الكلية: الطب

القسم او الفرع: الاحياء المجهرية

المرحلة: الثالثة

أستاذ المادة: د. ميساء إبراهيم محمد

اسم المادة باللغة العربية: احياء مجهرية

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اسم المحاضرة الأولى باللغة العربية: السل الرئوي

اسم المحاضرة الأولى باللغة الإنكليزية: Mycobacterium tuberculosis

Mycobacterium tuberculosis

DR-MAYSAAIBRAHIM

COLLEGE OF MEDICINE

DEPARTMENT OF MICROBIOLOGY

Learning Objectives

To understand the microbiology of M. tuberculosis.

To discuss pathogenesis and clinical features of TB.

To explain practical diagnostic methods for identifying M. tuberculosis.

Morphology and Characteristics

Shape:

Rod-shaped, slender bacilli.

Cell Wall:

Growth:

Rich in Acid-fast property (resistance to decolorization by acid alcohol).

mycolic acids, giving it a waxy appearance.

Microscopy Image:

Aerobic, slow-growing (forms visible colonies after 2–8 weeks).Requires special media (e.g., Löwenstein-Jensen).

Ziehl-Neelsen stain showing red M. tuberculosis bacilli against a blue background.

GENERAL CHARACTERISTICS

Mycobacterium tuberculosis (M. tuberculosis) is a species of pathogenic bacteria that is the causative agent of tuberculosis (TB).

It was first discovered in 1882 by Robert Koch and is characterized by an unusual, waxy coating on its cell surface, primarily due to the presence of mycolic acid.

This coating makes the cells impervious to Gram staining, and as a result, M. tuberculosis can appear weakly Gram-positive. Instead, acid-fast stains such as Ziehl–Neelsen, or fluorescent stains such as auramine are used to identify M. tuberculosis under a microscope.

General Characteristics

• mycobacterium are obligate aerobic (although some may grow in reduced oxygen concentrations)

• non–spore forming (except for m. marinum), non motile, very thin, slightly curved or straight rods (0.2 to 0.6 × 1 to 10 µm). some species may display a branching morphology.

• mycobacterium spp. have an unusual cell wall structure. the cell wall contains n glycolylmuramic acid instead of n-acetylmuramic acid, and it has a very high lipid content, which creates a hydrophobic permeability barrier.

• because of this cell wall structure, mycobacteria are difficult to stain with commonly used basic aniline dyes, such as those used in gram staining. although these organisms cannot be readily gram stained, they generally are considered gram positive and appear as ghost .

• the latter forms a strong complex with the mycolic acid content of the cell wall, therefore the

organism is only decolorized with 20% sulfuric acid and alcohol. this is the basis of the ziel

neelsen (zn) stain

they resist decolorization with acidified alcohol (3% hydrochloric acid) after prolonged

application of a basic fuchsin dye or with heating of this dye after its application. This

important property of mycobacteria, which derives from their cell wall structure, is

referred to as acid fastness; this characteristic distinguishes mycobacteria from other

genera. Rapid-growing mycobacteria (RGMs) may partially or completely lose this characteristic as a result of their growth characteristics.

Another important feature, they grow more slowly than most other human pathogenic

bacteria because of their hydrophobic cell surface. Because of this hydrophobicity, organisms tend to clump, so that nutrients are not easily allowed into the cell. single

cell's generation time) may range from approximately 20 hours to 36 hours.

For the most part, mycobacteria can be divided into two major groups, based on fundamental differences in epidemiology and association with disease: those belonging to

the Mycobacterium tuberculosis complex and nontuberculous mycobacteria (NTM)

Transmission and Pathogenesis

Transmission:

Droplet nuclei (1–5 microns) released during coughing, sneezing, or talking.Infectious dose: As few as 1–10 bacteria.

Pathogenesis Process:

Inhalation → alveoli → engulfment by macrophages → inhibition of phagosome-lysosome fusion.Formation of granulomas (caseous necrosis in the center).Latent TB vs. active TB (immune system determines progression).

Classification:

Mycobacteria are of many types:

TB complex bacilli:Mycobacterium tuberculosis, Mycobacterium -1 bovis

Lepromatous Mycobacteria : Mycobacteria lepri -2

Avian complex (Atypical mycobacterium) -3

Mycobacterium avium

Opportunistic Mycobacteria : Mycobacterium ulcerans, -4

Mycobacterium balani

non pathogenic Mycobacteria :Mycobacterium smegmatis-5

Antigenic structure

Cell Wall (insoluble) antigens

Peptidoglycan layer – maintains shape & rigidity

Arobinogalactan layer – survival of M.tuberculosis within macrophages

Mycolic acid layer – principal constituent, contres very low permeabilitiy acid fast and reduces the entry of most

antibiotics

Outermost layer – lipids, glycoliids & mycosides

Clinical Features

Pulmonary TB :Cough (lasting >3 weeks), hemoptysis. Fever, night sweats, weight loss, chest pain.

Extrapulmonary TB:

Lymphatic TB: Painless swelling of lymph nodes (most common).

CNS TB: Tuberculous meningitis (headache, confusion, stiff neck).

Skeletal TB: Pott's disease (spinal involvement).

Miliary TB: Disseminated form with small granulomas in multiple organs.

Images: X-rays showing cavity lesions, CT scans, or miliary TB patterns.

Diagnostic Techniques

Sample Collection.1

Pulmonary TB:

Sputum (early morning, deep cough).Induced sputum if necessary.

Microscopy: .2

Culture: .3

Extrapulmonary TB: CSF, pleural fluid, biopsy material.

Ziehl-Neelsen staining (classic method).

Auramine-rhodamine stain for fluorescent microscopy (higher sensitivity).

Limitations: Requires high bacterial load (>10^4 bacilli/ml).

Löwenstein-Jensen medium (solid): Visible colonies in 2–8 weeks.
MGIT system (mycobacterium growth indicator tube) (liquid): Faster detection (7–10 days).

Molecular Methods: .4
GeneXpert MTB/RIF: Rapid, detects rifampicin resistance.
PCR: Amplifies M. tuberculosis DNA.
Immune Tests: .5
Tuberculin Skin Test (TST): Hypersensitivity reaction to PPD.
Interferon-Gamma Release Assays (IGRAs): Detects
latent TB.
Imaging: .6
Chest X-ray: Consolidation, cavitary lesions.
CT scan: To confirm extrapulmonary TB.

A small amount of sputum was taken and brushed on the glass slide -1 until it dries
then the staining smear was immersed in constant temperature with -2 carbol fuchsin dye, and the slides were heated slowly until they were evaporated without bubbles for five minutes after the time was up. The patch is left for 5 minutes without heat, then washed with sterile water. Decolorizing (20-25%) H2SO4 was added to each slide and left for 2 -3 minutes. Then wash again with sterile water
Methylene blue was added to each slide for 2 min as a counterstain. -4
The smears were examined under a light microscope with a 100X oil

immersion objective.

Results were reported according to the standards of the World Health Organization (WHO).

Thank you