

## *Mycobacterium tuberculosis*

*Mycobacterium tuberculosis* is a species of pathogenic bacteria and the causative agent of tuberculosis. *M. tuberculosis* has a waxy coating on its cell surface primarily due to the presence of **mycolic acid**. Acid-fast stains such as Ziehl-Neelsen are used to identify *M. tuberculosis* with a microscope. *M. tuberculosis* is highly **aerobic**. Does not produce spores, and is **nonmotile**.

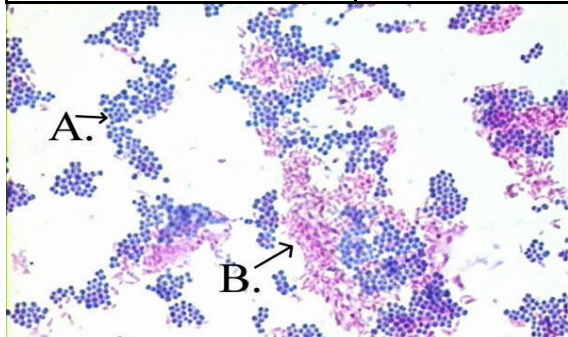
The most frequently used diagnostic methods for tuberculosis are the tuberculin skin test, acid-fast stain, culture, and polymerase chain reaction.

### Diagnosis

- Microscopy

Acid-fast stains such as Ziehl-Neelsen stain are used. Cells are curved rod-shaped and are often seen wrapped together, due to the presence of fatty acids in the cell wall that stick together.

Application of	Reagent	Cell color	
		Acid fast	Non-acid fast
Primary dye	Carbol fuchsin	Red	Red
Decolorizer (3% HCL in 95% alcohol)	Acid alcohol	Red	Colorless
Counter stain	Methylene blue	Red	Blue

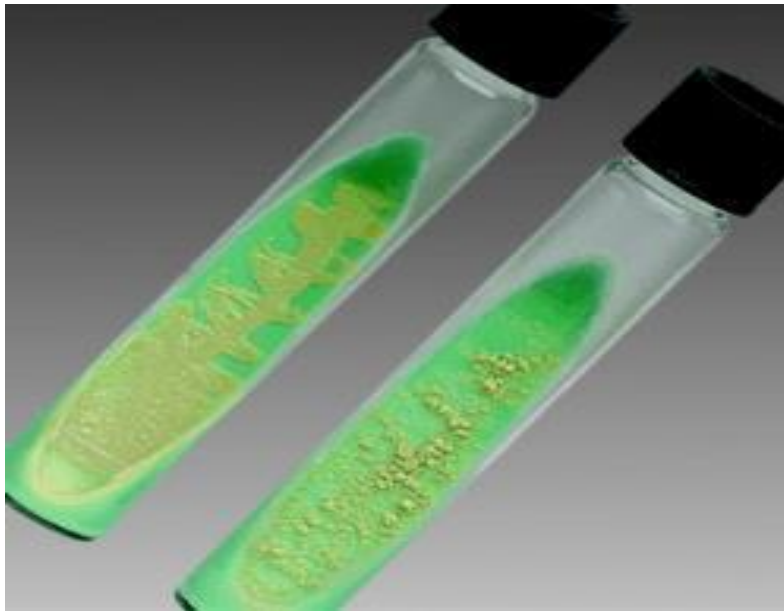


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## - Culturing

*M. tuberculosis* has a remarkably slow growth rate, doubling roughly once per day. Commonly used media include **Lowenstein-Jensen** media. *M. tuberculosis* grows slowly (*generation time of 16 to 20 hours*) and takes 3-6 weeks or longer to give visible colonies. It produces **dry, rough, raised, irregular colonies with a wrinkled surface**.

The medium appears green, opaque, and opalescent.



Low levels of penicillin and nalidixic acid are also present in LJ medium to inhibit growth of Gram-positive and Gram-negative bacteria, to limit growth to *Mycobacterium* species only. Presence of malachite green in the medium inhibits most other bacteria.

LJ medium containing glycerol favors the growth of *M. tuberculosis* while LJ medium without glycerol but containing pyruvate encourages the growth of *M. bovis*.

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## - Tuberculin skin test

- The standard recommended tuberculin test is the Mantoux test, which is administered by injecting a 0.1 mL of liquid containing 5 TU (tuberculin units) PPD (purified protein derivative) into the top layers of skin of the forearm.
- Doctors should read skin tests 48-72 hours after the injection.
- The basis of the reading of the skin test is the presence or absence and the amount of induration (localized swelling).

## - Niacin test

# BIOCHEMICAL REACTIONS

## NIACIN TEST :

- Niacin is detected by addition of 10% cyanogen bromide and 4% aniline in 96% ethanol to a suspension of the culture.
- Positive reaction – Canary yellow colour.
- *M. tuberculosis* – Positive.
- *M. bovis* – Negative.

