TURKISH
SLOVAK
SPANISH
ALBANIAN
SHARE BOOKMARK
PRINT THIS PAGE
Let us know what you think
FEEDBACK
SEARCH
<<Previous Next >>

KEYWORDS
Acid Fast
Tuberculosis
M. tuberculosis
M. bovis
Tubercle
PPD
Tuberculin
Mycobactin
Cord factor
BCG
Mycolic acids
Atypicals
M. avium - M. intracellulare complex
Runyon groups
Leprosy (Hansen's Disease)
M. leprae
Diphtheria
C. diphtheriae
Loeffler's agar
Tellurite agar
Metachromatic bodies
Diphtheria toxin
Schick test
Diphtheroids
Legionella pneumophila
Legionnaires disease
Pontiac Fever
Lepromin
Legionella (Tatlockia) micdadei
Charcoal yeast extract agar
Fatty acid profiles
Hybridization

BACTERIOLOGY - CHAPTER SIXTEEN

MYCOBACTERIA AND CORYNEBACTERIA

Dr Alvin Fox
Emeritus Professor
University of South Carolina School of Medicine

MYCOBACTERIA

In the 1980's, many experts felt that the days of tuberculosis as a threat to the US population had passed and the incidence of new cases (around 20,000 a year) was slowly decreasing, even though it was still the leading infectious cause of death world-wide. The situation in the 1990's has changed dramatically. The incidence of tuberculosis has slightly increased and the disease is certainly not going away (This is primarily due to the AIDS epidemic). At the same time multiple drug-resistant strains of *M. tuberculosis* are appearing regularly. The *M. avium - M. intracellulare* complex, long considered a group of organisms that only rarely infects man, is now recognized as one of the leading opportunists associated with AIDS. *M. leprae* is the causative agent of leprosy which remains a major disease in the third world. Due to eradication of infected cattle and pasteurization of milk *M. bovis* (a zoonotic cause of tuberculosis) is rarely seen in the United States.

Mycobacteria are obligate aerobic, acid-fast rods.

Mycobacterium tuberculosis

Tuberculosis is extremely common in third world countries and WHO estimates that about one third of the world's population is infected, although the rate of mortality due to tuberculosis around the world has fallen by 45% between 1990 and 2012. It is estimated that around the world in 2012:

- 1.3 million people died of the disease, including 320,000 HIV-infected patients.
- About 8.6 million people showed signs of infection and sickness, including 1.1 million HIV-infected people.
- About half of deaths among HIV-infected patients were in women.
- Among children, in 2012, 530,000 became sick with tuberculosis. 74,000 HIV-negative children died.

More data from WHO are given in figure 5.

In the United States there were 9,945 cases of tuberculosis in 2012 (3.2 cases peer 100,000 population). There was a decline of 5.4% in the number of cases over those reported in 2011 (figure 1). In 1953, there were 84,304 cases of tuberculosis (a case rate of 52.6 per 100,000 population). Rates were higher in the older population (figure 2) and an increasing proportion of cases is seen in foreign-born persons (figures 3 and 4). In the United States in 2013, 64% of tuberculosis cases and 91% of multidrug–resistant cases occurred in people born in other countries. Three quarters of these cases came from 15 countries.

Pathogenesis of tuberculosis

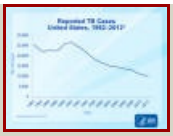


Figure 1 Reported cases of tuberculosis in the United States. 1982-2012. CDC

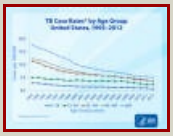


Figure 2 Tuberculosis cases by age group in the United States. CDC


Figure 3 Number of US tuberculosis cases. Foreign-born patients versus US-born. 1993-2012. CDC

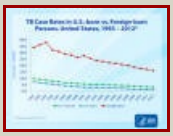


Figure 4 Tuberculosis case rate in US-born and foreign born persons. 1993-2012. CDC


Figure 4a Computer-generated image of a cluster of rod-shaped drug-resistant *Mycobacterium tuberculosis*. Image based on scanning electron microscopy. CDC

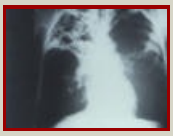


Figure 4b Anteroposterior x-ray of a patient's chest diagnosed with bilateral pulmonary tuberculosis. The x-ray revealed the presence of bilateral pulmonary infiltrate, and “caving formation” present in the right apical region. The diagnosis was "far-advanced" tuberculosis. CDC

Mycobacterium tuberculosis is spread via aerosols when an infected person coughs or sneezes; however, many people who become infected do not show any symptoms of the disease. This is a latent tuberculosis infection and these people are not infectious. The bacteria are kept under control by the immune system. In contrast, other people's immune system cannot control the bacteria and they show overt tuberculosis disease. Some, about 5 to 10%, people with latent tuberculosis may develop active disease many years after infection when their immune system weakens for a variety of reasons. Especially prone to activation of overt disease are people whose immune system has been weakened by HIV infection.

CDC lists the following categories of people who are at high risk for developing disease

- Close contacts of a person with infectious tuberculosis
- People who have immigrated from areas of the world with high rates of tuberculosis
- Children less than 5 years of age who have a positive tuberculosis test
- People with high rates of tuberculosis transmission including:
  homeless people
  intravenous drug users
  HIV-infected people
- People who work or reside with people who are at high risk for tuberculosis in facilities or institutions such as hospitals, homeless shelters, correctional facilities, nursing homes, and residential homes for those with HIV

Medical conditions that lead to a weakened immune system and therefore predispose people to overt tuberculosis disease include:

- HIV infection
- Drug abuse
- Silicosis
- Diabetes mellitus
- Severe kidney disease
- Low body weight
- Organ transplants
- Head and neck cancer
- Corticosteroids treatment
- Treatment for rheumatoid arthritis or Crohn’s disease

M. tuberculosis bacteria (figure 4a) infect the lungs (pulmonary tuberculosis) and are distributed systemically within macrophages where they survive intracellularly. Inhibition of phagosome-lysosome fusion and resistance to lysosomal enzymes have both been suggested to play a role. Cell-mediated immunity develops which causes infiltration of macrophages and lymphocytes with development of granulomas (tubercles). The disease can be diagnosed (figure 6) by skin testing for delayed hypersensitivity with tuberculin (also know as protein purified purified from *Mycobacterium tuberculosis*, PPD). A positive test does not indicate active disease; merely exposure to the organism.

Other pathogenesis factors (of considerably less importance than delayed hypersensitivity) include mycobactin (a siderophore) and cord factor which damages mitochondria.

Symptoms of tuberculosis

These depend on the site of infection. In pulmonary tuberculosis, symptoms include:

- a cough that lasts 3 weeks or longer
- chest pain
- blood or sputum (phlegm from deep inside the lungs)
- weakness/fatigue
- weight loss
- appetite loss
- chills
- fever
- night sweats

Diagnosis and identification

A positive skin (Mantoux) test shows whether a person has been infected by the bacteria but people who have been inoculated against tuberculosis using the BCG vaccine can also give a positive test. X-ray imaging (figure 4b) is also often used.

The presence of acid fast bacteria in sputum is a rapid presumptive test for tuberculosis. Subsequently, when cultured, *M. tuberculosis* will grow very slowly producing distinct non-pigmented colonies after several weeks. *M. tuberculosis* can be differentiated from most other mycobacteria by the production of niacin. A rapid alternative to culture is polymerase chain amplification (PCR).

There are also blood tests called interferon-gamma release assays (IGRAs) which are done on blood samples. These are not affected by prior BCG vaccination.

Treatment

Tuberculosis is usually treated for extensive time periods (9 months or longer) since the organism grows slowly and may become dormant. By using two or more antibiotics (including rifampin, rifapentine and isoniazid), the possibility of resistance developing during this extended time is minimized.

> Recommended treatment for overt tuberculosis includes some of ten approved drugs taken over a period of six to nine months. These drugs include, as a first line of attack:
>
> - isoniazid
> - rifampin
> - ethambutol
> - pyrazinamide

A new anti-tuberculosis drug

In 2013, a new anti-tuberculosis drug, bedaquiline (R207910), was approved by the Federal Drug Administration for use in patients infected with multi-drug resistant *Mycobacterium tuberculosis*. This is the first new drug to treat tuberculosis in over 40 years. The drug inhibits part of the F0 subunit of ATP synthase in the bacterial membrane and probably inhibits the proton pump leading to altered pH homeostasis and ATP depletion. Because this is a unique target, not inhibited by other anti-bacterial drugs, cross resistance with other anti-tuberculosis drugs does not occur. Nevertheless, since the bacterium can develop resistance to bedaquiline, it is only approved for use in combination therapy with other drugs

Vaccination

The BCG vaccine (*Bacillus de Calmette et Guerin*, an attenuated strain of *M. bovis*) has not been shown to be effective in many studies, yet in others a protective effect has been seen. In the United Kingdom, a protective effect of 60 to 80% has been reported. It is not known why the efficacy of BCG is so different in different studies. In most cases, however, efficacy seem to wane over a number of years. In the United Kingdom all school children received BCG vaccination until 2005. This was abandoned because it was not found to be cost effective since 94 children would have to be immunized in 1953 to prevent one case of tuberculosis but the fall in the incidence of the disease meant that by 1988 12,000 children would need vaccination to prevent one case.

In the United States, where the incidence of tuberculosis is low, widespread vaccination is not practiced. Indeed immunization (resulting in a positive tuberculin test) is felt to interfere with diagnosis.

In other countries, BCG vaccination has been carried out in school children and mass vaccination campaigns.


Figure 5a Estimated TB incidence rates 2012 WHO


Figure 5b Estimated TB mortality rates excluding TB deaths among HIV-positive people, 2012 WHO


Figure 5c Estimated HIV prevalence in new TB cases, 2012 WHO


Figure 5d Number of multidrug-resistant tuberculosis cases estimated to occur among notified pulmonary TB cases, 2012 WHO


Figure 5e Percentage of HIV-positive TB patients enrolled on antiretroviral therapy (ART), 2012 WHO


Figure 5f Percentage of new TB cases with multidrug-resistant tuberculosis WHO


Figure 5g Percentage of patients with known HIV status by country, 2012 WHO

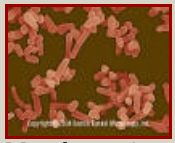

Figure 6 Mantoux intradermal tuberculin skin test for tuberculosis. CDC

Atypicals

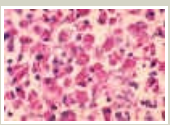
The "atypicals" generally infect the immunocompromised host and are thus not transmitted man-man. With the AIDS epidemic, the atypical mycobacteria have taken on new importance with the recognition that the *M. avium* complex (MAC) results in the most commonly associated systemic bacterial infection. Atypical mycobacteria can cause tuberculosis-like or leprosy-like, diseases, and are not susceptible to certain common anti-tuberculous antibiotics.

Mycobacterium avium complex and AIDS

*M. avium g*enerally infects AIDS patients when their CD4+ cell count decreases greatly (below 100/mm^3). *M. tuberculosis* infects AIDS patients much earlier in the disease. This clearly demonstrates the much greater virulence of *M. tuberculosis*. The incidence of systemic disease (versus primarily pulmonary) is much greater in tuberculosis associated with AIDS than in its absence. Furthermore, histologically lesions often appear lepromatous (non-granulomatous, with many organisms). It is rare to find a case of *M. avium* infection that is not AIDS associated. However, *M. tuberculosis* is a much more virulent organism. Approximately 20% of the total tuberculosis cases in the US are caused by AIDS. This helps explain why TB is no longer on the decline. Increased homelessness is also suggested to be a factor in the rise of tuberculosis.


Mycobacterium avium. Rod-shaped Bacterium (causes avian tuberculosis) (SEM x24,000) © Dennis Kunkel Microscopy, Inc. Used with permission

Treatment of *M. avium* also involves a long-term regimen of multiple drug combinations. However, this organism does not always respond to the drug regimens used to treat *M. tuberculosis*. Appropriate drug combinations are still under investigation in clinical trials. Since *M. tuberculosis* is the more virulent organism, the drug regimen selected is primarily against *M. tuberculosis*. If *M. avium* is suspected other agents effective against this organism are included.

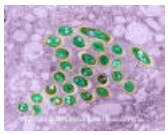

Mycobacterium avium-intracellulare infection of lymph node in patient with AIDS. Ziehl-Neelsen stain. Histopathology of lymph node shows tremendous numbers of acid-fast bacilli

Other atypicals

Presence or absence of pigmentation (and its dependence on growth in the light) and slow or fast growth rates of atypical mycobacteria allow some differentiation - the "Runyon Groups". Modern techniques allow ready speciation of mycobacteria based on their cellular fatty acid and/or mycolic acid profiles. This is only performed in reference laboratories.

within plump histiocytes.
CDC/Dr. Edwin P. Ewing, Jr.

Mycolic acids are components of a variety of lipids found only in mycobacteria, nocardia and corynebacteria. The chain length of these mycolic acids is longest in mycobacteria, intermediate in nocardia and shortest in corynebacteria. This explains why mycobacteria are generally acid fast; nocardia less acid fast; and corynebacteria are non-acid fast.


Mycobacterium avium complex (MAC) infection (human lung). Secondary infection to AIDS, HIV.


Global Leprosy Situation 2009 © World Health Organization


Leprosy: New case detection rate 2009

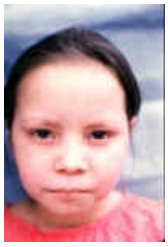
Mycobacterium leprae

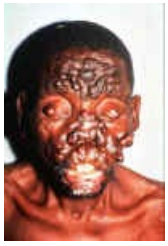
M. leprae is the causative agent of leprosy (Hansen's Disease), a chronic disease often leading to disfigurement.. It is rarely seen in the U.S. but common in the third world. The organism infects the skin, because of its growth at low temperature. It also has a strong affinity for nerves. In "tuberculoid" leprosy, there are few organisms due to control by active cell-mediated immunity. In "lepromatous" leprosy, due to immunosuppression by the organism, the opposite is found. Although uncommon in the U.S., millions of cases occur worldwide. Treatment with antibiotics (initially dapsone and now multi-drug) is effective and the overall disease incidence worldwide is down. The organism does not grow in culture media. However, it grows well in the armadillo (which has a low body temperature), allowing production of *M. leprae* antigens and pathogenesis studies. *M. leprae* has traditionally been identified on the basis of acid-fast stains of skin biopsies and clinical picture. Lepromin is used in skin testing.


Azadegan Clinic, Teheran: The foot of a woman that has been grossly disfigured through leprosy infection. © World Health Organization/TDR/Crump)


Deformity due to nerve damage with its consequent ulcers and resorption of bone. Such deformities can be worsened by careless use of the hands. © WHO/TDR


The face of a Peruvian man with active lepromatous leprosy. © WHO/ TDR/ McDougall


A young girl, 8 years old, with Burmese-Scots ancestry. The loss of eyebrows is an indication of diffuse lepromatous leprosy ©WHO/ TDR/ McDougall


The face of a patient with active, neglected nodulous lepromatous leprosy. With treatment, all nodules could be reversed. ©WHO/TDR/McDougall


The trunk of a patient showing abnormal lesions of dapsone-resistant leprosy


Most patients with leprosy can be cured with multi-drug therapy in just six months as shown in this image


Corynebacterium diphtheriae. Rod,clubed-shaped Bacterium (causes diphtheria), (SEM x24,000)


10-year-old child with severe diphtheria. CDC/NIP/Barbara Rice


This child has diphtheria resulting in a thick gray coating over back of throat. This coating can eventually expand down through airway and, if not treated, the child could die from suffocation CDC

CORYNEBACTERIA

Corynebacterium diphtheriae

C. diphtheriae grows best under strict aerobic conditions It is Gram positive and pleomorphic.

Colonization of the upper respiratory tract (pharynx and nose) and less commonly skin with *C. diphtheriae* can lead to diphtheria. The organism does not produce a systemic infection. However, in addition to a pseudomembrane being formed locally (which can cause choking), systemic and fatal injury results primarily from circulation of the potent exotoxin (diphtheria toxin). The latter begins over a period of a week. Thus treatment involves rapid therapy with anti-toxin. The gene for toxin synthesis is encoded on a bacteriophage (the tox gene). Corynebacteria that are not infected with phage, thus do not generally cause diphtheria. Diphtheria is now a disease of almost historic importance in the U.S. due to effective immunization of infants (in conjunction with pertussis and tetanus, DPT) with a toxoid (inactive toxin) which causes production of neutralizing antibodies. However, colonization is not inhibited and thus *C. diphtheriae* is still found in the normal flora (i.e. a carrier state exists). Immunity can be monitored with the Schick skin test. Treatment in non-immune individuals primarily involves injection of anti-toxin. Antibiotics are also administered at this time.

The toxin consists of two types of polypeptide. One binds to host cells; the other then becomes internalized and inhibits protein synthesis. The exotoxin catalyses the covalent attachment of the ADP-ribose moiety of NADH to a rare amino acid, diphthamide, present in EF2 (elongation factor 2). The toxin is not synthesized in the presence of iron as an iron-repressor complex forms which inhibits expression of the tox gene.

C. diphtheriae are identified by growth on Loeffler's medium followed by staining for metachromatic bodies (polyphosphate granules, Babes-Ernst bodies). The term "metachromatic" refers to the color difference of the intracellular polyphosphate granules (pink) compared to the rest of the cell (blue). Characteristic black colonies are seen on tellurite agar from precipitation of tellurium on reduction by the bacteria. Production of exotoxin can be determined by *in vivo* or *in vitro* tests.

Other organisms which morphologically resemble *C. diphtheriae* ("diphtheroids" which include other corynebacteria and also propionibacteria) are found in the normal flora. Isolates should not be confused with these organisms.

SOME MAJOR EXOTOXINS

Organism	Disease	Toxin	Further Information
Bacillus anthracis	Anthrax	Edema toxin	Edema factor/protective antigen complex
		Lethal toxin	Lethal factor/protective antigen complex

Clostridium botulinum	Botulism	Botulism .toxin	Blocks release of acetylcholine
Clostridium difficile	Pseudo membranous colitis	Enterotoxin	
Clostridium perfringens	Gas gangrene	Alpha toxin Hyaluronidase	Phospholipase, (lecithinase)
	Food poisoning	Enterotoxin	
Clostridium tetani	Tetanus	Tetanospasmin	Blocks action of inhibitory neurones
Corynebacterium diphtheriae	Diphtheria	Diphtheria toxin	Inhibits elongation factor- 2 (EF2) by ADP ribosylation
Escherichia coli	Diarrhea (ETEC)	Heat labile toxin	Activates adenyl cyclase
		Heat stable toxins	Activates adenyl cyclase
	Hemorrhagic colitis	Vero toxin	
Pseudomonas aeruginosa	Diseases of compromised host	Exotoxin A	Inhibits EF2
Staphylococcus aureus	Opportunistic infections	Alpha-gamma toxins, leucocidin	
	Toxic shock	Toxic shock toxin	
	Food poisoning	Enterotoxin	
	Scalded skin syndrome	Exfoliatin	
Streptococcus pyogenes	Scarlet fever Toxic shock	Erythrogenic/pyrogenic toxin	
Shigella dysenteriae	Bacillary dysentery	Shiga toxin	Inhibits protein synthesis by lysing 28S rRNA
Vibrio cholerae	Cholera	Choleragen	Activates adenyl cyclase by ADP-ribosylation

 Return to the Bacteriology Section of Microbiology and Immunology On-line

This page last changed on Saturday, March 05, 2016
Page maintained by Richard Hunt