease and in urine during the later stages. Clearance of leptospires occurs when humoral immunity develops. However, some clinical manifestations may be related to

immunological reactions with the organisms. For example, meningitis develops after the organisms have been removed from the cerebrospinal fluid and immune complexes have been detected in renal lesions.

Epidemiology

Leptospirosis has worldwide distribution. However, the incidence of disease is significantly underestimated because **most infections are mild and misdiagnosed as a "viral syndrome" or viral aseptic meningitis**.

Many wild and domestic animals are colonized with leptospires, with as many as 10% to 50% of some species infected. **Dogs, cattle, rodents, and wild animals are the most common sources for human disease**. Chronic carriage in humans has not been demonstrated.

Leptospires usually cause asymptomatic infections in their reservoir host, where the spirochetes colonize the renal tubules and are shed in urine in large numbers. **Streams, rivers, standing water, and moist soil can be contaminated with urine from infected animals and serve as a source for human infection, with organisms surviving for as long as 6 weeks**. A moist, alkaline environment is required for survival of leptospires. Most human infections are due to either **recreational exposure** to contaminated water or **occupational exposure** to infected animals (farmers, slaughterhouse workers, veterinarians). Most human infections are reported during the warm months of the year when recreational exposure is greatest. Person-to-person spread has not been documented.

Clinical Syndromes

The majority of infections with *L. interrogans* are clinically inapparent and detected only by demonstration of specific antibodies. **Symptomatic infections develop after a 1- to 2-week incubation period**. The **initial presentation is similar to a flu-like illness, with fever and myalgias**. During this phase the patient is **bacteremic** with the leptospires, and the organisms can frequently be isolated in cerebrospinal fluid even though no meningeal symptoms are present. The **fever and myalgias** may remit after 1 week with no further difficulties, or the patient may progress to more advanced disease – including **aseptic meningitis** – or to a **generalized illness, with headache, rash, vascular collapse, thrombocytopenia, hemorrhage, and hepatic and renal dysfunction (Weil's disease).**

Leptospirosis confined to the central nervous system can be mistaken for viral meningitis because the course of disease is generally uncomplicated with a very low mortality. Culture of the cerebrospinal fluid is usually negative at this stage. In contrast, the icteric form of generalized disease (approximately 10% of all symptomatic infections) is more severe and has a mortality approaching 10%. Although hepatic involvement with jaundice is striking in severe leptospirosis, hepatic necrosis is not seen and surviving patients do not suffer permanent hepatic damage. Similarly, most patients recover full renal function.

Congenital leptospirosis can also occur. The disease is characterized by a sudden onset with headache, fever, myalgias, and a diffuse rash.

Treatment, Prevention, and Control

Leptospirosis is usually not fatal, particularly in the absence of icteric disease. Treatment with either **penicillin or tetracycline** can shorten the clinical symptoms and complications of leptospirosis. **Doxycycline** has also been used both to treat infections and to prevent disease in individuals exposed to infected animals or water contaminated with urine. The total eradication of leptospirosis is difficult because the disease is widespread in wild and domestic animals. However, **vaccination of livestock and pets** has proved successful in reducing disease in these populations and therefore subsequent human exposure. Rodent control is also effective in eliminating leptospirosis in communities.

26. MYCOBACTERIA

Mycobacteria are **aerobic, acid-fast bacilli (rods).** They are **neither gram-positive nor gram-negative**; ie, they are **stained poorly by the dyes used in Gram's stain**. They are virtually the only bacteria that are **acid-fast**. The term "acid-fast" refers to an organism's **ability to retain the carbolfuchsin stain despite subsequent treatment with an ethanol-hydrochloric acid mixture. The high lipid content** (approximately 60%) of their cell wall makes mycobacteria acid-fast.

The major pathogens are ***Mycobacterium tuberculosis*, the cause of tuberculosis**, and ***Mycobacterium leprae*, the cause of leprosy. Atypical mycobacteria**, such as *Mycobacterium avium-intracellulare* complex and *Mycobacterium kansasii*, can cause tuberculosislike disease but are less frequent pathogens. Rapidly growing mycobacteria, such as *Mycobacterium chelonei*, are saprophytes that occasionally cause human disease in immunocompromised hosts (Table 8).

Table 8

Species	Growth on Bacteriologic Media	Preferred Temperature in Vivo (˚C)	Source or Mode of Transmission
M. tuberculosis	Slow (weeks)	37	Respiratory droplets
M. bovis	Slow (weeks)	37	Milk from infected animals
M. leprae	None	32	Prolonged close contact
Atypical mycobacteria (ex. *M. kansasii)*	Slow (weeks)	37	Soil and water
M. marinum	Slow (weeks)	32	Water
M. avium-intracellulare complex	Slow (weeks)	37	Soil and water
M. fortuitum-chelonei complex	Rapid (days)	37	Soil and water

26.1 Mycobacterium tuberculosis

This organism causes tuberculosis.

Important Properties *M. tuberculosis* **grows slowly** (ie, it has a doubling time of 18 hours, in contrast to most bacteria, which can double in number in 1 hour or less). Because growth is so slow, **cultures of clinical specimens must be held for 6-8 weeks before being recorded as negative**. *M. tuberculosis* **can be cultured on bacteriologic media, whereas *M. leprae* cannot.** Media used for its growth (eg, **Lowenstein-Jensen medium**) contain complex nutrients (eg, egg yolk) and dyes (eg, malachite green). The dyes inhibit the unwanted normal flora present in sputum samples.

M. tuberculosis is an **obligate aerobe**; this explains its predilection for causing disease in highly oxygenated tissue such as the upper lobe of the lung and the kidney. Its cell wall contains several complex lipids.

Cord factor (trehalose dimycolate) is correlated with virulence of the organism. Virulent strains grow in a characteristic "serpentine" cordlike pattern, whereas avirulent strains do not. The organism also contains **several proteins**, which, elicit delayed hypersensitivity. These proteins are the antigens in the **PPD (purified protein derivative) skin test.**

***M. tuberculosis* is relatively resistant to acids and alkalis**. NaOH is used to concentrate clinical specimens; it destroys unwanted bacteria, human cells, and mucus but not the organism. *M. tuberculosis* is **resistant to dehydration and so survives in dried expectorated sputum**; this property may be important in its transmission by aerosol.

Strains of *M. tuberculosis* resistant to the main antimycobacterial drug, isoniazid, as well as strains resistant to multiple antibiotics (**called multiple-drug-resistant or MDR strains**), have become a worldwide problem. This resistance is attributed to one or more chromosomal mutations, because no plasmids have been found in this organism.

Transmission & Epidemiology

M. tuberculosis is **transmitted from person to person** by **respiratory aerosol**, and its **initial site of infection is the lung.** In the body, it resides chiefly within reticuloendothelial cells, eg, macrophages. **Humans are the natural reservoir of *M. tuberculosis*; there is no animal reservoir.**

In developed countries, tuberculosis is almost exclusively a human disease. In developing countries, ***Mycobacterium bovis* also causes tuberculosis in humans. *M. bovis* is found in cow's milk, which, unless pasteurized, can cause gastrointestinal tuberculosis in humans**. The disease tuberculosis occurs in only a small proportion of infected individuals. In developed countries, **most tuberculosis is due to reactivation in elderly, malnourished men**. The risk of infection and

disease is highest among socio-economically disadvantaged people, who have poor housing and poor nutrition.

Pathogenesis

M. tuberculosis produces no exotoxins and does not contain endotoxin in its cell wall. In fact, no mycobacteria produce toxins. **Lesions are dependent on the presence of the organism and the host response.** There are two types of lesions:

(1) exudative lesions, which consist of an acute inflammatory response and occur chiefly in the lungs at the initial site of infection; and

(2) granulomatous lesions, which consist of a central area of giant cells containing tubercle bacilli surrounded by a zone of epithelioid cells. A tubercle is a granuloma surrounded by fibrous tissue that has undergone central caseation necrosis. Tubercles heal by fibrosis and calcification.

The primary lesion of tuberculosis usually occurs in the lungs. The parenchymal exudative lesion and the draining lymph nodes together are called a Ghon complex. Primary lesions usually occur in the lower lobes, whereas reactivation lesions usually occur in the apices. Reactivation lesions also occur in other well-oxygenated sites such as the kidneys, brain, and bone. Reactivation is seen primarily in immunocompromised or debilitated patients.

Spread of the organism within the body occurs by two mechanisms:

(1) **A tubercle can erode into a brochus**, empty its caseous contents, and thereby spread the organism to other parts of the lungs, to the gastrointestinal tract if swallowed, and to other persons if expectorated.

(2) **It can disseminate via the bloodstream to many internal organs**. Dissemination can occur at an early stage if cell-mediated immunity fails to contain the initial infection or at a late stage if a person becomes immunocompromised.

Immunity & Hypersensitivity

After recovery from the primary infection, resistance to the organism is mediated by **cellular immunity**, ie, by CD4-positive T cells and macrophages. Circulating antibodies also form, but they play no role in resistance and are not used for diagnostic purposes.

Prior infection can be detected by a positive tuberculin skin test, which is due to a **delayed hypersensitivity reaction**. **PPD is used as the antigen in the tuberculin skin test**. The intermediate-strength preparation of PPD, which contains **5 tuberculin units**, is usually used. **The test is positive if 10 mm of induration occurs 48-72 hours after intradermal injection of the PPD**. Induration (thickening), not simply erythema (reddening), must be observed. **Reactions of 5 to 9 mm in people known to be exposed and who have not received the BCG vaccine indicate that the person is probably infected.** In AIDS patients, a 5-mm reaction is considered positive.

A positive skin test indicates previous infection by the organism but not necessarily active disease. The tuberculin test becomes positive 4-6 weeks after infection. **Immunization with BCG vaccine can cause a positive test** but the reactions are usually only 5 to 10 mm and tend to decrease with the passage of time. PPD reactions of 15 mm or more are assumed to be infected with *M tuberculosis* even if they have received the BCG vaccine. The skin test itself does not induce a positive response. Tuberculin reactivity is mediated by the cellular arm of the immune system; it can be transferred by CD4-positive T cells but not by serum. Infection with measles virus can suppress cell-mediated immunity, resulting in a loss of tuberculin skin test reactivity and, in some instances, reactivation of dormant organisms and clinical disease.

Clinical Findings

Many organs can be involved. Fever**, fatigue, night sweats, and weight loss are common**. Pulmonary tuberculosis causes **cough and hemoptysis**. Scrofula is **mycobacterial cervical adenitis** that presents as swollen nontender lymph nodes, usually unilaterally. Both *M. tuberculosis* and *M. scrofulaceum* cause scrofula. **Miliary tuberculosis is characterized by multiple disseminated lesions that resemble millet seeds**. **Tuberculous meningitis and tuberculous osteomyelitis, especially vertebral osteomyelitis (Pott's disease), are important disseminated forms.**

Laboratory Diagnosis

Acid-fast staining of sputum or other specimens is the usual initial test. For rapid screening purposes, **auramine stain, which can be visualized by fluorescence microscopy**, can be used.

After digestion of the specimen by treatment with NaOH and concentration by centrifugation, the material is **cultured on special media, such as Lowenstein-Jensen agar**, for **up to 8 weeks**. It will not grow on a blood agar plate. In **liquid BACTEC medium**, radioactive metabolites are present and growth can be detected by the production of radioactive carbon dioxide in about 2 weeks. **A liquid medium is preferred for isolation because the organism grows more rapidly and reliably** than it does on agar. If growth in the culture occurs, the organism can be identified by **biochemical tests**. More rapid identification tests using **DNA probes are also available.**

Because drug resistance, especially to isoniazid (see below), is a problem, susceptibility tests should be performed. However, the organism grows very slowly and susceptibility tests usually take several weeks, which is too long to guide the initial choice of drugs. A recently devised test called the luciferase assay, which can detect drug-resistant organisms in a few days, is a distinct improvement. Luciferase is an enzyme isolated from fireflies that produces flashes of light in the presence of ATP. If the organism isolated from the patient is resistant, it will not be damaged by the drug; ie, it will make a normal amount of ATP, and the luciferase will produce the normal amount of light. If the organism is sensitive to the drug, less ATP will be made and less light produced.

Treatment & Resistance

Multiple-drug therapy is used to prevent the emergence of drug-resistant mutants during the long (**6-9 month**) duration of treatment. (Organisms that become resistant to one drug will be inhibited by the other.) **Isoniazid** (isonicotinic acid hydrazide, INH), a bactericidal drug, is the mainstay of treatment. Treatment for most patients with pulmonary tuberculosis is with three drugs: **INH, rifampin, and pyrazinamide**. INH and rifampin are given **for 6 months**, but **pyrazinamide treatment is stopped after 2 months**. In patients who are immunocompromised (eg, AIDS patients), who have disseminated disease, or who are likely to have INH-resistant organisms, a fourth drug, ethambutol, is added and all four drugs are given for 9-12 months.

Prevention

The incidence of tuberculosis began to decrease markedly even before the advent of drug therapy in the 1940s. This is attributed to better housing and nutrition, which have improved host resistance.

The risk of progression to active tuberculosis is reduced by chemoprophylaxis with INH for 6-9 months. It is prescribed for (1) asymptomatic patients whose PPD skin test has recently converted to positive, (2) children exposed to patients with symptomatic pulmonary tuberculosis, and (3) patients with a positive PPD skin test who undergo immunosuppression. Patients receiving INH prophylaxis should be evaluated for drug-induced hepatitis, especially those over age 35 years, in view of the he-patotoxicity of the drug.

A vaccine containing a strain of live, attenuated *M. bovis* (bacillus Calmette-Guerin or BCG) can be used to induce partial resistance to tuberculosis. Pasteurization of milk and destruction of infected cattle are important in preventing intestinal tuberculosis.

26.2 Atypical mycobacteria

Several species of mycobacteria are characterized as atypical, because they differ in certain respects from the prototype, *M. tuberculosis*. For example, **atypical mycobacteria are widespread in the environment** and are not pathogenic for guinea pigs, whereas *M. tuberculosis* is found only in humans and is highly pathogenic for guinea pigs.

The atypical mycobacteria are **classified into four groups according to their rate of growth and whether they produce pigment under certain conditions** (Table 9). Group I organisms produce a yellow-orange-pigmented colony only when exposed to light (photochromogens), whereas group II organisms produce the pigment chiefly in the dark (scotochromogens). Group III mycobacteria produce little or no yellow-orange pigment, irrespective of the presence or absence of light (nonchromogens). In contrast to the organisms

in the previous three groups, which grow slowly, group IV organisms grow rapidly, producing colonies in less than 7 days.

Table 9

Group	Growth rate	Pigment Formation in:		Typical Species
		Light	Dark	
I	Slow	+	-	**M. kansasii, M. marinum**
II	Slow	+	+	***M.scrofulaceum***
III	Slow	-	-	*M. avium-intracellulare* complex
IV	Rapid	-	-	*M. fortuitum-chelonei* complex

26.3 Mycobacterium leprae

This organism causes leprosy.

Important Properties *M. leprae* has not been grown in the laboratory, either on artificial media or in cell culture. Humans are the natural hosts. The optimal temperature for growth (30 °C) is lower than body temperature; it therefore grows preferentially in the skin and superficial nerves.

Transmission: Infection is acquired by **prolonged contact with patients with lepromatous leprosy,** who discharge *M. leprae* in large numbers in nasal secretions and from skin lesions. The disease occurs worldwide, with most cases in the tropical areas of Asia and Africa.

Pathogenesis The organism replicates intracellularly typically within skin histiocytes, endothelial cells, and the Schwann cells of nerves. There are **two distinct forms of leprosy – tuberculoid and lepromatous** – with several intermediate forms between the two extremes (Table 10).

Table 10

Feature	Tuberculoid Leprosy	Lepromatous Leprosy
Type of Lesion	One or few lesions with little tissue destruction	Many lesions with marked tissue destruction
Number of Acid-Fast Bacilli	Few	Many
Likelihood of Transmitting Leprosy	Low	High
Cell-mediated Response to *M. leprae*	Present	Reduced or absent
Lepromin Skin Test	Positive	Negative

(1) In tuberculoid leprosy, the cell-mediated immune response to the organism limits its growth, very few acid-fast bacilli are seen, granulomas containing giant cells form, and the lepromin skin test is positive. The lepromin skin test is similar to the tuberculin test (see above). An extract of *M. leprae* is injected intradermally, and induration is observed 48 hours later in those in whom a cell-mediated immune response against the organism exists.

(2) In lepromatous leprosy, the cell-mediated response to the organism is poor, the skin and mucous membrane lesions contain large numbers of organisms, foamy histiocytes rather than granulomas are found, and the lepromin skin test is negative. Note that in lepromatous leprosy only the cell-mediated response to *M. leprae* is defective; ie, the patient is anergic to *M. leprae*. The cell-mediated response to other organisms is unaffected and the humoral response to *M. leprae* is intact. However, these antibodies are not protective.

Clinical Findings The **incubation period averages several years**, and the onset of the disease is gradual. **In tuberculoid leprosy, hypopigmented macular skin lesions, thickened superficial nerves, and significant anesthesia of the skin lesions occur**. **In lepromatous leprosy, multiple nodular skin lesions occur, resulting in the typical leonine (lionlike) fades.**

The disfiguring appearance of the disease is due to several factors:

(1) the skin anesthesia results in bums and other traumas, which often become infected;
(2) resorption of bone leads to loss of features such as the nose and fingertips;
(3) infiltration of the skin and nerves leads to thickening and folding of the skin. In the majority of patients with a single skin lesion, the disease resolves spontaneously.

Patients with forms of the disease intermediate between tuberculoid and lepromatous can progress to either extreme.

Laboratory Diagnosis In lepromatous leprosy, the bacilli are easily demonstrated by performing an **acid-fast stain of skin lesions or nasal scrapings**. In the tuberculoid form, very few organisms are seen and the appearance of typical granulomas is sufficient for diagnosis. No serologic tests are useful. False-positive serologic tests for syphilis occur frequently.

Treatment The mainstay of therapy is **dapsone** (diaminodiphenylsulfone), but sufficient resistance to the drug has emerged that combination therapy is now recommended, eg, dapsone, rifampin, and clofazimine for lepromatous leprosy and dapsone and rifampin for the tuberculoid form. Treatment is given for at least 2 years or until the lesions are free of organisms.

Prevention Isolation of all lepromatous patients, coupled with chemoprophylaxis with dapsone for exposed children, is required.

27. MYCOPLASMAS

Mycoplasmas are a group of small, wall-less organisms, of which *Mycoplasma pneumoniae* is the major pathogen.

27.1 Mycoplasma pneumoniae

M. pneumoniae causes "atypical" pneumonia. Mycoplasmas are the smallest free-living organisms; many are as small as 0.3 μm in diameter. Their most striking feature is the absence of a cell wall. Consequently, mycoplasmas stain poorly with Gram's stain, and antibiotics that inhibit cell wall synthesis, eg, penicillins and cephalosporins, are ineffective. Their outer surface is a flexible three-layer cell membrane; hence, these organisms can assume a variety of shapes.

Mycoplasmas **can be grown in the laboratory on artificial media, but they have complex nutritional requirements,** including several lipids. They grow slowly and require at least 1 week to form a visible colony. **The colony frequently has a characteristic "fried-egg"** shape, with a raised center and a thinner outer edge.

Pathogenesis & Epidemiology

M. pneumoniae, a **pathogen only for humans, is transmitted by respiratory droplets.**

M. pneumoniae has only one serotype and is antigenically distinct from other species of Mycoplasma. Immunity is incomplete, and second episodes of disease can occur. During *M. pneumoniae* infection, **autoantibodies are produced against red cells** (cold agglutinins) and brain, lung, and liver cells. These antibodies may be the source of the extrapulmonary manifestations of infection.

M. pneumoniae infections **occur worldwide, with an increased incidence in the winter.** This organism is the most **frequent cause of pneumonia in young adults and is responsible for outbreaks in groups with close contacts such as families, military personnel, and college students**. It is estimated that only 10% of infected individuals actually get pneumonia. *Mycoplasma pneumonia* accounts for about 5-10% of all community-acquired pneumonia.

Clinical Findings

Mycoplasma pneumonia is the most common type of atypical pneumonia. It was formerly called primary atypical pneumonia. (The term "atypical" means that a causative bacterium cannot be isolated on routine media in the diagnostic laboratory or that the disease does not resemble pneumococcal pneumonia.) The onset of *Mycoplasma pneumonia* is gradual, usually **beginning with a nonproductive cough, sore throat, or ear ache**. Small amounts of **whitish,**

nonbloody sputum are produced. Constitutional symptoms of **fever, headache, malaise, and myalgias** are pronounced. The paucity of findings on chest examination is in marked contrast to the prominence of the chest X-ray infiltrates. The disease resolves spontaneously in 10-14 days.

Laboratory Diagnosis

Serologic testing is the mainstay of diagnosis. **A cold-agglutinin titer of 1: 128 or higher is indicative of recent infection.** Cold agglutinins are IgM autoantibodies against type 0 red blood cells that agglutinate these cells at 4 °C but not at 37 °C. However, only half of patients with *Mycoplasma pneumonia* will be positive for cold agglutinins. The test is nonspecific; false-positive results occur in influenza virus and adenovirus infections. The diagnosis of *M. pneumoniae* infection can be confirmed by a 4-fold or greater rise in specific antibody titer in the complement fixation test.

Treatment

The treatment of choice is either **erythromycin or tetracycline**. These drugs can shorten the duration of symptoms, although, as mentioned above, the disease resolves spontaneously. Penicillins and cephalosporins are inactive because the organism has no cell wall.

27.2 Other mycoplasmas

Mycoplasma hominis has been implicated as an **infrequent cause of pelvic inflammatory disease.** ***Ureaplasma urealyticum*** may cause approximately 20% of cases of **nongonococcal urethritis**. Ureaplasmas can be distinguished from mycoplasmas by their ability to produce the enzyme urease, which degrades urea to ammonia and carbon dioxide.

28. CHLAMYDIAE

Chlamydiae are **obligate intracellular parasites**. They are the **agents of psittacosis, trachoma, lymphogranuloma venereum**, and other infections.

Chlamydia psittaci* causes psittacosis**; ***Chlamydia trachomatis causes eye, respiratory, and genital tract infections. *C. trachomatis* is the most common cause of sexually transmitted disease in the United States. *Chlamydia pneumoniae* (formerly called the TWAR strain) causes atypical pneumonia (Table 11).

Table 11

Medically Important Species	Disease	Natural Hosts	Mode of Transmission to Humans	Number of Immunologic Types	Diagnosis	Treatment
C. psittaci	Psittacosis (pneumonia)	Birds	Inhalation of dried bird feces	1	Serologic test (cell culture rarely done)	Tetracycline
C. trachomatis	Urethritis, pneumonia, conjunctivitis lymphogranu-loma venereum, trachoma	Humans	Sexual contact; perinatal transmission	More than 15	Inclusions in epithelial cells seen with Giemsa's stain or by immunofluorescence; also cell culture	Tetracycline erythromycin
C. pneumoniae	Atypical pneumonia	Humans	Respiratory droplets	1	Serologic test	Tetracycline

Chlamydiae are **obligate intracellular bacteria**. They **lack the ability to produce sufficient energy to grow independently and therefore can grow only inside host cells**. They **have a rigid cell wall but do not have a typical peptidoglycan layer. Their cell walls resemble those of gram-negative bacteria but lack muramic acid.**

Chlamydiae have a **replicative cycle different from that of all other bacteria**. The cycle begins when the extracellular, metabolically inert, "sporelike," **elementary body** enters the cell and reorganizes into a larger, metabolically active reticulate body. The latter undergoes repeated binary fission to form daughter elementary bodies, which are released from the cell. Within cells, the site of replication appears as an inclusion body, which can be stained and visualized microscopically. These inclusions are useful in the diagnosis of these organisms in the clinical laboratory.

All chlamydiae share a group-specific lipopolysaccharide antigen, which is detected by complement fixation tests. They also possess species-specific and immunotype-specific antigens (proteins), which are detected by immunofluorescence. *C. psittaci* and *C. pneumoniae* each have one immunotype, whereas *C. trachomatis* has at least 15.

Transmission & Epidemiology

***C. psittaci* infects birds and many mammals.** Humans are infected primarily by **inhaling organisms in dry bird feces**. ***C. trachomatis* infects only humans and is usually transmitted by close personal contact**, eg, sexually or by passage through the birth canal. Individuals with asymptomatic genital tract infections are an important reservoir of infection for others. In trachoma, ***C. trachomatis* is transmitted by finger-to-eye or fomite-to-eye contact**. ***C. pneumoniae* infects only humans and is transmitted from person to person by aerosol.**

Disease caused by these organisms occurs worldwide, but trachoma is most frequently found in developing countries in dry, hot regions. Nongonococcal urethritis caused by *C. trachomatis* is said to occur more frequently in higher socioeconomic groups, in contrast to gonorrhea, which is found predominantly in lower socioeconomic groups. However, the two diseases commonly occur simultaneously in the same individual.

Pathogenesis & Clinical Findings

Chlamydiae **infect primarily epithelial cells of the mucous membranes or the lungs.** They **rarely cause invasive, disseminated infections**.

***C. psittaci* infects the lungs primarily**. The infection may be asymptomatic (detected only by a rising antibody titer) or may produce high fever and pneumonia. Human psittacosis is not generally communicable.

C. pneumoniae causes upper and lower respiratory tract infections, especially bronchitis and pneumonia, in young adults.

C. trachomatis exists in more than **15 immunotypes (A-L).** Types **A, B, and C cause trachoma, a chronic conjunctivitis endemic** in Africa and Asia. Trachoma may recur over many years and may lead to blindness but causes no systemic illness. Types **D-K cause genital tract infections,** which are **occasionally transmitted to the eyes or the respiratory tract**. In men, it is a common cause of **non-gonococcal urethritis**, which may progress to epididymitis, prostatitis,

or proctitis. In women, **cervicitis** develops and may progress to salpingitis and pelvic inflammatory disease (PID). Repeated episodes of salpingitis or PID can result in infertility or ectopic pregnancy. Infants born to infected mothers often develop mucopurulent eye infections (neonatal inclusion conjunctivitis) 7-12 days after delivery, and some develop chlamydial pneumonitis 2-12 weeks after birth. Patients with genital tract infections caused by *C. trachomatis* have a high incidence of Reiter's syndrome, which is characterized by urethritis, arthritis, and uveitis. Reiter's syndrome is an autoimmune disease caused by antibodies formed against *C. trachomatis* cross-reacting with antigens on the cells of the urethra, joints, and uveal tract.

***C. trachomatis* L1-L3 immunotypes cause lymphogranuloma venereum**, a sexually transmitted disease with lesions on genitalia and in lymph nodes.

Infection by *C. trachomatis* leads to formation of antibodies and cell-mediated reactions but not to resistance to reinfection or elimination of organisms.

Laboratory Diagnosis

Chlamydiae form cytoplasmic inclusions, which can be seen with **special stains (eg, Giemsa's stain) or by immunofluorescence**. The Gram stain is not useful. In exudates, the organism can be identified within epithelial cells by **fluorescent antibody staining or hybridization with a DNA probe.** Chlamydial antigens can also be detected in exudates or urine by **ELISA.**

Serologic tests are used to diagnose infections by *C. psittaci* and *C. pneumoniae*, but are rarely helpful in diagnosing disease caused by *C. trachomatis* because the frequency of infection is so high that many people already have antibodies.

Treatment

All chlamydiae are **susceptible to tetracyclines and erythromycin**. Treatment suppresses signs and symptoms but does not regularly eradicate the organisms. *C. trachomatis* is susceptible to either tetracycline or erythromycin. The treatment of choice for *C. psittaci* and *C. pneumoniae* infections is tetracycline. The drug of choice for lymphogranuloma venereum is a tetracycline. The treatment of choice for *C. trachomatis* sexually transmitted diseases is azithromycin.

Prevention

Psittacosis in humans is controlled by **restricting the importation of psittacine birds, destroying sick birds, and adding tetracycline to bird feed.** Domestic flocks of turkeys and ducks are surveyed for the presence of *C. psittaci*.

C. trachomatis infection in humans should be diagnosed and treated early both in clinically manifest cases and in asymptomatic sexual contacts. Several types of sexually transmitted diseases are often present simultaneously. Thus, diagnosis of one requires a search for other etiologic

agents. Erythromycin given to newborn infants of infected mothers can prevent inclusion conjunctivitis and pneumonitis. There is no vaccine available against any chlamydial disease.

29. RICKETTSIAE

Rickettsiae are **obligate intracellular parasites**. They are the **agents of typhus, spotted fevers, and Q fever.** In the United States, there are two rickettsial diseases of significance: Rocky Mountain spotted fever, caused by *Rickettsia rickettsii*, and Q fever, caused by *Coxiella burnetii*. Several other rickettsial diseases such as epidemic, endemic, and scrub typhus are important in developing countries. Rickettsialpox, caused by *Rickettsia akari*, is a rare disease found in certain densely populated cities in the United States.

Rickettsiae are very short rods that are **barely visible in the light microscope**. Structurally, their cell wall resembles that of **gram-negative rods**, but **they stain poorly with the standard Gram stain. Rickettsiae are obligate intracellular parasites** because they are unable to produce sufficient energy to replicate extracellularly. Therefore, rickettsiae must be **grown in cell culture, embryonated eggs, or experimental animals**. Rickettsiae divide by binary fission within the host cell, in contrast to chlamydiae, which are also obligate intracellular parasites but replicate by a distinctive intracellular cycle.

Several rickettsiae, such as *Rickettsia prowazekii*, *Rickettsia tsutsugamushi*, and *R. rickettsii*, **possess antigens that cross-react with antigens of the OX strains of *Proteus vulgaris*.** The **Weil-Felix test,** which detects antirickettsial antibodies in a patient's serum by agglutination of the *Proteus* organisms, is based on this cross-reaction.

Transmission

The most striking aspect of the life cycle of the rickettsiae is that **they are maintained in nature in certain arthropods such as ticks, lice, fleas, and mites and, with one exception, are transmitted to humans by the bite of the arthropod**. The exception to arthropod transmission is ***C. burnetii*, the cause of Q fever, which is transmitted by aerosol and inhaled into the lungs**. Virtually **all rickettsial diseases are zoonoses** (ie, they have an animal reservoir), with the prominent exception of epidemic typhus, which occurs only in humans. A summary of the vectors and reservoirs for selected rickettsial diseases is presented in Table 12.

The incidence of the disease depends on the geographic distribution of the arthropod vector and on the risk of exposure, which is enhanced by such things as poor hygienic conditions and camping in wooded areas. These factors are discussed below with the individual diseases.

Pathogenesis

The typical lesion caused by the rickettsiae is a **vasculitis, particularly in the endothelial lining of the vessel wall where the organism is found.** Damage to the vessels of the skin results in the **characteristic rash and in edema and hemorrhage due to increased capillary permeability.** The basis for pathogenesis by these organisms is unclear. There is some evidence that endotoxin is involved, which is in accord with the nature of some of the lesions such as fever

and petechiae, but its role has not been confirmed. No exotoxins or cytolytic enzymes have been found.

Table 12

Disease	Organism	Arthropod Vector	Mammalian Reservoir
Spotted fevers Rocky Mountain spotted fever	*R. rickettsii*	Tikcs	Dogs, rodents
Rickettsialpox	*R. akarii*	Mites	Mice
Typhus group Epidemic	*R. prowazekii*	Lice	Humans
Endemic	*R. typhi*	Fleas	Rodents
Scrub	*R. tsutsugamushi*	Mites	Rodents
Others Q fever	*C. burnetii*	None	Cattle, sheep, goats

Clinical Findings & Epidemiology

A. Rocky Mountain Spotted Fever*:* This disease is characterized by the acute onset of nonspecific symptoms, eg, fever, severe headache, myalgias, and prostration. The typical rash, which appears 2-6 days later, begins with macules that frequently progress to petechiae. The rash usually appears first on the hands and feet and then moves inward to the trunk. In addition to headache, other profound central nervous system changes such as delirium and coma can occur. Disseminated intravascular coagulation, edema, and circulatory collapse may ensue in severe cases. The diagnosis must be made on clinical grounds and therapy started promptly, because the laboratory diagnosis is delayed until a rise in antibody titer can be observed.

The name "Rocky Mountain spotted fever" is derived from the region in which the disease was first found. The tick is an important reservoir of *R. rickettsii* as well as the vector; the organism is passed by the transovarian route from tick to tick, and a lifetime infection results. Certain mammals, such as dogs and rodents, are also reservoirs of the organism. Humans are accidental hosts and are not required for the perpetuation of the organism in nature; there is no person-to-person transmission. Most cases occur in children during spring and early summer, when the ticks are active. Rocky Mountain spotted fever accounts for 95% of the rickettsial disease in the United States; there are about 1000 cases per year. It can be fatal if untreated, but if it is diagnosed and treated, a prompt cure results.

B. Q Fever: Unlike the other rickettsial diseases, the main focus of disease in Q fever is in the lungs. It begins suddenly with fever, severe headache, and influenzalike symptoms. This is all

that occurs in many patients, but pneumonia ensues in about half. Hepatitis is frequent enough that the combination of pneumonia and hepatitis should suggest Q fever. A rash is rare, unlike in the other rickettsial diseases. In general, Q fever is an acute disease and recovery is expected even in the absence of antibiotic therapy. Rarely, chronic Q fever characterized by a life-threatening endocarditis occurs.

Q fever is the one rickettsial disease that is not transmitted to humans by the bite of an arthropod. The important **reservoirs for human infection are cattle, sheep, and goats**. The agent, *C. burnetii*, which causes an inapparent infection in these reservoir hosts, is found in high concentrations in the urine, feces, placental tissue, and amniotic fluid of the animals. It is transmitted to humans by inhalation of aerosols of these materials. The disease occurs worldwide, chiefly in individuals whose occupations expose them to livestock, such as shepherds, abattoir employees, and farm workers. Cows' milk is usually responsible for subclinical infections rather than disease in humans. Pasteurization of milk kills the organism.

***C. Typhus**:* There are several different forms of typhus, namely louse-bome epidemic typhus caused by *R. prowazekii*, flea-borne endemic typhus caused by *Rickettsia typhi*, chigger-borne scrub typhus caused by *R. tsutsugamushi*, and several other quite rare forms. The following description is limited to epidemic typhus, the most important of the typhus group of diseases.

Typhus begins with the **sudden onset of chills, fever, headache, and other influenzalike symptoms** approximately 1-3 weeks after the louse bite occurs. Between the fifth and ninth days after the onset of symptoms, a maculopapular rash begins on the trunk and spreads peripherally. **Signs of severe meningoencephalitis, including delirium and coma, begin with the rash and continue into the second and third weeks.** In untreated cases, death occurs from peripheral vascular collapse or from the most frequent complication, bacterial pneumonia.

Epidemic typhus is transmitted from person to person by the human body louse, *Pediculus*. When a bacteremic patient is bitten, **the organism is ingested by the louse and multiplies in the gut epithelium**. It is **excreted in the feces** of the louse during the act of biting the next person and **autoinoculated by the person while scratching the bite.** The infected louse dies after a few weeks, and there is no louse-to-louse transmission, so that human infection is an obligatory stage in the cycle. Epidemic typhus is associated with wars and poverty; at present it is found in developing countries in Africa and South America but not in the United States.

A recurrent form of epidemic typhus is called **Brill-Zinsser disease.** The signs and symptoms are similar to those of epidemic typhus but are less severe, of shorter duration, and rarely fatal. Recurrences can appear as long as 50 years later and can be precipitated by another intercurrent disease. In the United States, the disease is seen in older people who had epidemic typhus during World War II in Europe. Brill-Zinsser disease is epidemiologically interesting; persistently infected patients can serve as a source of the organism should a louse bite occur.

Laboratory Diagnosis

Laboratory diagnosis of rickettsial diseases is based on **serologic analysis** rather than isolation of the organism. Although rickettsiae can be grown in cell culture or embryonated eggs, this is a hazardous procedure that is not available in the standard clinical laboratory.

Of the two serologic tests, **complement fixation is more frequently used and provides more specific data than the Weil-Felix reaction**. (A microagglutination test is used by public health laboratories but is not widely available.) As usual, a 4-fold rise in antibody titer found in a convalescent-phase serum sample is significant. However, in the presence of the clinical picture of Rocky Mountain spotted fever, a single acute-phase serum titer of 1:16 or greater is diagnostic.

The Weil-Felix test is based on the cross-reaction of an antigen present in many rickettsiae with the O antigen polysaccharide found in *P. vulgaris* OX-2, OX-19, and OX-K. The test measures the **presence of antirickettsial antibodies in the patient's serum by their ability to agglutinate *Proteus* bacteria.** The specific rickettsial organism can be identified by the agglutination observed with one or another of these three different strains of *P. vulgaris*. However, since there can be false-positive reactions following *Proteus* urinary tract infections and false-negative reactions due to variable antibody responses, and since no Weil-Felix agglutinins are made in Q fever, the test is of limited value.

Treatment: The treatment of choice for all rickettsial diseases is **tetracycline, with chloramphenicol as the second choice.**

Prevention

Prevention of many of these diseases is based on **reducing exposure to the arthropod vector by wearing protective clothing and using insect repellent**. Frequent examination of the skin for ticks is important in preventing Rocky Mountain spotted fever; the tick must be attached for several hours to transmit the disease. Prevention of typhus is based on personal hygiene and "delousing" with DDT. A typhus vaccine containing formalin-killed *R. prowazekii* organism is effective and useful in the military during wartime but is not available to civilians. **Persons at high risk of contacting Q fever, such as veterinarians, shepherds, abbatoir workers, and laboratory personnel exposed to *C. burnetii*, should receive the vaccine that consists of killed organisms.**