CHAPTER 21

Mycobacteria

CHAPTER CONTENTS

Introduction
Mycobacterium tuberculosis
Atypical Mycobacteria
Mycobacterium leprae
Self-Assessment Questions
Summaries of Organisms
Practice Questions: USMLE & Course Examinations

INTRODUCTION

Mycobacteria are aerobic, **acid-fast** bacilli (rods) (Figure 21–1). They are neither gram-positive nor gram-negative (i.e., they are stained poorly by the dyes used in Gram stain). They are virtually the only bacteria that are acid-fast. (One exception is *Nocardia asteroides*, the major cause of nocardiosis, which is also 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 tuberculosis-like disease but are less frequent pathogens. Rapidly growing mycobacteria, such as *Mycobacterium chelonae*, occasionally cause human disease in immunocompromised patients or those in whom prosthetic devices have been implanted (Table 21–1). The clinical features of three important mycobacteria are described in Table 21–2.

Additional information regarding the clinical aspects of infections caused by the organisms in this chapter is provided in Part IX entitled Infectious Diseases beginning on page 593.

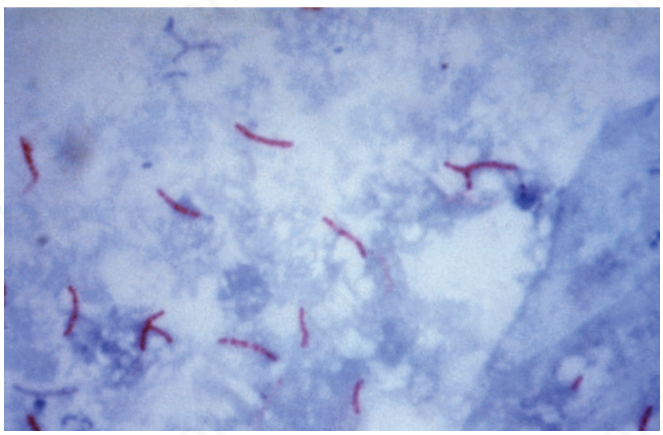

FIGURE 21–1 *Mycobacterium tuberculosis*—acid-fast stain. Long red rods of *M. tuberculosis* are seen on a blue background. (Source: Dr. George Kubica, Public Health Image Library, Centers for Disease Control and Prevention.)

MYCOBACTERIUM TUBERCULOSIS

Disease

This organism causes tuberculosis. Worldwide, *M. tuberculosis* causes more deaths than any other single microbial agent. Approximately one-third of the world's population is infected with this organism. Each year, it is estimated that 1.7 million people die of tuberculosis and that 9 million new cases occur. An estimated 500,000 people are infected with a multidrug-resistant strain of *M. tuberculosis*.

Important Properties

Mycobacterium tuberculosis **grows slowly** (i.e., 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 to 8 weeks before being recorded as negative. *Mycobacterium tuberculosis* can be cultured on bacteriologic media,

TABLE 21–1 Medically Important Mycobacteria

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[1]	Slow (weeks)	37	Soil and water
M. kansasii			
M. marinum	Slow (weeks)	32	Water
M. avium–intracellulare complex	Slow (weeks)	37	Soil and water
M. fortuitum–chelonae complex	Rapid (days)	37	Soil and water

[1]Only representative examples are given.

whereas *M. leprae* cannot. Media used for its growth (e.g., Löwenstein-Jensen medium) contain complex nutrients (e.g., egg yolk) and dyes (e.g., malachite green). The dyes inhibit the unwanted normal flora present in sputum samples.

Mycobacterium tuberculosis is an **obligate aerobe**; this explains its predilection for causing disease in highly oxygenated tissues such as the upper lobe of the lung and the kidney. The acid-fast property of *M. tuberculosis* (and other mycobacteria) is attributed to long-chain (**C78–C90**) fatty acids called **mycolic acids** in the cell wall.

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, when combined with waxes, elicit delayed hypersensitivity. These proteins are the antigens in the **purified protein derivative (PPD)** skin test (also known as the tuberculin skin test). A lipid located in the bacterial cell wall called phthiocerol dimycocerosate is required for pathogenesis in the lung.

Mycobacterium 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 therefore 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 (**isonicotinic acid hydrazide, INH**), as well as strains resistant to multiple antibiotics (called **multidrug-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. One of these mutations is in a gene for mycolic acid synthesis, and another is in a gene for catalase-peroxidase, an enzyme required to activate INH within the bacterium.

Transmission & Epidemiology

Mycobacterium tuberculosis is transmitted from person to person by respiratory aerosols produced by coughing. The source of the organism is a cavity in the lung that has eroded into a bronchus. The portal of entry is the respiratory tract and the initial site of infection is the lung. In tissue, it resides chiefly within reticuloendothelial cells (e.g., **macrophages**). Macrophages kill most, but not all, of the infecting organisms. The ones that survive can continue to infect other adjacent cells or can disseminate to other organs.

Humans are the natural reservoir of *M. tuberculosis*. Although some animals, such as cattle, can be infected, they are not the main reservoir for human infection. Most transmission occurs by aerosols generated by the coughing of "smear-positive" people (i.e., those whose sputum contains detectable bacilli in the acid-fast stain). However, about 20% of people are infected by aerosols produced by the coughing of "smear-negative" people.

In the United States, tuberculosis is almost exclusively a human disease. In developing countries, *Mycobacterium*

TABLE 21–2 Clinical Features of Important Mycobacteria

Organism	Main Site of Infection	Skin Test in Common Use	Multiple-Drug Therapy Used	Vaccine Available
M. tuberculosis	Lungs	Yes	Yes	Yes
M. avium–intracellulare	Lungs	No	Yes	No
M. leprae	Skin, nerves	No	Yes	No

bovis also causes tuberculosis in humans. *Mycobacterium bovis* is found in cow's milk, which, unless pasteurized, can cause gastrointestinal tuberculosis in humans. The disease tuberculosis occurs in only a small number of infected individuals. In the United States, most cases of tuberculosis are associated with reactivation in elderly, malnourished men. The risk of infection and disease is highest among socioeconomically disadvantaged people, who have poor housing and poor nutrition. These factors, rather than genetic ones, probably account for the high rate of infection among Native Americans, African Americans, and Native Alaskans.

Pathogenesis

An overall scheme of pathogenesis by *M. tuberculosis* is shown in Figure 21–2. It describes primary tuberculosis which typically results in a Ghon focus in the lower lung. Primary tuberculosis can heal by fibrosis, can lead to progressive lung disease, can cause bacteremia and miliary

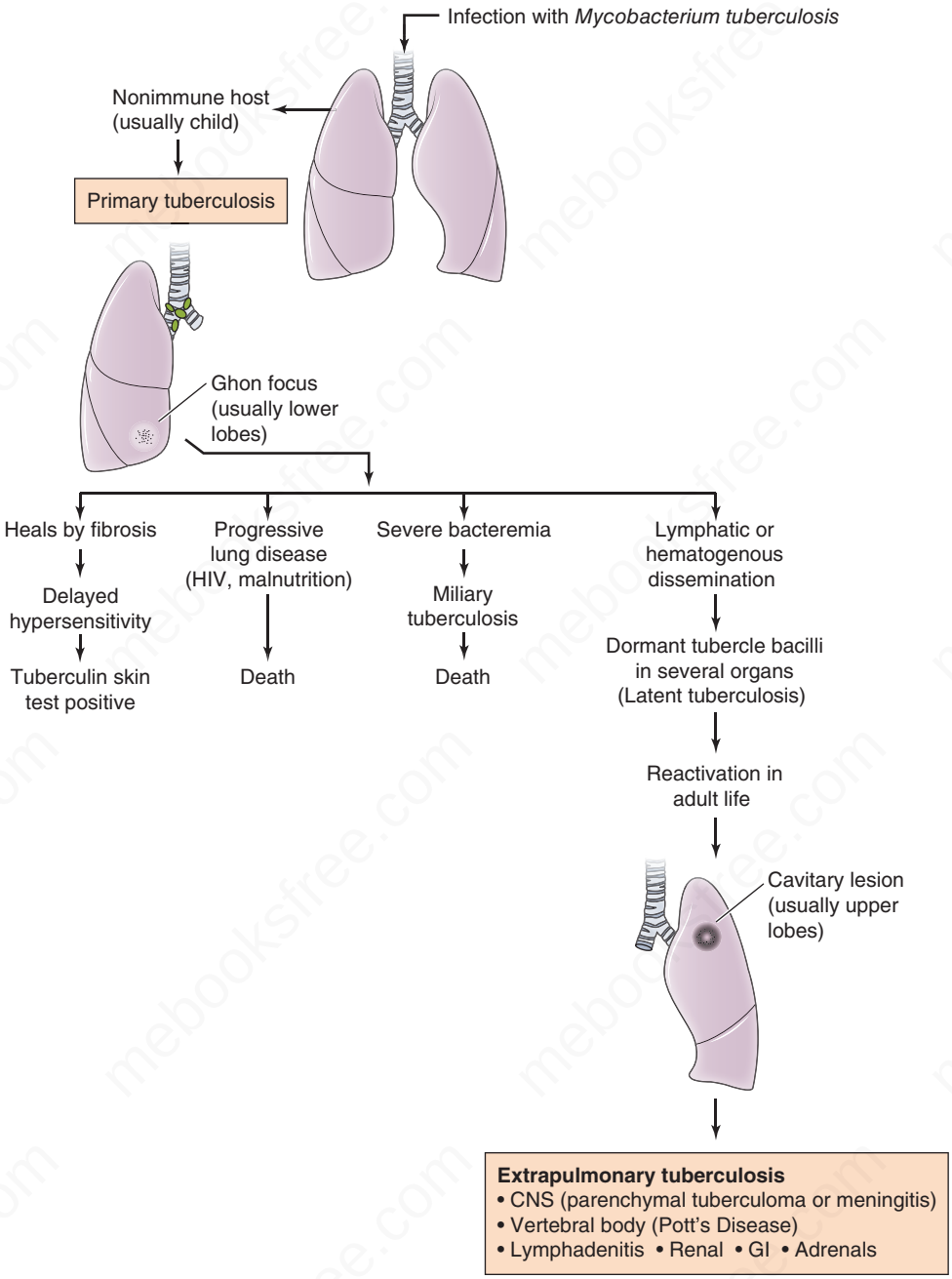

FIGURE 21–2 Pathogenesis by *Mycobacterium tuberculosis*. (Reproduced with permission from Le T, Bhushan V, and Sochat, M. *First Aid for the USMLE Step 1*. 25th ed. New York: McGraw-Hill, 2015.)

tuberculosis, or cause hematogenous dissemination resulting in no immediate disease but with the risk of reactivation in later life.

Figure 21–2 also describes secondary tuberculosis with a cavity in the upper lobes. This can cause disease directly or result in reactivation disease in later life with central nervous system lesions, vertebral osteomyelitis (Pott's disease), or involvement of other organs.

Mycobacterium tuberculosis produces no exotoxins and does not contain endotoxin in its cell wall. In fact, no mycobacteria produce toxins. The organism preferentially infects macrophages and other reticuloendothelial cells. *Mycobacterium tuberculosis* survives and multiplies within a cellular vacuole called a phagosome. The organism produces a protein called "exported repetitive protein" that prevents the phagosome from fusing with the lysosome, thereby allowing the organism to escape the degradative enzymes in the lysosome.

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.

(2) **Granulomatous lesions,** which consist of a central area of giant cells containing tubercle bacilli surrounded by a zone of epithelioid cells. These giant cells, called **Langhans' giant cells,** are an important pathologic finding in tuberculous lesions. 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 bronchus, 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** (i.e., by CD4-positive T cells and macrophages). The CD4-positive T cells are Th-1 helper T cells (see Chapter 58).

Circulating antibodies also form, but they play no role in resistance and are not used for diagnostic purposes. Patients deficient in cellular immunity, such as patients with acquired immunodeficiency syndrome (AIDS), are at much higher risk for disseminated, life-threatening tuberculosis. Mutations in the interferon-γ receptor gene are another cause of defective cellular immunity that predisposes to severe tuberculosis. This emphasizes the importance of activation of macrophages by interferon-γ in the host defense against *M. tuberculosis*.

Prior infection can be detected by a positive tuberculin skin test result, 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 five tuberculin units, is usually used. The skin test is evaluated by measuring the diameter of the **induration** surrounding the skin test site (Figure 21–3). Note that induration (thickening), not simply erythema (reddening), must be observed.

The diameter required to judge the test as positive varies depending on the status of the individual being tested. Induration of 15 mm or more is positive in a person who has no known risk factors. Induration of 10 mm or more is positive in a person with high-risk factors, such as a homeless person, intravenous drug users, or nursing home residents. Induration of 5 mm or more is positive in a person who has deficient cell-mediated immunity (e.g., AIDS patients) or has been in close contact with a person with active tuberculosis.

A positive skin test result indicates previous infection by the organism but not necessarily active disease. The tuberculin test becomes positive 4 to 6 weeks after infection. Immunization with bacillus Calmette-Guérin (BCG) vaccine (see page 189) can cause a positive test, but the reactions are usually only 5 to 10 mm and tend to decrease with

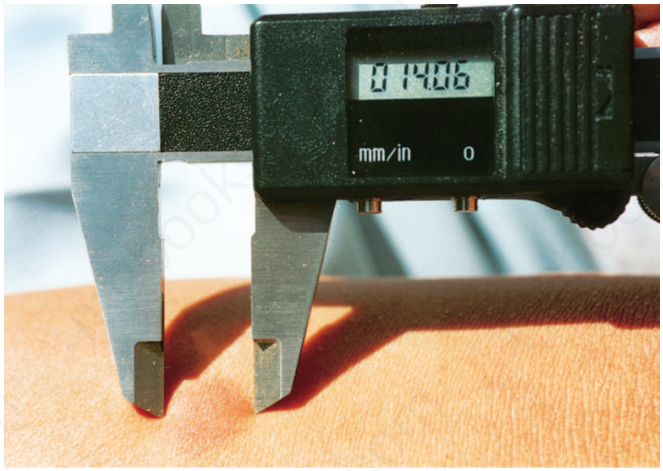

FIGURE 21–3 Tuberculin skin test. Purified protein derivative (PPD) was injected intradermally, and 48 hours later, the diameter of induration was measured with a caliper. (Reproduced with permission from Talaro KP. *Foundations in Microbiology*. 8th ed. New York: McGraw-Hill, 2011.)

time. People with PPD reactions of 15 mm or more are assumed to be infected with *M. tuberculosis* even if they have received the BCG vaccine. A positive skin test reverts to negative in about 5% to 10% of people. Reversion to negative is more common in the United States now than many years ago because now a person is less likely to be exposed to the organism and therefore less likely to receive a boost to the immune system.

The skin test itself does *not* induce a positive response in a person who has not been exposed to the organism. It can, however, "boost" a weak or negative response in a person who has been exposed to produce a positive reaction. The clinical implications of this "booster effect" are beyond the scope of this book.

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.

A gene called *Nramp* determines natural resistance to tuberculosis. People who have mutations in the *Nramp* gene have a much higher rate of clinical tuberculosis than those with a normal allele. The NRAMP protein is located in the membrane of the phagosome in macrophages and plays an important role in killing the organism within the phagosome.

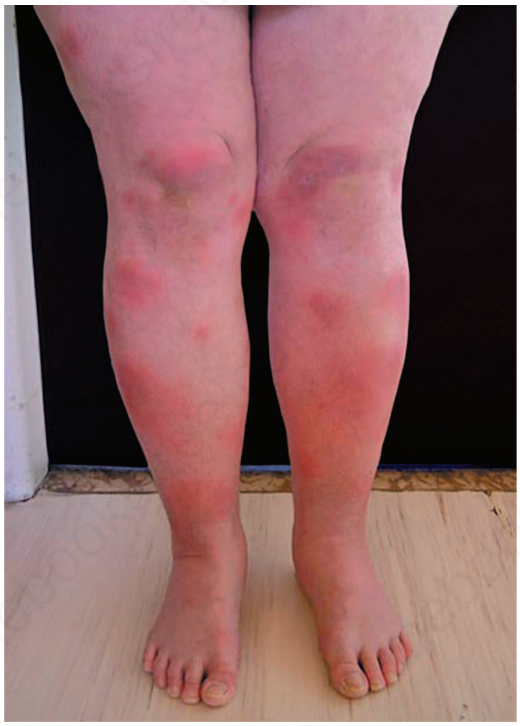

FIGURE 21–4 Erythema nodosum. Note erythematous nodules over the anterior surface of the tibia bilaterally. (Used with permission from Dr. Hanus Rozsypal.)

Clinical Findings

Clinical findings are protean; many organs can be involved. Fever, fatigue, night sweats, and weight loss are common. The main findings in pulmonary tuberculosis are cough and hemoptysis.

Scrofula is mycobacterial cervical lymphadenitis that presents as swollen, nontender lymph nodes, usually unilaterally. *Mycobacterium tuberculosis* causes most cases of scrofula, but nontuberculous *Mycobacteria*, such as *Mycobacterium scrofulaceum*, can also cause scrofula. Lymphadenitis is the most common extrapulmonary manifestation of tuberculosis. Patients infected with human immunodeficiency virus (HIV) are more likely to have multifocal lymphadenitis than those not infected with HIV.

Erythema nodosum, characterized by tender nodules along the extensor surfaces of the tibia and ulna, is a manifestation of primary infection seen in patients who are controlling the infection with a potent cell-mediated response (Figure 21–4). **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.

Gastrointestinal tuberculosis is characterized by abdominal pain and diarrhea accompanied by more generalized symptoms of fever and weight loss. Intestinal obstruction or hemorrhage may occur. The ileocecal region is the site most often involved. Tuberculosis of the gastrointestinal tract can be caused by either *M. tuberculosis* when it is swallowed after being coughed up from a lung lesion or by *M. bovis* when it is ingested in unpasteurized milk products. **Oropharyngeal tuberculosis** typically presents as a painless ulcer accompanied by local adenopathy.

In **renal tuberculosis**, dysuria, hematuria, and flank pain occur. "Sterile pyuria" is a characteristic finding. The urine contains white blood cells, but cultures for the common urinary tract bacterial pathogens show no growth. However, mycobacterial cultures are often positive.

Note that most (approximately 90%) infections with *M. tuberculosis* are asymptomatic. Asymptomatic infections, also known as **latent infections**, can reactivate and cause symptomatic tuberculosis. **The most important determinant of whether overt disease occurs is the adequacy of the host's cell-mediated immune (CMI) response**. For example, AIDS patients have a very high rate of reactivation of prior asymptomatic infection and of rapid progression of the disease. In these patients, untreated disease caused by *M. tuberculosis* has a 50% mortality rate. Furthermore, administration of infliximab (Remicade), a monoclonal antibody that neutralizes tumor necrosis factor (TNF), has activated latent tuberculosis in some patients. Remicade is used in the treatment of rheumatoid arthritis (see Chapter 66). Diabetics also are predisposed to reactivation and progression of disease.

In some patients with AIDS who are infected with *M. tuberculosis*, treating the patient with highly active antiretroviral therapy (HAART) causes an exacerbation of symptoms. This phenomenon is called immune reconstitution inflammatory syndrome (IRIS). The explanation of the exacerbation of symptoms is that HAART increases the number of CD4 cells, which increases the inflammatory response. To prevent this, patients should be treated for the underlying infection before starting HAART.

Laboratory Diagnosis

Acid-fast staining of sputum or other specimens is the usual initial test (see Figure 21–1). Either the Kinyoun version of the acid-fast stain or the older Ziehl-Neelsen version can be used. For rapid screening purposes, auramine stain, which can be visualized by fluorescence microscopy, is used.

After digestion of the specimen by treatment with NaOH and concentration by centrifugation, the material is cultured on special media, such as Löwenstein-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. For example, *M. tuberculosis* produces **niacin,** whereas almost no other mycobacteria do. It also produces catalase.

Nucleic acid amplification tests can be used to detect the presence of *M. tuberculosis* directly in clinical specimens such as sputum. Tests are available that detect either the ribosomal RNA or the DNA of the organism. These tests are highly specific, but their sensitivity varies. In sputum specimens that are acid-fast stain positive, the sensitivity is high, but in "smear-negative" sputums, the sensitivity is significantly lower. These tests are quite useful in deciding whether to initiate therapy prior to obtaining the culture results.

Because drug resistance, especially to isoniazid (see later), 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. To address this problem, molecular tests are available, which detect mutations in the chromosomal genes that encode either the catalase gene that mediates resistance to isoniazid or the RNA polymerase gene that mediates resistance to rifampin. The **luciferase assay,** which can detect drug-resistant organisms in a few days, is also used. Luciferase is an enzyme isolated from fireflies that produces flashes of light in the presence of adenosine triphosphate (ATP). If the organism isolated from the patient is resistant, it will not be damaged by the drug (i.e., 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.

There are two approaches to the diagnosis of latent infections. One is the PPD skin test as described in the "Immunity & Hypersensitivity" section earlier in this chapter. Because there are problems both in the interpretation of the PPD test and with the person returning for the skin test to be read, a quantifiable laboratory-based test is valuable. This laboratory test is an **interferon-γ release assay (IGRA),** and there are two versions available: Quantiferon-TB and T-spot-TB. In this assay, blood cells from the patient are exposed to antigens from *M. tuberculosis*, and the amount of interferon-γ released from the cells is measured. The sensitivity and specificity of the IGRA are as good as the PPD skin test. Because the antigens used in the test are specific for *M. tuberculosis* and are not present in BCG, the test is not influenced by whether a person has been previously immunized with the BCG vaccine.

Treatment & Resistance

Multidrug therapy is used to prevent the emergence of drug-resistant mutants during the long (6- to 9-month) duration of treatment. (Organisms that become resistant to one drug will be inhibited by the other.) **Isoniazid** (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. A somewhat different regimen can also be used. A convenient way to remember that regimen is to give four drugs (isoniazid, rifampin, pyrazinamide, and ethambutol) for 2 months and two drugs (isoniazid and rifampin) for 4 months. In patients who are immunocompromised (e.g., 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 to 12 months.

Although therapy is usually given for months, the patient's sputum becomes **noninfectious within 2 to 3 weeks.** The necessity for protracted therapy is attributed to (1) the intracellular location of the organism; (2) caseous material, which blocks penetration by the drug; (3) the slow growth of the organism; and (4) metabolically inactive "persisters" within the lesion. Because metabolically inactive organisms may not be killed by antitubercular drugs, treatment may not eradicate the infection, and reactivation of the disease may occur in the future.

Lymphadenitis, including cervical lymphadenitis (scrofula) caused by *M. tuberculosis*, should be treated with the drug regimens described earlier for disseminated disease. Scrofula caused by *M. scrofulaceum* can be treated by surgical excision of the single cervical lymph node, but alternative approaches exist. A complete discussion of these is beyond the scope of this book.

Treatment of **latent (asymptomatic) infections** consists of INH taken for 6 to 9 months or INH plus rifapentine for 3 months. This approach is most often used in asymptomatic patients whose PPD skin test recently converted to positive. The risk of symptomatic infection is greatest within the first 2 years after infection, so INH is particularly indicated for these "recent converters." INH is also used in children exposed to patients with symptomatic tuberculosis. Patients who receive INH should be evaluated for drug-induced hepatitis, especially those over the age of 35 years. Rifampin can be used in those exposed to INH-resistant strains. A combination of rifampin and pyrazinamide should not be used because it causes a high rate of severe liver injury.

Resistance to INH and other antituberculosis drugs is being seen with increasing frequency in the United States, especially in immigrants from Southeast Asia and Latin America. Strains of *M. tuberculosis* **resistant to multiple drugs** (MDR strains) have emerged, primarily in AIDS patients. The most common pattern is resistance to both INH and rifampin, but some isolates are resistant to three or more drugs. The treatment of MDR organisms usually involves the use of four or five drugs, including ciprofloxacin, amikacin, ethionamide, and cycloserine. The precise recommendations depend on the resistance pattern of the isolate and are beyond the scope of this book.

In 2013, a new drug, bedaquiline, was approved for the treatment of MDR strains. It should be used in combination with other drugs, not as monotherapy. It is a diarylquinoline that inhibits an ATP synthase unique to *M. tuberculosis*.

Previous treatment for tuberculosis predisposes to the selection of these MDR organisms. **Noncompliance** (i.e., the failure of patients to complete the full course of therapy) is a major factor in allowing the resistant organisms to survive. One approach to the problem of noncompliance is **directly observed therapy (DOT),** in which health care workers observe the patient taking the medication.

The strains of *M. tuberculosis* resistant to INH, rifampin, a fluoroquinolone, and at least one additional drug are called extensively drug-resistant (XDR) strains. XDR strains emerged in 2005 among HIV-infected patients in South Africa.

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. At present, prevention of the spread of the organism depends largely on the prompt identification and adequate treatment of patients who are coughing up the organism. The use of masks and other respiratory isolation procedures to prevent spread to medical personnel is also important. Contact tracing of individuals exposed to patients with active pulmonary disease who are coughing should be done.

An important component of prevention is the use of the PPD skin test to detect recent converters and to institute treatment for latent infections as described earlier. Groups that should be screened with the PPD skin test include people with HIV infection, close contacts of patients with active tuberculosis, low-income populations, alcoholics and intravenous drug users, prison inmates, and foreign-born individuals from countries with a high incidence of tuberculosis.

Because there are some problems associated with PPD skin tests, such as the measurement and the interpretation of results and the inconvenience of the patient having to return for the skin test to be read, a laboratory test to detect latent infections was developed. This test, called Quantiferon-TB (QFT), measures the amount of interferon-γ released from the patient's lymphocytes after exposure to PPD in cell culture. QFT requires only a single blood specimen and determines the amount of interferon-γ by an enzyme-linked immunosorbent assay (ELISA) test.

BCG vaccine can be used to induce partial resistance to tuberculosis. The vaccine contains a strain of live, attenuated *M. bovis* called bacillus Calmette-Guérin. The vaccine is effective in preventing the appearance of tuberculosis as a clinical disease, especially in children, although it does not prevent infection by *M. tuberculosis*. However, a major problem with the vaccine is its variable effectiveness, which can range from 0% to 70%. It is used primarily in areas of the world where the incidence of the disease is high. It is *not* usually used in the United States because of its variable effectiveness and because the incidence of the disease is low enough that it is not cost-effective.

The skin test reactivity induced by the vaccine given to children wanes with time, and the interpretation of the skin test reaction in adults is not altered by the vaccine. For example, skin test reactions of 10 mm or more should not be attributed to the vaccine unless it was administered recently. In the United States, use of the vaccine is limited to young children who are in close contact with individuals with active tuberculosis and to military personnel. BCG vaccine should not be given to immunocompromised people because the live BCG organisms can cause disseminated disease.

BCG vaccine is also used to treat bladder cancer. The vaccine is instilled into the bladder and serves to nonspecifically stimulate cell-mediated immunity, which can inhibit the growth of the carcinoma cells.

Pasteurization of milk and destruction of infected cattle are important in preventing intestinal tuberculosis.

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

TABLE 21–3 Runyon's Classification of Atypical Mycobacteria

Group	Growth Rate	Pigment Formation		Typical Species
		Light	Dark	
I	Slow	+	–	*M. kansasii, M. marinum*
II	Slow	+	+	*M. scrofulaceum*
III	Slow	–	–	*M. avium–intracellulare complex*
IV	Rapid	–	–	*M. fortuitum–chelonae complex*

for guinea pigs, whereas *M. tuberculosis* is found only in humans and is highly pathogenic for guinea pigs. The atypical mycobacteria are sometimes called mycobacteria other than tuberculosis (MOTTs) or non-tuberculous mycobacteria (NTM).

The atypical mycobacteria are classified into four groups according to their rate of growth and whether they produce pigment under certain conditions (Table 21–3). The atypical mycobacteria in groups I, II, and III grow slowly, at a rate similar to that of *M. tuberculosis*, whereas those in group IV are "rapid growers," producing colonies in fewer than 7 days. 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**).

Group I (Photochromogens)

Mycobacterium kansasii causes lung disease clinically resembling tuberculosis. Because it is antigenically similar to *M. tuberculosis*, patients are frequently tuberculin skin test–positive. Its habitat in the environment is unknown, but infections by this organism are localized to the midwestern states and Texas. It is susceptible to the standard antituberculosis drugs.

Mycobacterium marinum causes "swimming pool granuloma," also known as "fish tank granuloma." These granulomatous, ulcerating lesions occur in the skin at the site of abrasions incurred at swimming pools and aquariums. The natural habitat of the organism is both fresh and salt water. Treatment with a tetracycline such as minocycline is effective.

Group II (Scotochromogens)

Mycobacterium scrofulaceum causes scrofula, a granulomatous cervical adenitis, usually in children. (*M. tuberculosis* also causes scrofula.) The organism enters through the oropharynx and infects the draining lymph nodes. Its natural habitat is environmental water sources, but it has also been isolated as a saprophyte from the human respiratory tract. Scrofula can often be cured by surgical excision of the affected lymph nodes.

Group III (Nonchromogens)

Mycobacterium avium–intracellulare complex (MAI, MAC) is composed of two species, *M. avium* and *M. intracellulare*, that are very difficult to distinguish from each other by standard laboratory tests. They cause pulmonary disease clinically indistinguishable from tuberculosis, primarily in immunocompromised patients such as those with AIDS who have CD4 cell counts of less than 200/μL. MAI is the most common bacterial cause of disease in AIDS patients. The organisms are widespread in the environment, including water and soil, particularly in the southeastern United States. They are highly resistant to antituberculosis drugs, and as many as six drugs in combination are frequently required for adequate treatment. Current drugs of choice are azithromycin or clarithromycin plus one or more of the following: ethambutol, rifabutin, or ciprofloxacin. Azithromycin is currently recommended for preventing disease in AIDS patients.

Group IV (Rapidly Growing Mycobacteria)

Mycobacterium fortuitum–chelonae complex is composed of two similar species, *M. fortuitum* and *M. chelonae*. They are saprophytes, found chiefly in soil and water, and rarely cause human disease. Infections occur chiefly in two populations: (1) immunocompromised patients and (2) individuals with prosthetic hip joints and indwelling catheters. Skin and soft tissue infections occur at the site of puncture wounds (e.g., at tattoo sites). They are often resistant to antituberculosis therapy, and therapy with multiple drugs in combination plus surgical excision may be required for effective treatment. Current drugs of choice are amikacin plus doxycycline.

Mycobacterium abscessus is another rapidly growing mycobacteria acquired from the environment. It causes chronic lung infections, as well as infections of the skin, bone, and joints. It is highly antibiotic-resistant. Current drugs of choice are amikacin plus imipenem or cefoxitin plus clarithromycin.

Mycobacterium smegmatis is a rapidly growing mycobacterium that is not associated with human disease. It is part of the normal flora of smegma, the material that collects under the foreskin of the penis.

MYCOBACTERIUM LEPRAE

Disease

This organism causes leprosy (Hansen's disease).

Important Properties

Mycobacterium leprae has **not been grown** in the laboratory, either on artificial media or in cell culture. It can be grown in experimental animals, such as mice and armadillos. Humans are the natural hosts, although the armadillo appears to be a reservoir for human infection in the Mississippi Delta region where these animals are common. In view of this, leprosy can be thought of as a zoonotic disease, at least in certain southern states, such as Louisiana and Texas.

The optimal temperature for growth (30°C) is lower than body temperature; therefore, *M. leprae* grows preferentially in the skin and superficial nerves. It grows very slowly, with a doubling time of 14 days. This makes it the slowest-growing human bacterial pathogen. One consequence of this is that antibiotic therapy must be continued for a long time, usually several years.

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. In the United States, leprosy occurs primarily in Texas, Louisiana, California, and Hawaii. Most cases are found in immigrants from Mexico, the Philippines, Southeast Asia, and India. The disease occurs worldwide, with most cases in the tropical areas of Asia and Africa. The armadillo is unlikely to be an important reservoir because it is not found in many areas of the world where leprosy is endemic.

Pathogenesis

The organism replicates intracellularly, typically within skin histiocytes, endothelial cells, and the Schwann cells of nerves. The nerve damage in leprosy is the result of two processes: damage caused by direct contact with the bacterium and damage caused by CMI attack on the nerves.

There are two distinct forms of leprosy—**tuberculoid** and **lepromatous**—with several intermediate forms between the two extremes (Table 21–4).

(1) In tuberculoid (also known as **paucibacillary**) leprosy, the CMI response to the organism limits its growth, very few acid-fast bacilli are seen, and granulomas containing giant cells form. The nerve damage seems likely to be caused by cell-mediated immunity as there are few organisms and the CMI response is strong.

The CMI response consists primarily of CD4-positive cells and a Th-1 profile of cytokines, namely, interferon-γ, interleukin-2, and interleukin-12. It is the CMI response that causes the nerve damage seen in tuberculoid leprosy.

The lepromin skin test result is positive. The lepromin skin test is similar to the tuberculin test (see earlier). An extract of *M. leprae* is injected intradermally, and induration is observed 48 hours later in those in whom a CMI response against the organism exists.

(2) In lepromatous (also known as **multibacillary**) 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 result is negative. The nerve damage seems likely to be caused by direct contact as there are many organisms and the CMI response is poor.

There is evidence that people with lepromatous leprosy produce interferon-β (antiviral interferon) in response to *M. leprae* infection, whereas people with tuberculoid leprosy produce interferon-γ. Interferon-β inhibits the synthesis of interferon-γ thereby reducing the CMI response needed to contain the infection.

Note that in lepromatous leprosy, only the cell-mediated response to *M. leprae* is defective (i.e., 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. The T-cell response consists primarily of Th-2 cells.

Clinical Findings

The incubation period averages several years, and the onset of the disease is gradual. In tuberculoid leprosy, hypopigmented macular or plaquelike skin lesions, thickened superficial nerves, and significant anesthesia of the skin lesions occur (Figure 21–5). In lepromatous leprosy, multiple nodular skin lesions occur, resulting in the typical

TABLE 21–4 Comparison of Tuberculoid and Lepromatous Leprosy

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

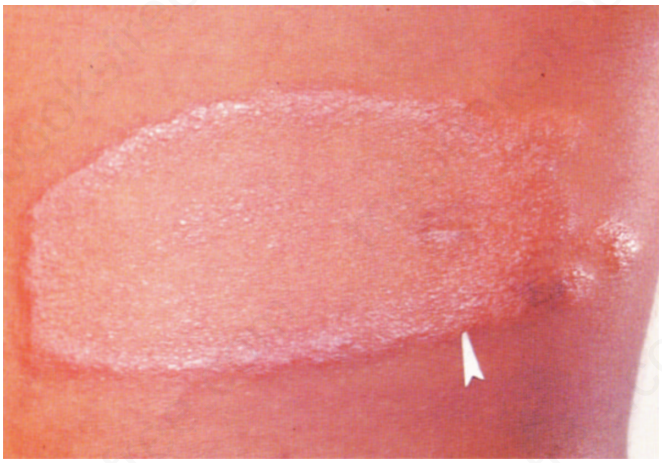

FIGURE 21–5 Tuberculoid leprosy. The tuberculoid form is characterized by a single, flat, hypopigmented lesion that has lost sensation. (Reproduced with permission from Longo DL et al [eds]. *Harrison's Principles of Internal Medicine*. 18th ed. New York: McGraw-Hill, 2012. Copyright © 2012 by The McGraw-Hill Companies, Inc.)

leonine (lionlike) **facies** (Figure 21–6). After the onset of therapy, patients with lepromatous leprosy often develop **erythema nodosum leprosum** (ENL), which is interpreted as a sign that cell-mediated immunity is being restored. ENL is characterized by painful nodules, especially along the extensor surfaces of the tibia and ulna, neuritis, and uveitis.

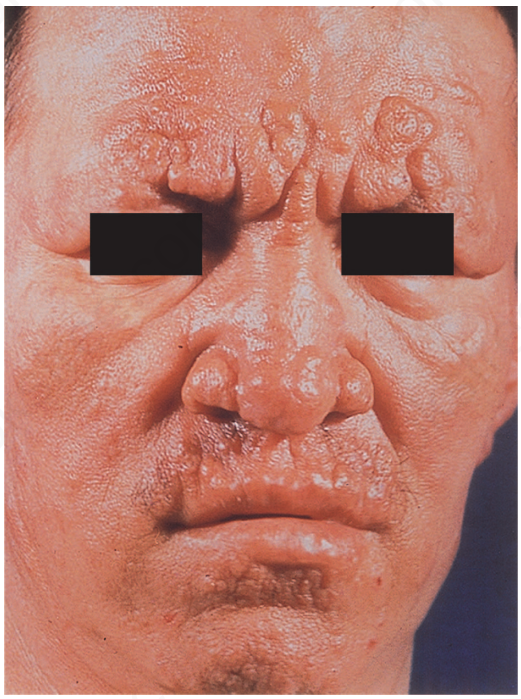

FIGURE 21–6 Lepromatous leprosy. The lepromatous form is characterized by multiple, raised lesions, often with the appearance of leonine facies (the face resembles a lion with a prominent brow). (Used with permission from Robert H. Gelber, MD.)

The disfiguring appearance of the disease results from several factors: (1) the skin anesthesia results in burns and other traumas, which often become infected; (2) resorption of bone leads to loss of features such as the nose and fingertips; and (3) infiltration of the skin and nerves leads to thickening and folding of the skin. In most 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. Lipid-laden macrophages called "foam cells" containing many acid-fast bacilli are seen in the skin. In the tuberculoid form, very few organisms are seen, and the appearance of typical granulomas is sufficient for diagnosis. Cultures are negative because the organism does not grow on artificial media.

A serologic test for IgM against phenolic glycolipid-1 is useful in the diagnosis of lepromatous leprosy but is not useful in the diagnosis of tuberculoid leprosy. The diagnosis of lepromatous leprosy can be confirmed by using the polymerase chain reaction (PCR) test on a skin sample. False-positive results in the nonspecific serologic tests for syphilis, such as the Venereal Disease Research Laboratory (VDRL) and rapid plasma reagin (RPR) tests, occur frequently in patients with lepromatous leprosy.

Treatment

The mainstay of therapy is **dapsone** (diaminodiphenylsulfone), but because sufficient resistance to the drug has emerged, combination therapy is now recommended. For tuberculoid (paucibacillary) leprosy, dapsone and rifampin are given for 6 to 12 months whereas for lepromatous (multibacillary) leprosy, a combination of dapsone, rifampin, and clofazimine is given for 12 to 24 months. A combination of ofloxacin plus clarithromycin is an alternative regimen. Thalidomide is the treatment of choice for severe ENL reactions.

Prevention

Isolation of all lepromatous patients, coupled with chemoprophylaxis with dapsone for exposed children, is required. There is no vaccine.

SELF-ASSESSMENT QUESTIONS

1. Your patient is a 25-year-old homeless man who complains of a cough for the past month. The cough is now productive of several tablespoons of blood-streaked sputum per day. The sputum is not foul-smelling. He has lost 10 pounds but says that he doesn't eat regularly. On physical exam, temperature is 38°C, and coarse rales were heard in the apex of the left lung. An acid-fast stain of

the sputum reveals acid-fast rods. Culture of the sputum shows no growth at 7 days, but buff-colored colonies are visible at 21 days. Of the following organisms, which one is most likely to be the cause of this infection?

(A) *Mycobacterium fortuitum-chelonae*
(B) *Mycobacterium leprae*
(C) *Mycobacterium marinum*
(D) *Mycobacterium tuberculosis*

2. Which one of the following regimens is optimal initial treatment for the patient in Question 1?

(A) Isoniazid for 9 months
(B) Isoniazid and gentamicin for 2 weeks
(C) Isoniazid and rifampin for 4 months
(D) Isoniazid, rifampin, ethambutol, and pyrazinamide for 2 months

3. Your patient is a 70-year-old man with progressive weakness in both legs that began about a week ago. He reports back pain and fever for the past month. Magnetic resonance imaging (MRI) of the spine reveals destruction of the seventh thoracic vertebra and a paravertebral mass. Surgical decompression and debridement was performed. Histologic examination of the mass revealed caseating granulomas, and Langhans' giant cells were observed in the granulomas. Gram stain revealed no organisms, but an acid-fast stain showed red rods. Culture shows no growth at 7 days, but growth is seen at 28 days. Of the following, which one is the most likely cause?

(A) *Mycobacterium fortuitum-chelonae*
(B) *Mycobacterium leprae*
(C) *Mycobacterium marinum*
(D) *Mycobacterium tuberculosis*

4. Your patient is a 30-year-old woman who is infected with HIV and has a low CD4 count. She now has the findings of pulmonary tuberculosis, but you are concerned that she may be infected with *Mycobacterium avium–intracellulare* (MAI). Regarding MAI, which one of the following is most accurate?

(A) Disseminated disease caused by MAI is typically the result of decreased antibody production, whereas disseminated disease caused by *M. tuberculosis* is typically caused by reduced cell-mediated immunity.
(B) Immigrants from Southeast Asia are more likely to be infected with MAI than with *M. tuberculosis*.
(C) In the clinical laboratory, MAI forms colonies in 7 days, whereas *M. tuberculosis* colonies typically require at least 21 days of incubation for colonies to appear.
(D) MAI is typically susceptible to a drug regimen of isoniazid and rifampin, whereas *M. tuberculosis* is often resistant.
(E) The natural habitat of MAI is the environment, whereas the natural habitat of *M. tuberculosis* is humans.

5. Regarding the patient in Question 4, if MAI was shown to be the cause of her symptoms, which one of the following is the best choice of antibiotics to prescribe?

(A) Amikacin and doxycycline
(B) Clarithromycin, ethambutol, and rifabutin
(C) Dapsone, rifampin, and clofazimine
(D) Isoniazid and gentamicin
(E) Isoniazid, rifampin, ethambutol, and pyrazinamide

6. Your patient is a 20-year-old man with a single, slowly expanding, nonpainful scaly lesion on his chest for the past 2 months. The lesion is nonpruritic, and he has lost sensation at the site of the lesion. He is otherwise well. He is a recent immigrant from Central America. An acid-fast stain of a scraping of the lesion is positive. Which one of the following diseases is he most likely to have?

(A) Cutaneous tuberculosis
(B) Fish tank granuloma
(C) Lepromatous leprosy
(D) Scrofula
(E) Tuberculoid leprosy

ANSWERS

1. **(D)**
2. **(D)**
3. **(D)**
4. **(E)**
5. **(B)**
6. **(E)**

SUMMARIES OF ORGANISMS

Brief summaries of the organisms described in this chapter begin on page 664. Please consult these summaries for a rapid review of the essential material.

PRACTICE QUESTIONS: USMLE & COURSE EXAMINATIONS

Questions on the topics discussed in this chapter can be found in the Clinical Bacteriology section of Part XIII: USMLE (National Board) Practice Questions starting on page 713. Also see Part XIV: USMLE (National Board) Practice Examination starting on page 751.