Tuberculosis

BY:

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History of Tuberculosis

- Tuberculosis has been found in the remains of ancient skeleton and Egyptian mummies 5000 years B.C.
- in the early 19th century it was known as “White plague”.

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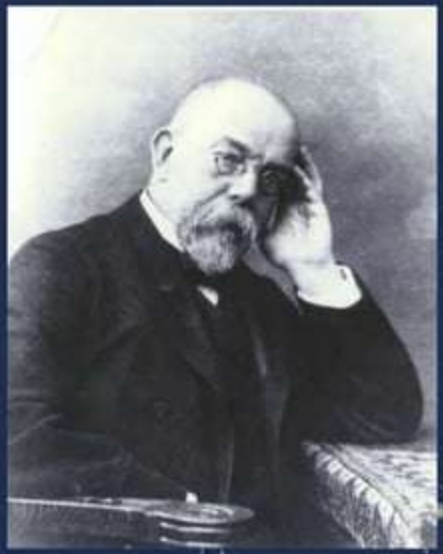
- Eventually the name Tuberculosis was acquired with discovery of tubercle bacillus by Robert Koch in 1882



What are Mycobacteria?

Scientific classification:

- •Kingdom: Bacteria
- •Phylum : Actinobacteria
- •Order : Actinomycetales
- •Suborder: Corynebacterineae
- •Family : Mycobacteriaceae
- •Genus : Mycobacterium
- •Species : M. tuberculosis

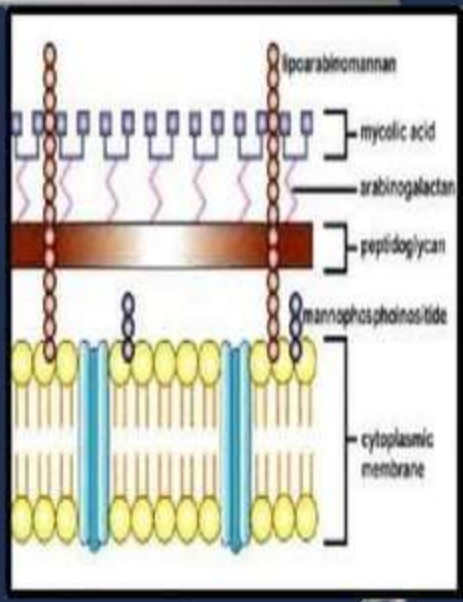
What are Mycobacteria?

- Facultative intracellular pathogens: usually infecting mononuclear phagocytes (e.g. macrophages).
- Rod shape Non-motile
- Non spore forming
- Obligate aerobic: growing most successfully in tissues with a high oxygen content, such as the lungs.


Scanning Electron Micrograph of Mycobacterium tuberculosis

What are Mycobacteria?

- Known as **"Acid-fast bacilli"**:
  - ✓ Because of their lipid-rich cell walls (**mycolic acid and wax D**), which are relatively impermeable to various basic dyes.
  - ✓ Thus they resist decolourization with acidified organic solvents.
- (Other bacteria which also contain mycolic acids, such as ***Nocardia***, can also exhibit this feature.)



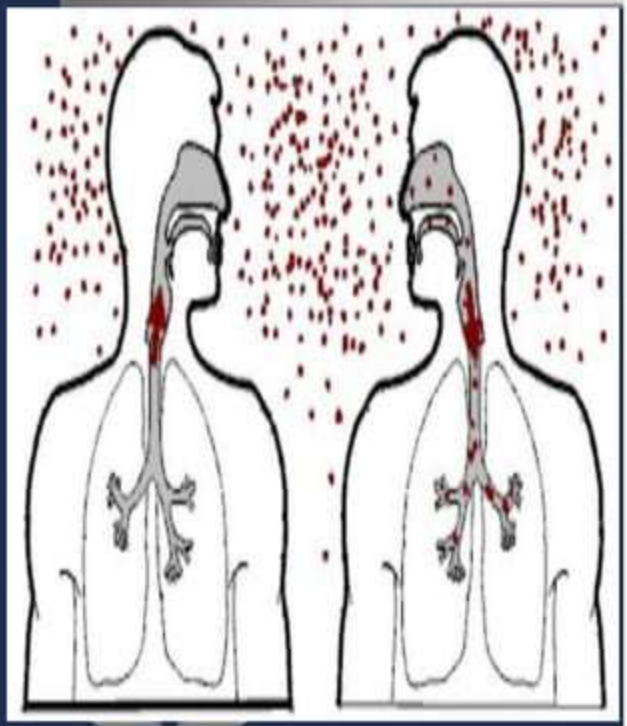
Mycobacteria Tuberculosis

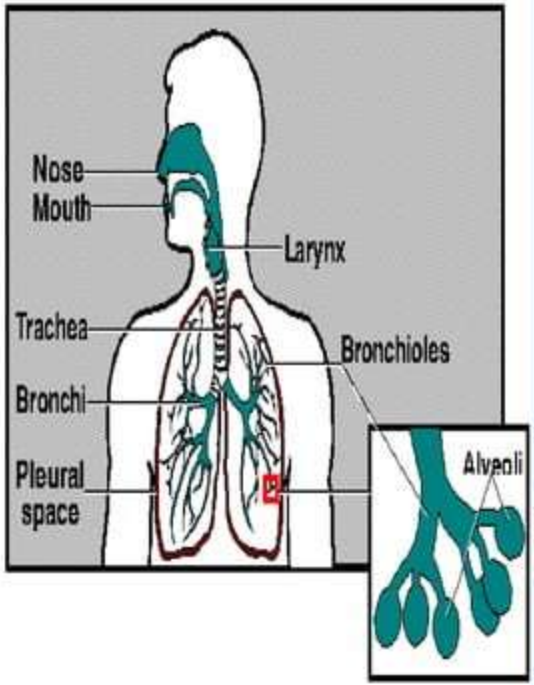
- **Mycobacterium Tuberculosis is the commonest to cause T.B. in man.**
- Worldwide, ***M. tuberculosis*** causes more deaths than any other single microbial agent.
- Tuberculosis highly Communicable Disease.
- Someone in the world is newly infected with TB bacilli every second.
- Approximately one-third 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.

Transmission (Portal of entry)

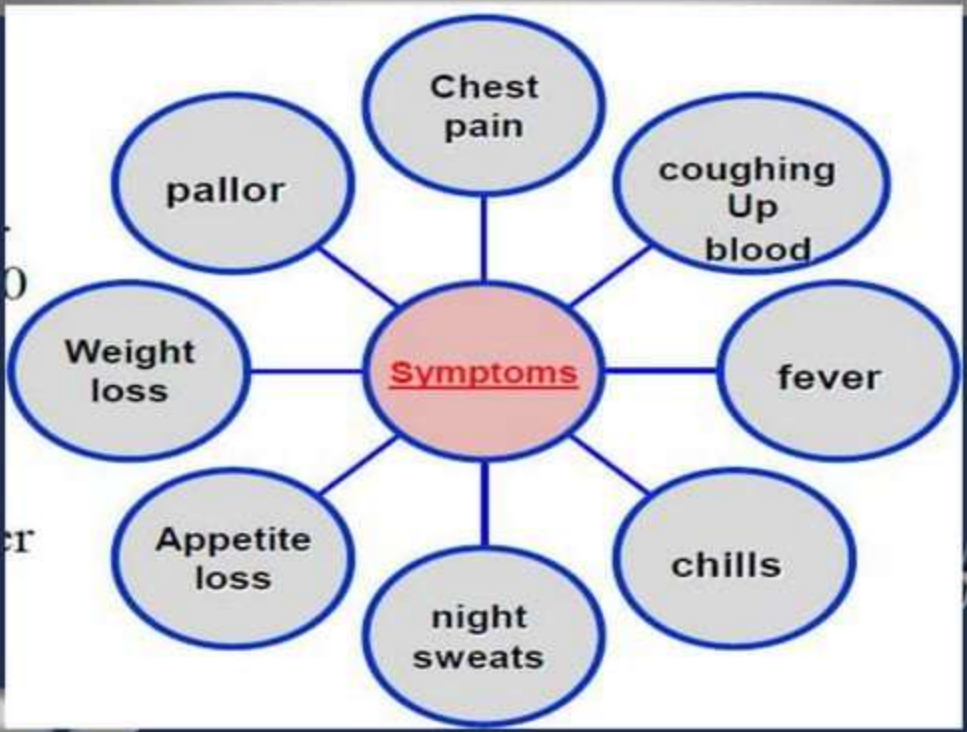
- Inhalation of droplet nuclei.
- Ingestion of contaminated milk.
- Infection after trauma.
- Congenital primary T.B.

Spread of Tuberculosis

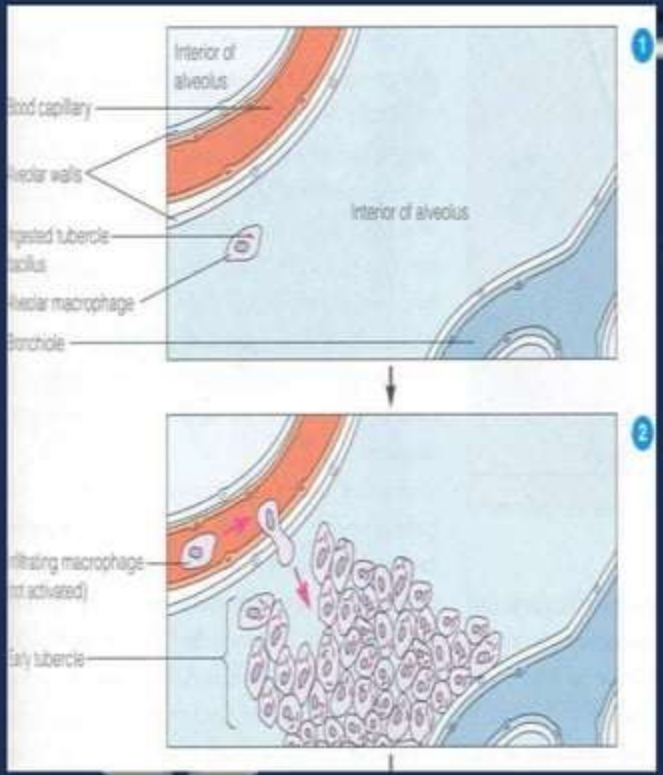




Symptoms and Sings of Tuberculosis



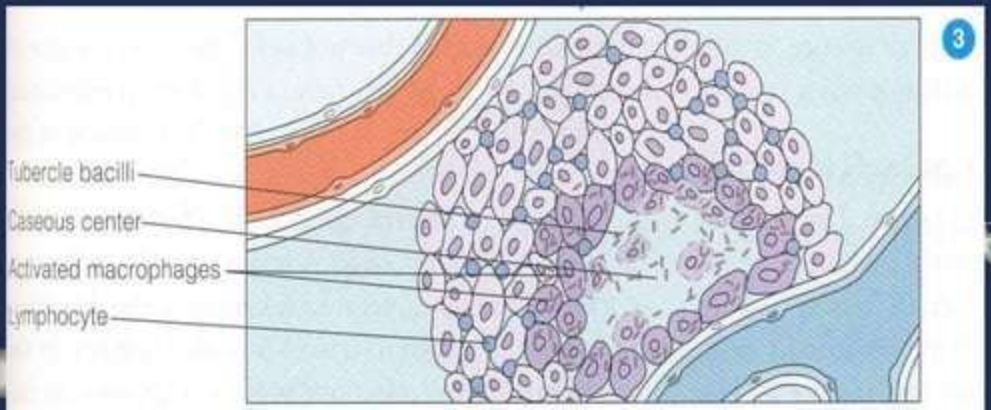
Pathogenesis of Tuberculosis

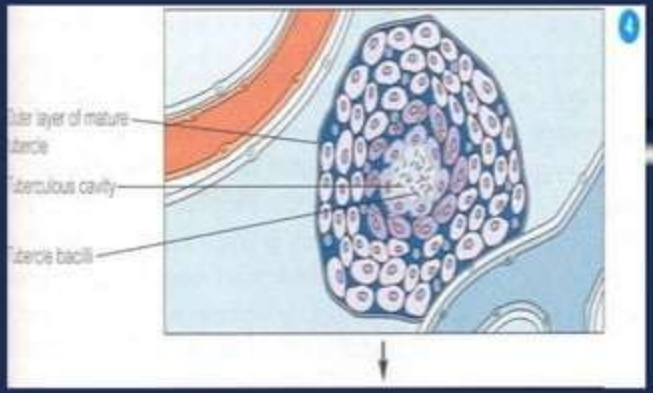


Tubercle bacilli that reach the alveoli of the lung are ingested by macrophages

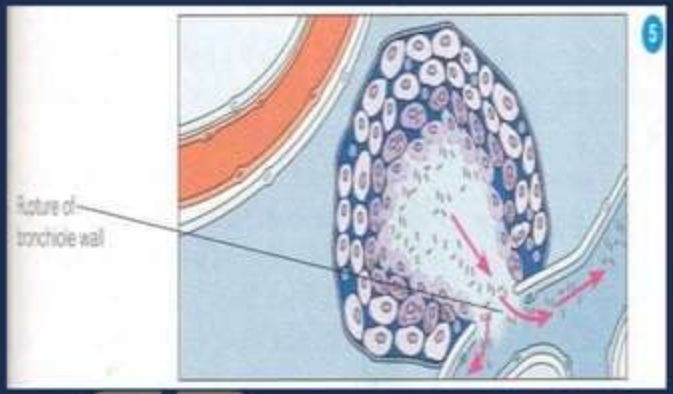
- The organisms multiply and cause a chemotactic response that recruits other macrophages and T cells to the area.
- Macrophages release enzymes and cytokines to start an inflammatory response to wall off the organism (tubercle formation), but the inflammatory response also causes lung damage.

- After a few weeks many of the macrophages die, releasing tubercle bacilli and forming a caseous center in the tubercle.
- In healthy individuals, the disease is usually arrested at this time and the lesions may become calcified (Ghon complexes).
- Tubercle bacilli may remain dormant in the lesion and serve as a basis for later reactivation of the disease.





- When the defenses fail, a mature tubercle may form whereby the caseous center will enlarge and liquify to form a tuberculous cavity where the bacilli multiply outside the macrophages.

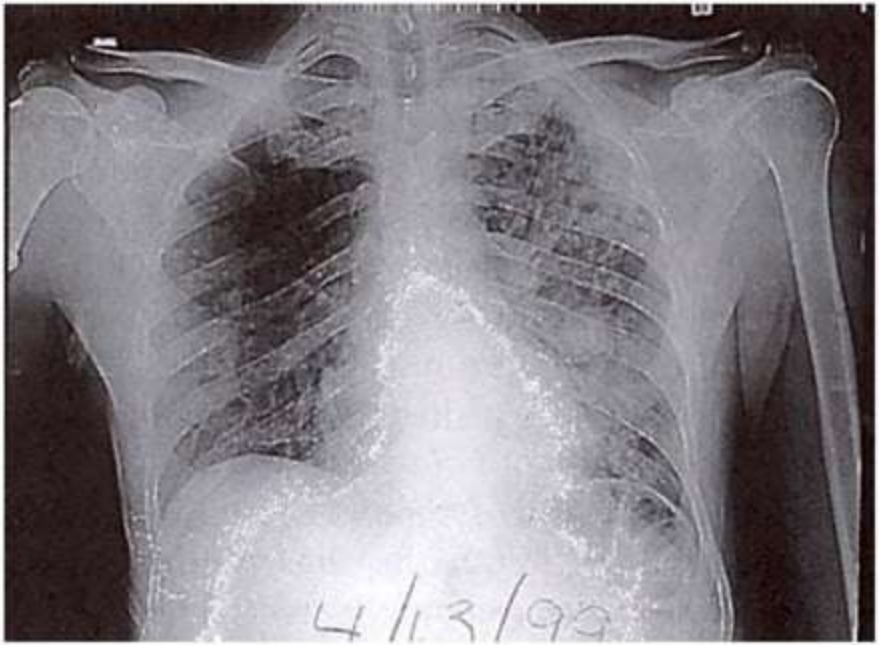


- The tubercle eventually ruptures, releasing tubercle bacilli that can disseminate throughout the lungs and then to the circulatory and lymphatic systems.

Diagnosis

- Tuberculosis is diagnosed definitively by identifying the causative organism in a clinical sample.
- ► When this is not possible , a probable diagnosis may be made using imaging (X-rays or scans) or a tuberculin skin test.
- ►Currently, infection is diagnosed in a non-immunized person by recent techniques as PCR.

Chest X-Ray of Patient with Active Pulmonary Tuberculosis



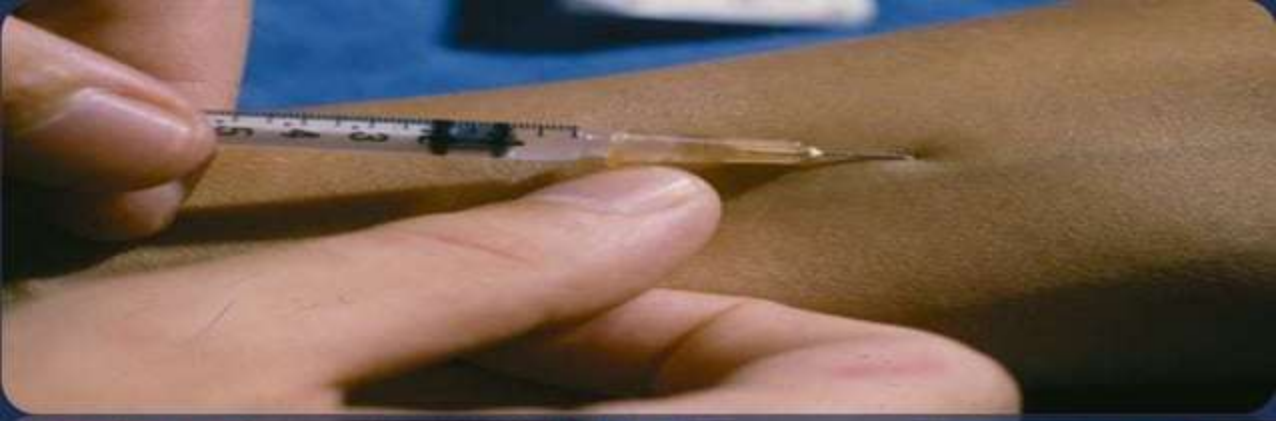
Tuberculin Skin Testing

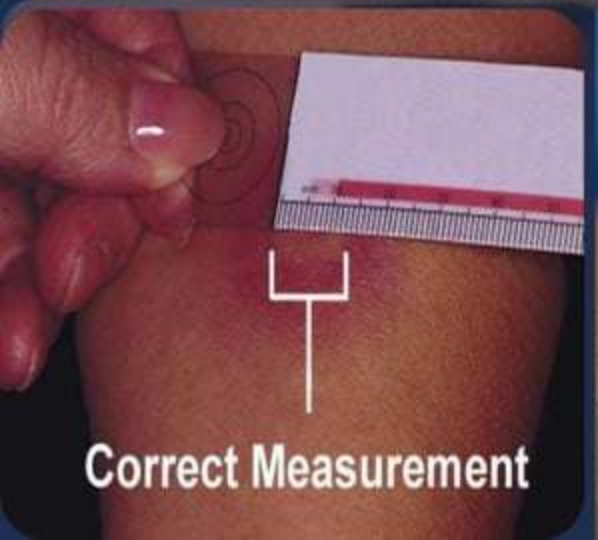
- **Intradermal test** used to detect the state of hypersensitivity to tubercle bacilli.
- **Mantoux tuberculin skin test :**

- is the standard method of determining whether a person is infected with *Mycobacterium tuberculosis*.
- **Purified protein derivative (PPD)** or **Old Tuberculin (OT)** injected intracutaneously into skin of the forearm.
- After 72 hours, the site of injection examined (nodules).
- If nodules diameter greater than 10 mm → positive.

- **Interpretation:**

- **Positive** test indicates previous exposure and carriage of T.B.
- **Negative** tuberculin test excludes infection in suspected persons.
- Tuberculin positive persons may develop reactivation type of T.B.
- Tuberculin negative persons are at risk of gaining new infection.
- Positive test in Children below 5 years of age with no exposure history must be regarded suspicious.



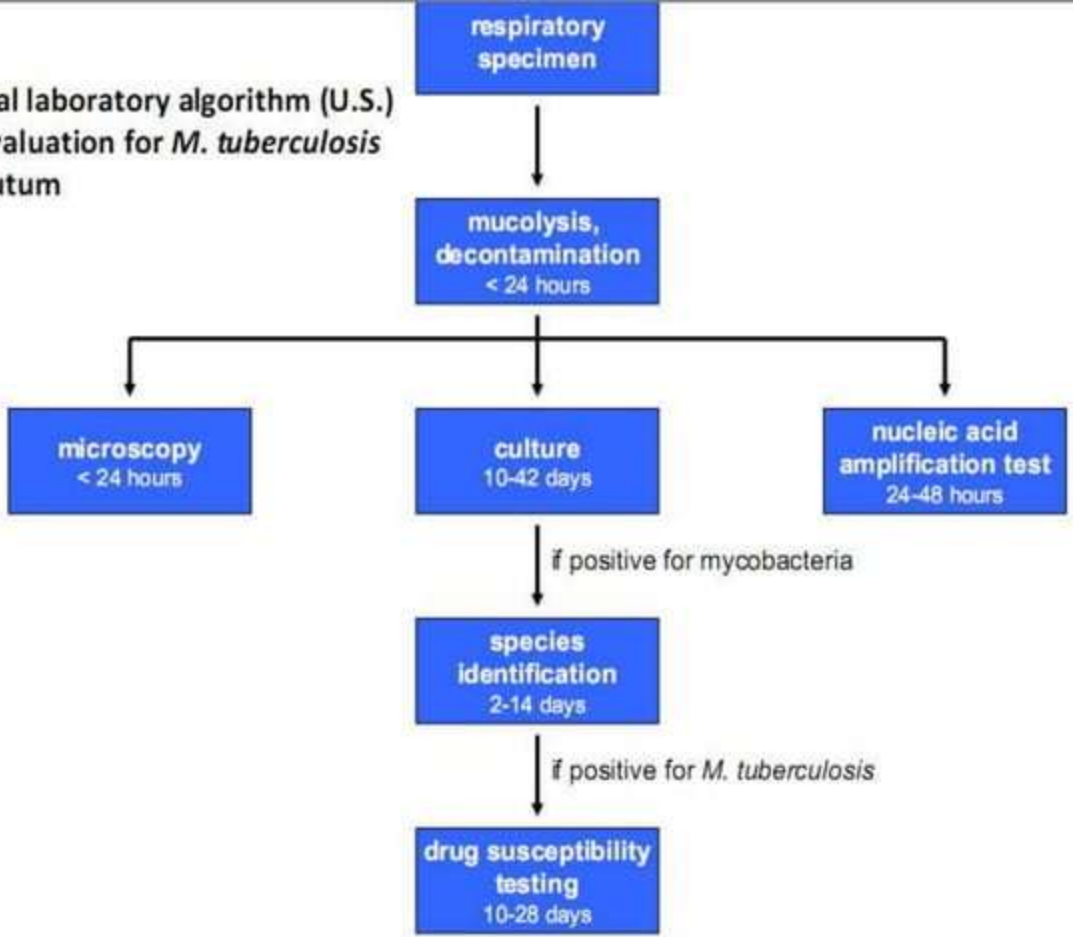

Correct Measurement

Types of specimens:

- Sputum.
- Urine
- Pleural fluid
- Peritoneal fluid
- CSF

Laboratory Diagnosis

Typical laboratory algorithm (U.S.) for evaluation for *M. tuberculosis* in sputum



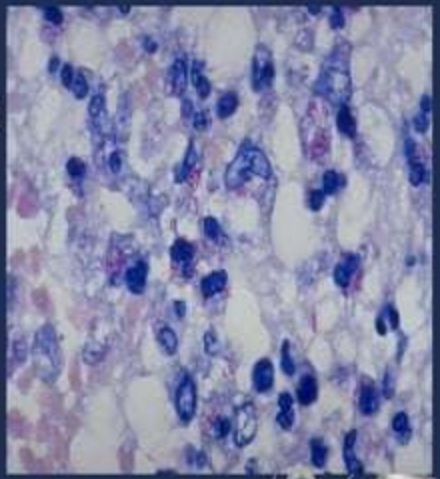
1. Microscopic examination

- Need to contain large number of bacilli (5000-10000 AFB/ml).
- Direct smear stained by Ziehl-Neelsen (hot stain) or Kinyoun's-cold stain
- Direct smear stained by fluorescence dyes.
- Under Microscope:

  slender, straight or curved rod

  Size: 0.4 μm wide and 3–4 μm long

  Arranged singly, in pairs or clusters



Acid fast Bacilli seen as in Florescent Microscope

Acid Fast Bacilli seen in a Sputum

Acid Fast Stain

Sputum smear microscope the only cost-effective tool for diagnosing patients with infectious tuberculosis. and to monitor their progress in treatment.

Sputum smear microscopy is a simple inexpensive,
appropriate technology method which is relatively easy to perform and to read

It yields timely results with a very high sensitivity of detection of tubercle bacilli

Sputum smear microscope also used after cultuering sputum to identify *Mycobacterium tuberculosis*

Principle of ZN Stain - 1

M tuberculosis

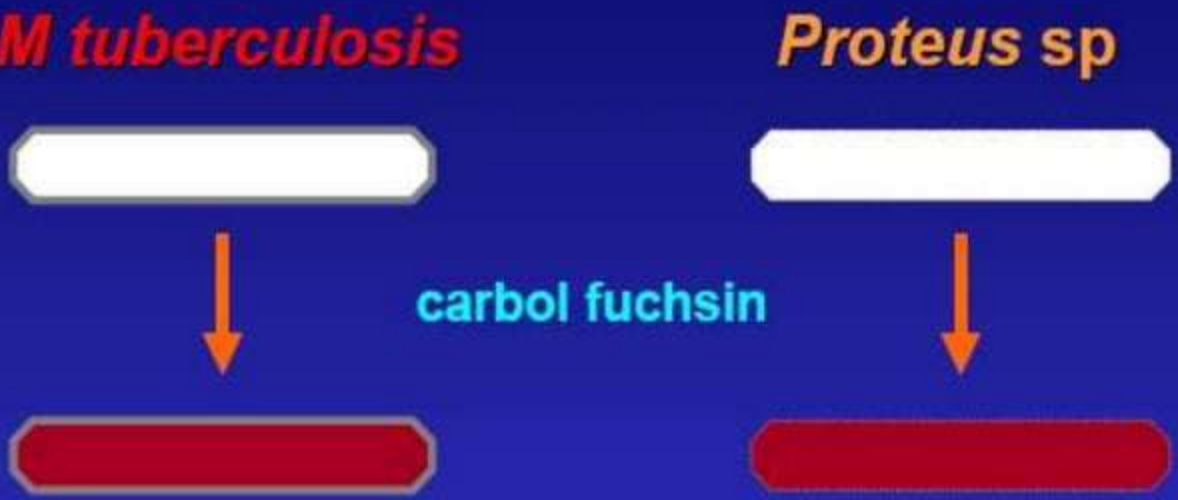
complex waxy cell wall

Proteus sp e.g.

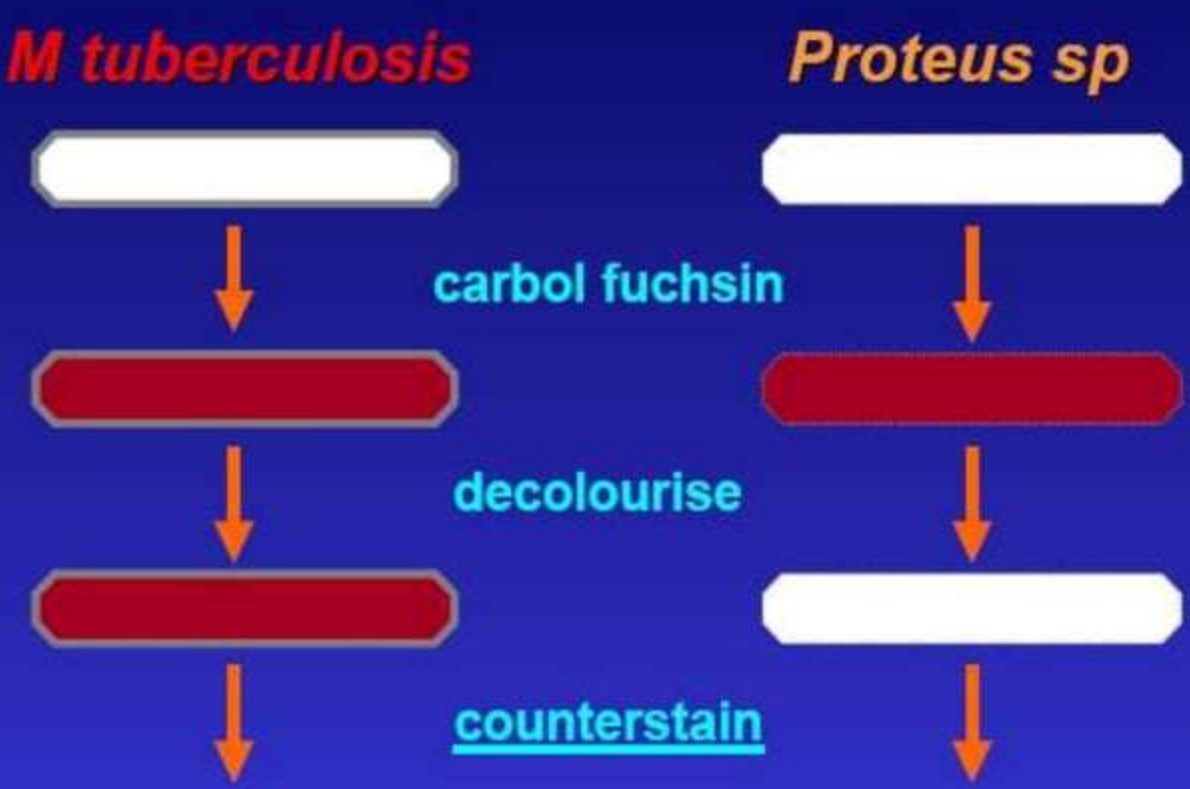
carbol fuchsin

HEAT

Principle of ZN Stain - 2



Principle of ZN Stain - 3



2. Culture

Culture characters

- Obligate aerobes
- Growth generally slow
- The generation time of TB is approximately 12–18 hours (20-30 minutes for Escherichia coli)
- Optimum Temp. 37°C
- Incubated for 6-8 weeks

They can grow on media containing complex substances include:

1. **Dorset egg** and **egg saline media**: contain egg yolk.
2. Selective media:
   a) **Lowenstein Jensen media**: contain malachite green dye to inhibit the unwanted normal flora present in sputum samples.
   b) **Lowenstein Jensen Glycerol media**: Glycerol enhance the growth of *Human Tubercle bacilli (TB).*
   c) **Lowenstein Jensen Pyruvate media**: used for isolation of *M.bovis*

On L J Medium

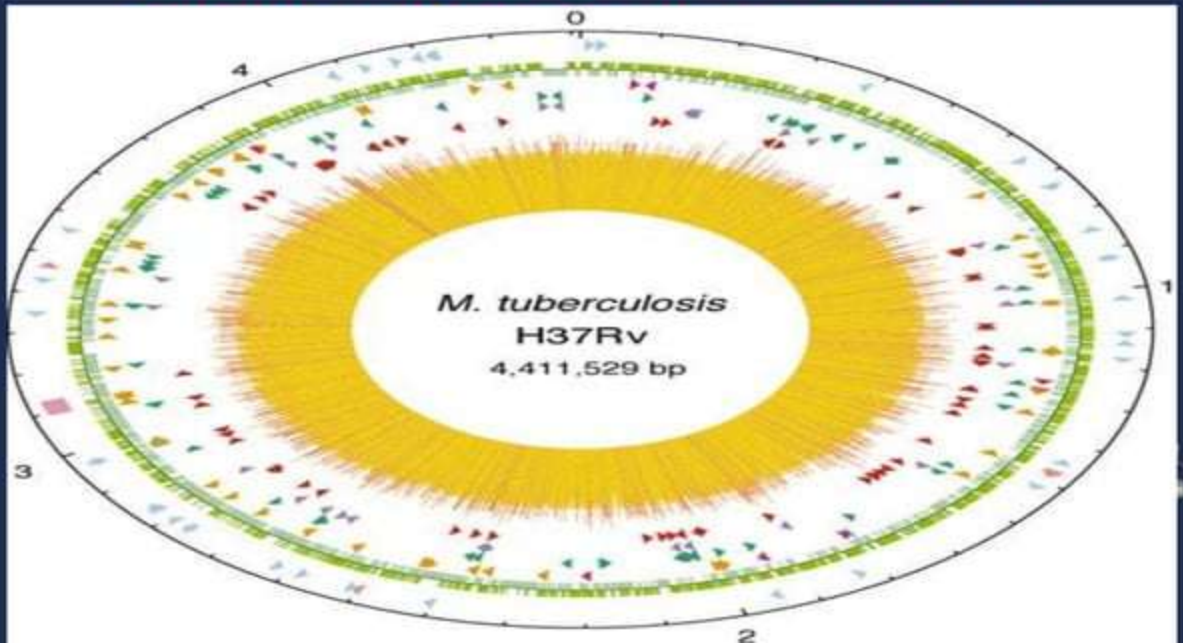
- M.tuberculosis appear dry, rough raised irregular colonies
- Appear wrinkled
- They appear creamy white
- Become yellowish

Eight Week Growth of Mycobacterium tuberculosis on Lowenstein-Jensen Agar

Mycobacterium Tuberculosis on Lewenstien Jensen medium

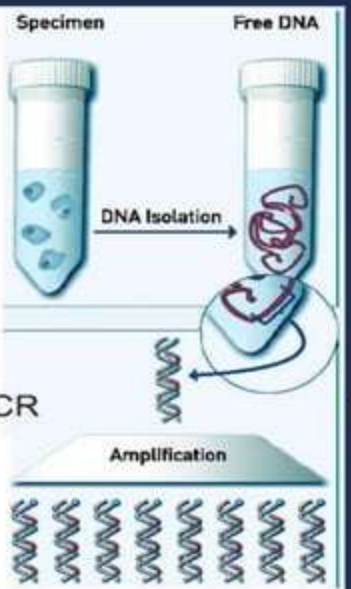
3. Molecular diagnosis

Genome of Mycobacterium tuberculosis

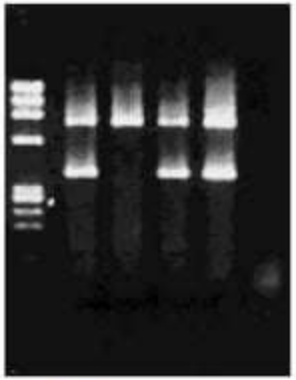


Polymerase Chain Reaction (PCR)

1) DNA extraction



2) Amplification by PCR



T.B. Situation in Palestine

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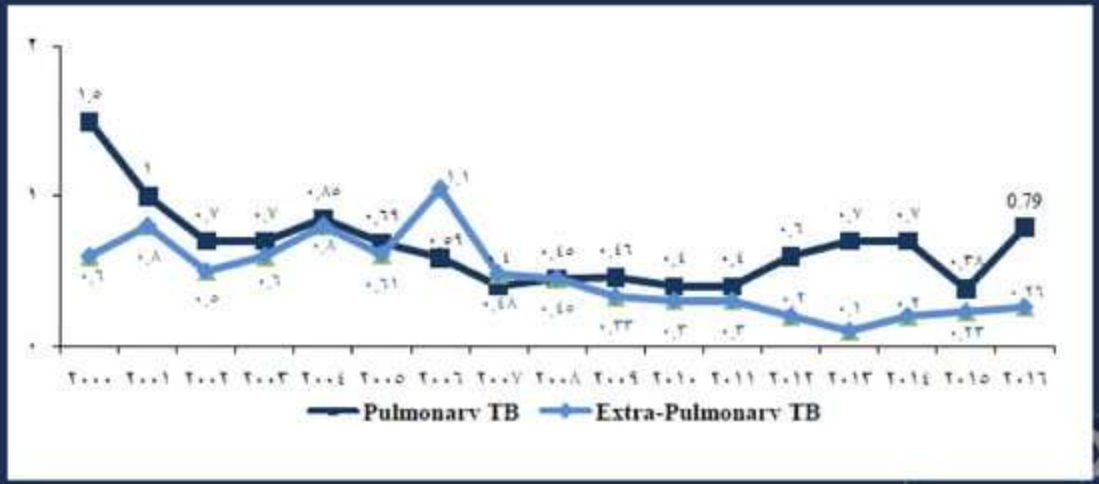
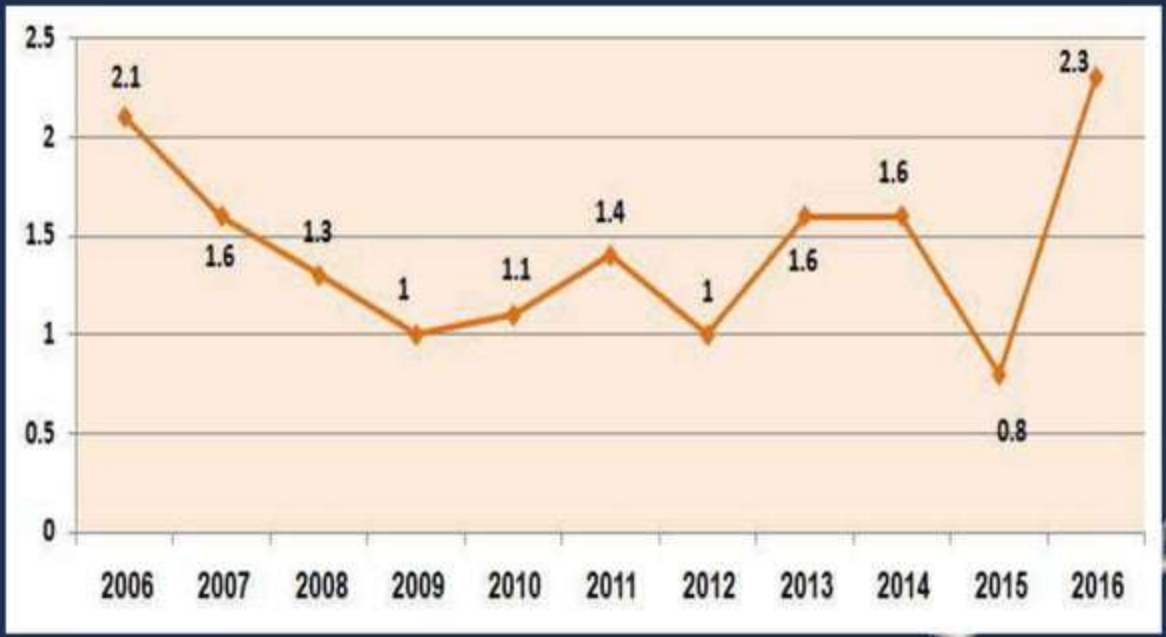
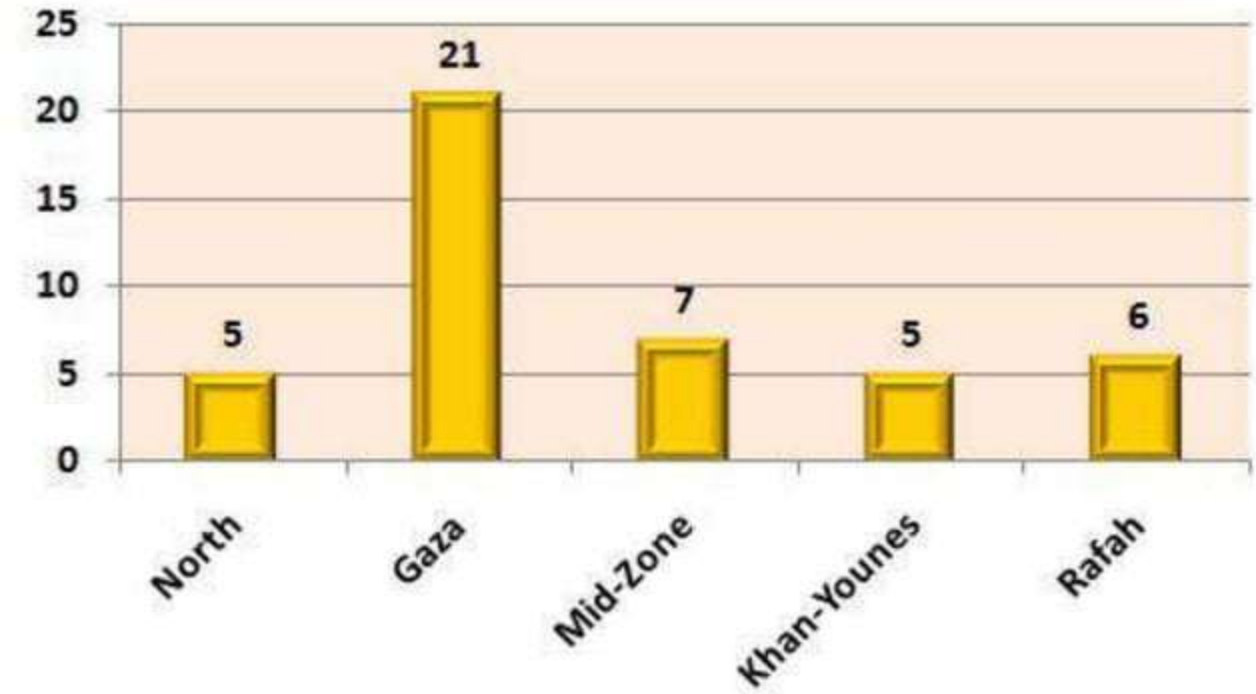
Incidence Rate of Tuberculosis per 100,000 Pop., Palestine 2000- 2016



Annual incidence rate per 100.000 of TB in GS, years 2006-2016



Geographical distribution of TB reported cases in GS, year 2016



Percentage of reported cases of TB by type of disease in GS, 2016