# *Mycobacterium tuberculosis* and Non-tuberculous *Mycobacteria*

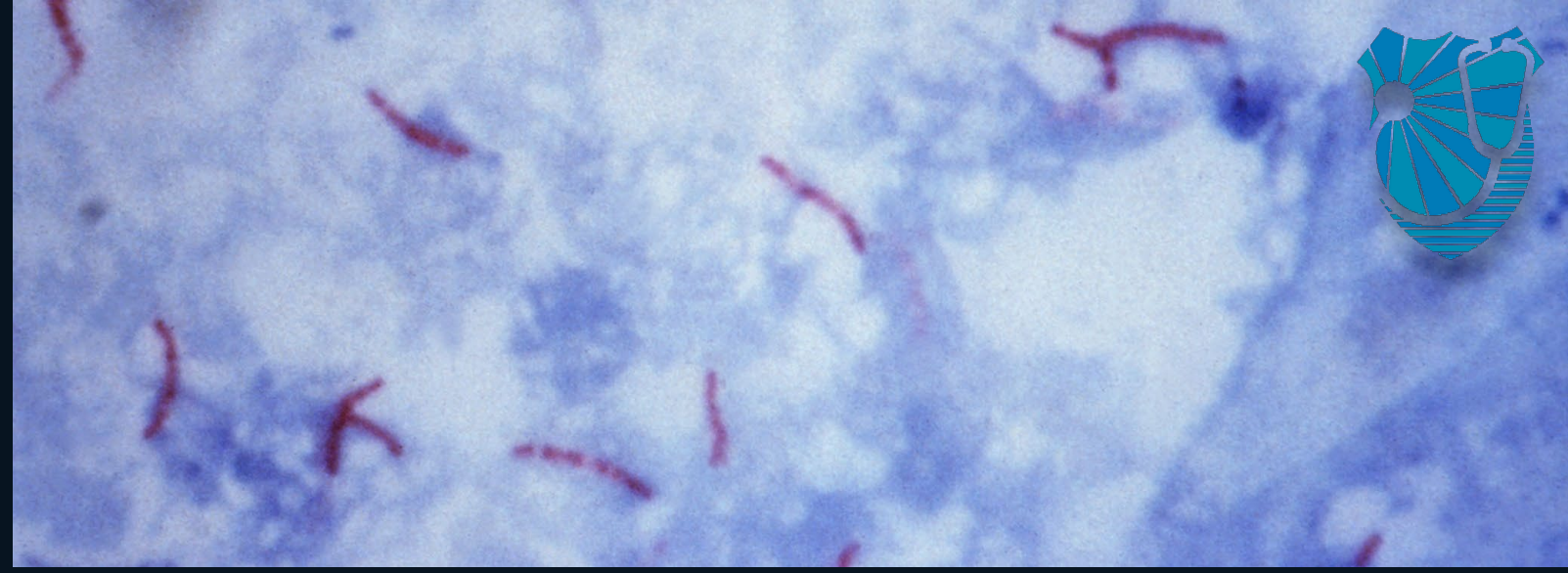
**University of Jordan**

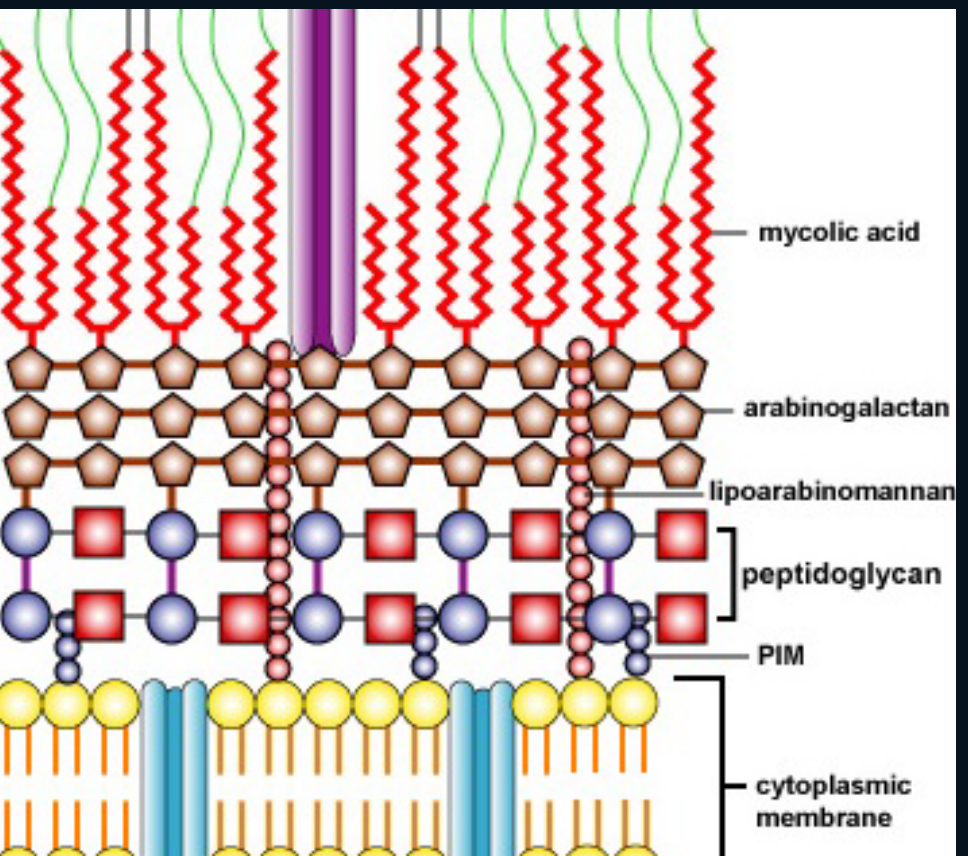
Malik Sallam, M.D., Ph.D.

School of Medicine

Department of Pathology, Microbiology and Forensic Medicine

# Biology of *Mycobacteria*



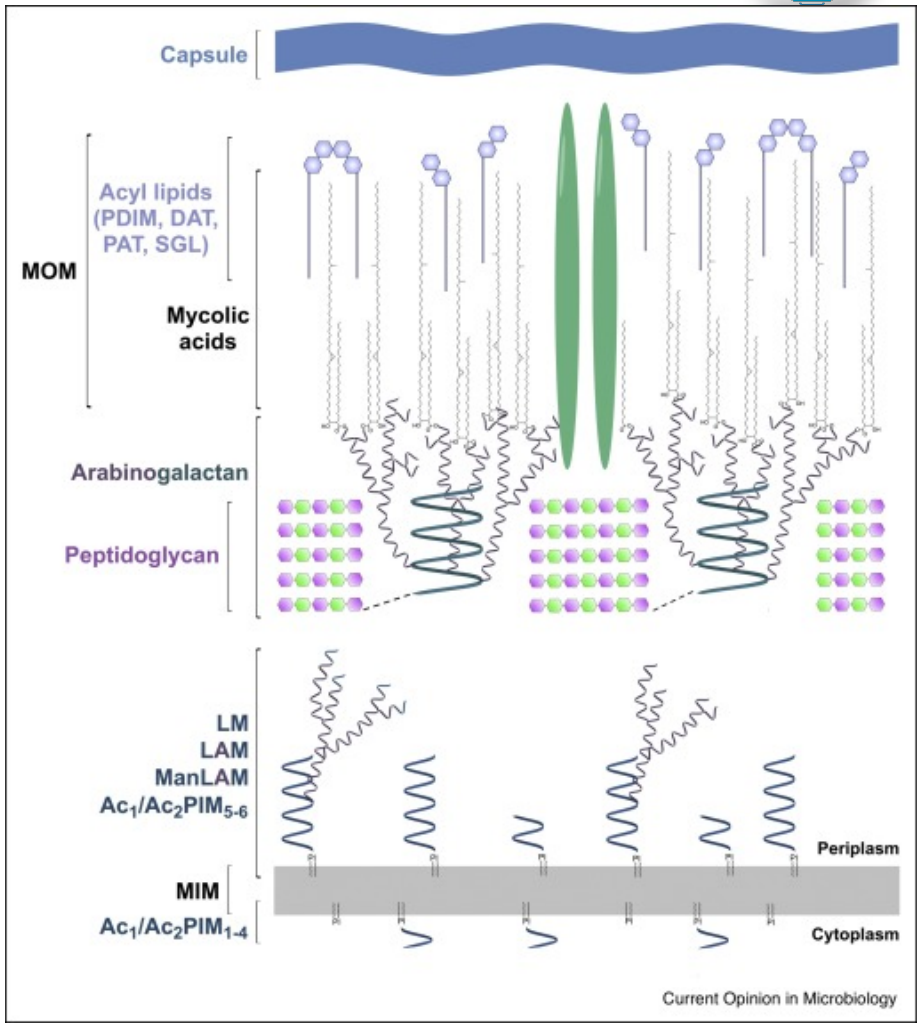



- **Acid-fast bacilli (AFB)**, obligate aerobes
- **Slow growth**: 15-20 hours per replication
- Non-spore forming but **extremely resistant**
- Cell wall is the most defining feature, as they have thick mycolic acids (long-chain fatty acids) and an arabinogalactan layer composed of peptidoglycan core that gives resistance to drying, disinfectants.
- **Ability to persist intracellularly**

# Mycobacterial Cell Wall

- The MTB cell envelope has multiple layers that mediate **chronicity** and **intracellular survival**.

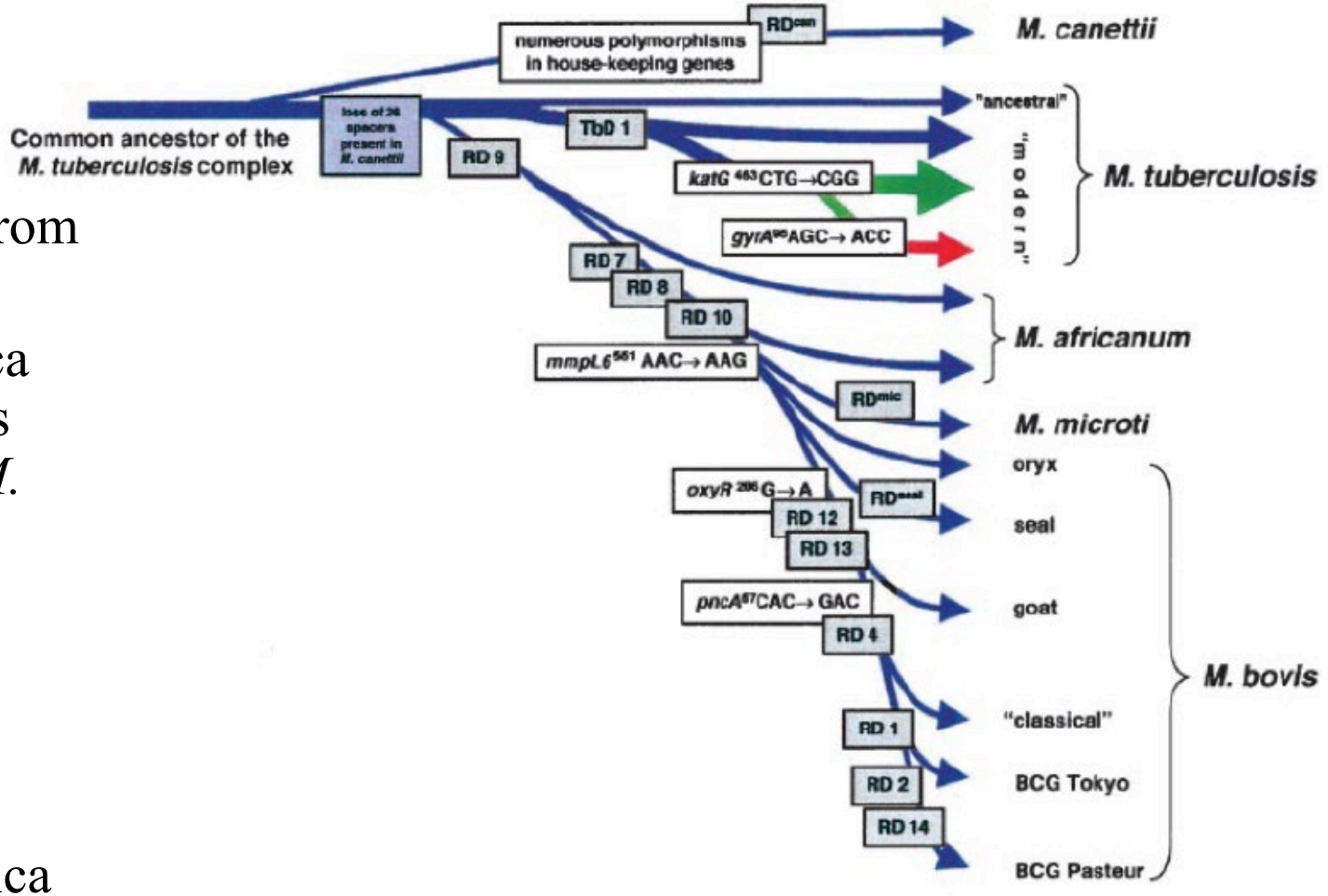
A. Inner cell membrane, periplasmic space
B. Peptidoglycan providing structural rigidity
C. Arabinogalactan links peptidoglycan to mycolic acids
D. Mycolic acid layer composed of waxy hydrophobic barrier. Key to acid-fastness. Impermeable to many antibiotics. Surface lipids & glycolipids: **Cord factor (trehalose dimycolate)**. Toxic to macrophages. Drives granuloma formation.
E. Capsule: mediate interactions with phagocytes that favor bacterial survival




Source: Abrahams KA, Besra GS. Synthesis and recycling of the mycobacterial cell envelope. Curr Opin Microbiol. 2021 Apr;60:58-65. doi: 10.1016/j.mib.2021.01.012. A schematic representation of the mycobacterial cell wall, highlighting the key features. Abbreviations: mycobacterial inner membrane (MIM), mycobacterial outer membrane (MOM), phosphatidyl-myo-inositol mannosides, (PIMs, with acylation sites Ac1/Ac2), lipomannan (LM), lipoarabinomannan (LAM), mannosylated lipoarabinomannan (ManLAM), diacyl-trehalose (DAT), polyacyl-trehalose (PAT), phthiocerol dimycocerosate (PDIM), and sulfoglycolipid (SGL). An outer membrane protein has been included (green) to depict how solutes traverse the hydrophobic layer.

# *Mycobacterium tuberculosis* Complex (MTBC)

- *M. tuberculosis*: the main human pathogen
- *M. bovis*: Causes zoonotic TB. Transmitted via unpasteurized milk, direct animal contact; BCG derived from attenuated strain
- *M. africanum*: Endemic in West Africa and causes human TB. Generally, less transmissible and less virulent than *M. tuberculosis*
- *M. microti*: Primarily infects rodents
- *M. caprae*: Infects goats and cattle. Occasional zoonotic transmission to humans
- *M. canetti*: Found mainly in East Africa




Scheme of the proposed evolutionary pathway of the tubercle bacilli illustrating successive loss of DNA in certain lineages (gray boxes).

# Non-tuberculous *Mycobacteria* (NTM)

- Environmental organisms, do not spread person-to-person.
- Major groups:
- **Slow growers** (take more than 2 weeks to grow): *M. avium* complex (MAC), *M. kansasii*, *M. xenopi*
- **Rapid growers** (grow in liquid media within 7 days): *M. abscessus*, *M. fortuitum*, *M. chelonae*
- Clinical significance: Pulmonary disease in COPD/bronchiectasis. Disseminated disease in immunocompromised. High-level of intrinsic drug resistance

## NTM vs. *Mycobacterium tuberculosis*: Key distinctions

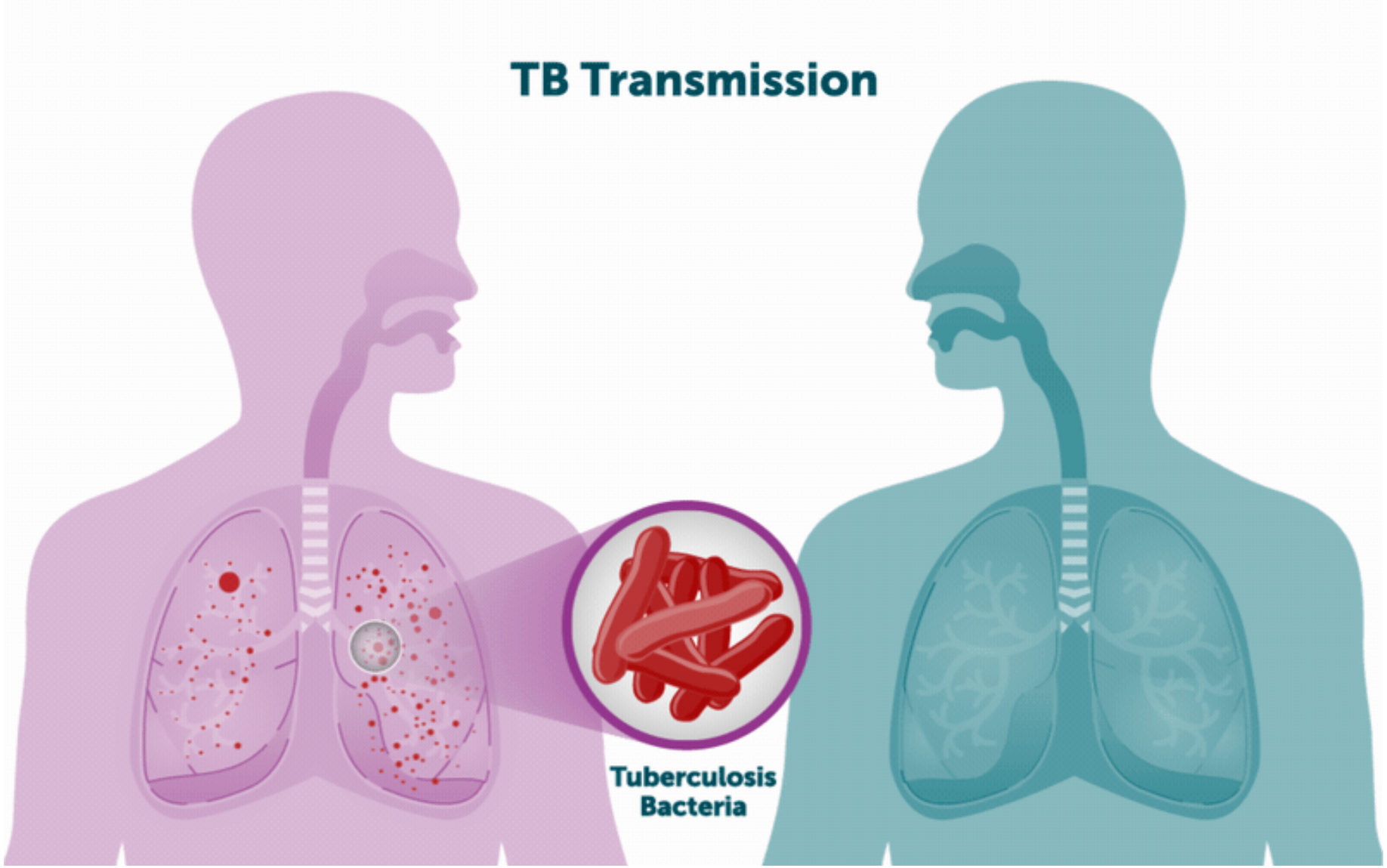
| NTM | *Mycobacterium tuberculosis* |
|---|---|
| Not obligate pathogens – normally live free in the environment[1] | Obligate pathogens: require host[1] |
| Low virulence: not usually pathogenic in the absence of predisposing conditions[2,3] | Pathogenic[3,6] |
| Human-to-human transmission extremely rare, but some evidence of this in the cystic fibrosis community[4] | Human-to-human transmission[3] |
| Infection rates increasing, especially in developed countries[6] | Infection rates decreasing, especially in developed countries[6] |
| Large heterogeneous group of species[6] | *Mycobacterium tuberculosis* complex contains small group of closely related subspecies[6,7] |

NTM and *Mycobacterium tuberculosis* differ in terms of pathogenicity, infection rates and transmission routes

NTM, non-tuberculous mycobacteria.
1. Primm TP, et al. Clin Microbiol Rev 2004;17:98-106; 2. Tortoli E. Clin Microbiol Infect 2009;15:906-10;3. Tortoli E. FEMS Immunol Med Microbiol 2006;48:159-78;4. MacShane PJ, Glassroth J. Chest 2015;148:1617-27; 5. Brode SK, et al. Int J Tuberc Lung Dis 2014;18:1370-7;6. Van Soolingen D. J Intern Med 2001;249:1-26; 7. Cole ST. Microbiology 2002;148:2019-28.

# MTBC Immunopathogenesis

- **Early TB infection**
- Inhalation of aerosolized droplet nuclei containing *Mycobacterium tuberculosis*
- Bacilli reach terminal bronchioles and alveoli



# MTBC Immunopathogenesis

- **Initial human-MTB interaction**
- Mediated via PRRs TLRs 2, 4, 9, C-Type Lectin Receptors (CLRs) like Dectin-1, Mannose Receptor, DC-SIGN, and NOD-Like Receptors (NLRs), that recognize MTB components to trigger innate immunity.
- **MTB is a facultative intracellular pathogen**

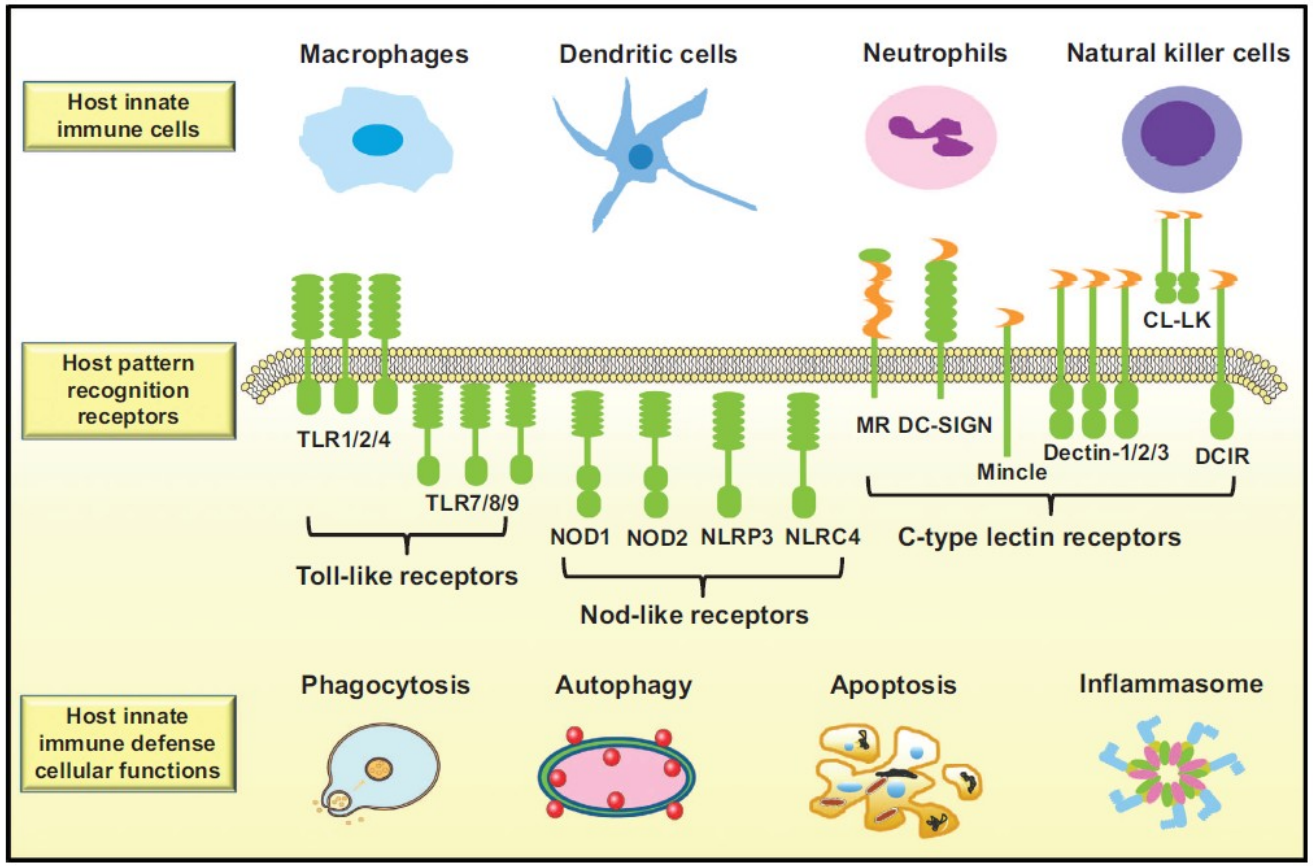
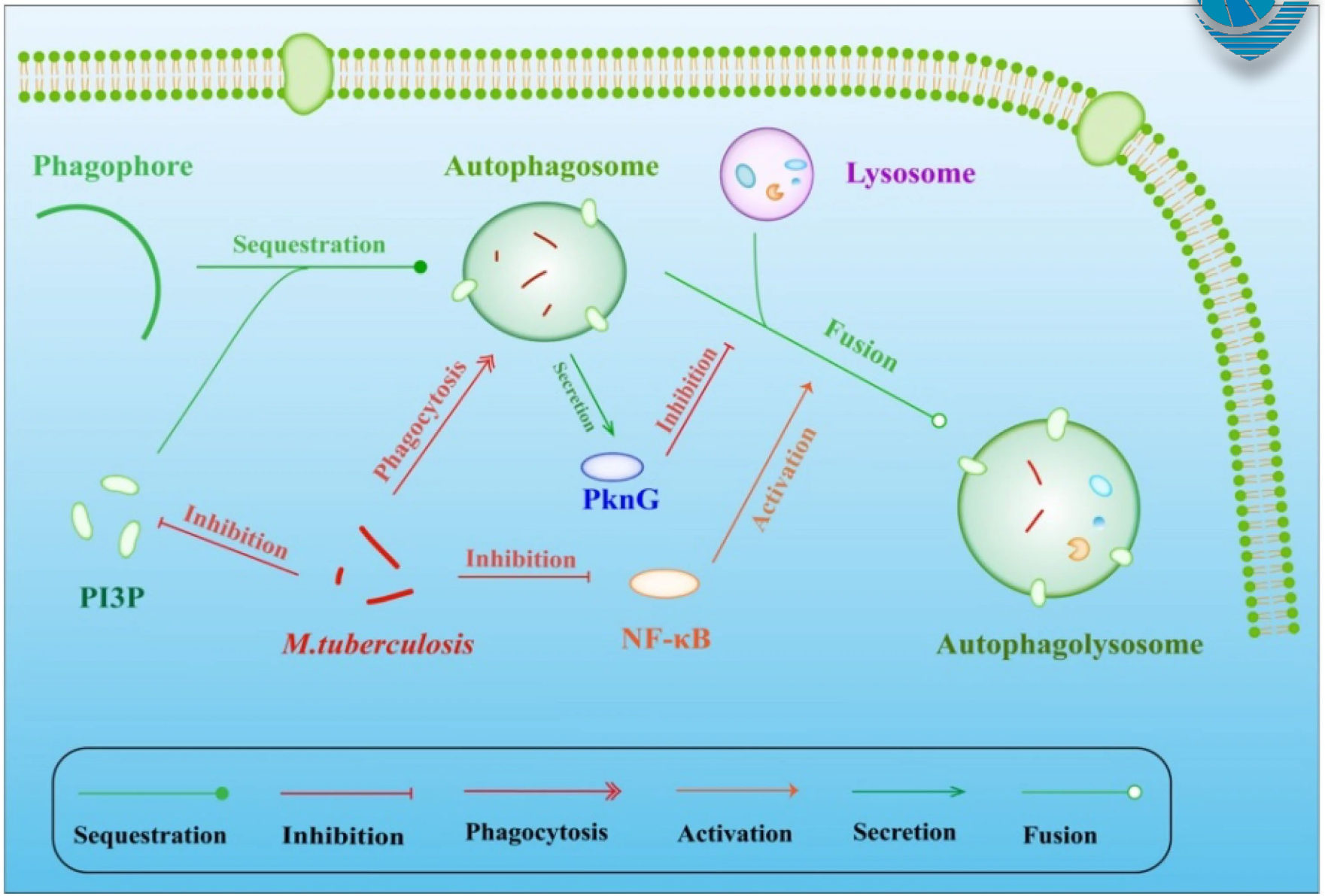



**Figure 1** Major host immune cells, pattern recognition receptors and cellular functions involved in innate immune defense against Mtb. Mtb mainly infects innate immune cells, including macrophages, dendritic cells, neutrophils and natural killer cells. Those immune cells recognize Mtb through various pattern recognition receptors, including Toll-like receptors (such as TLR1, TLR2, TLR4, TLR7, TLR8 and TLR9), Nod-like receptors (such as NOD1, NOD2, NLRP3 and NLRC4) and C-type lectin receptors (such as MR, DC-SIGN, Mincle, Dectin-1 and Dectin-2, Dectin-3, CL-LK and DCIR). During Mtb infection, the host orchestrates signaling from those PRRs and launches a variety of cellular functions, such as phagocytosis, autophagy, apoptosis and inflammasome activation, to control or eliminate Mtb.

Source: Liu, C., Liu, H. & Ge, B. Innate immunity in tuberculosis: host defense vs pathogen evasion. Cell Mol Immunol 14, 963–975 (2017).

# MTBC Immunopathogenesis

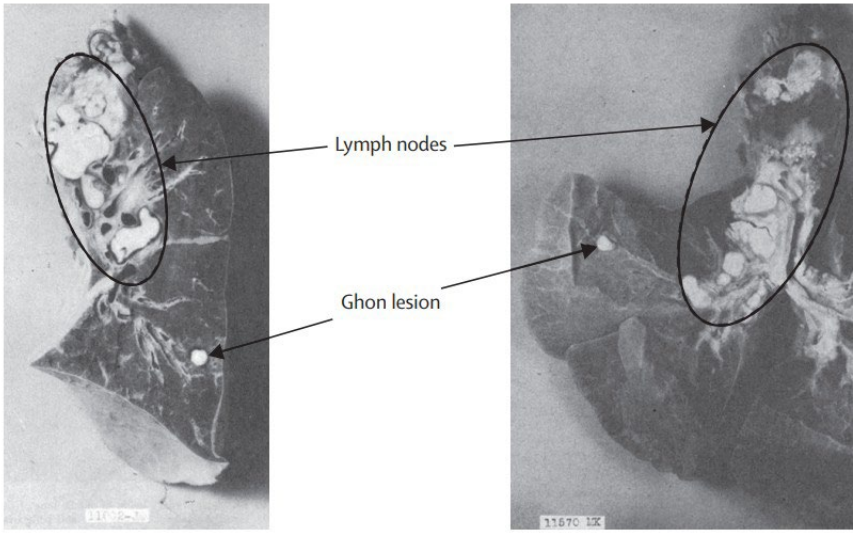
- **Intracellular MTBC survival mechanisms:**
- Inhibits phagosome-lysosome fusion
- Resists oxidative stress
- Alters phagosomal pH preventing acidification



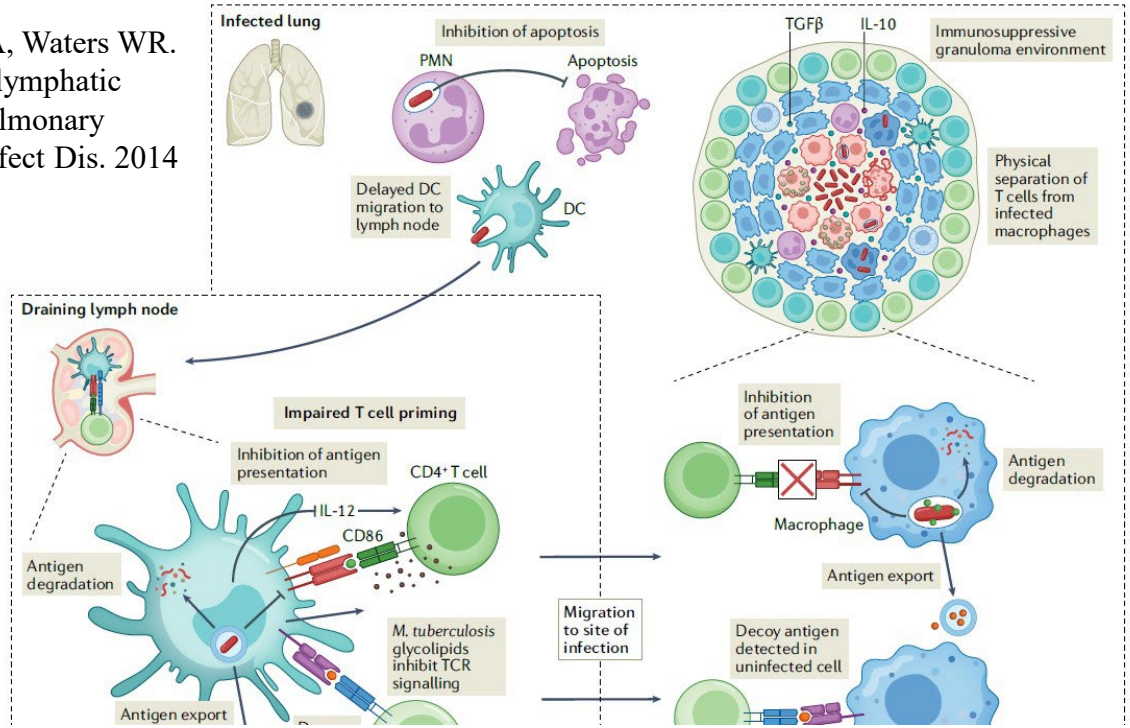

Source: Zhai, W.; Wu, F.; Zhang, Y.; Fu, Y.; Liu, Z. The Immune Escape Mechanisms of Mycobacterium Tuberculosis. Int. J. Mol. Sci. 2019, 20, 340.

# MTBC Immunopathogenesis

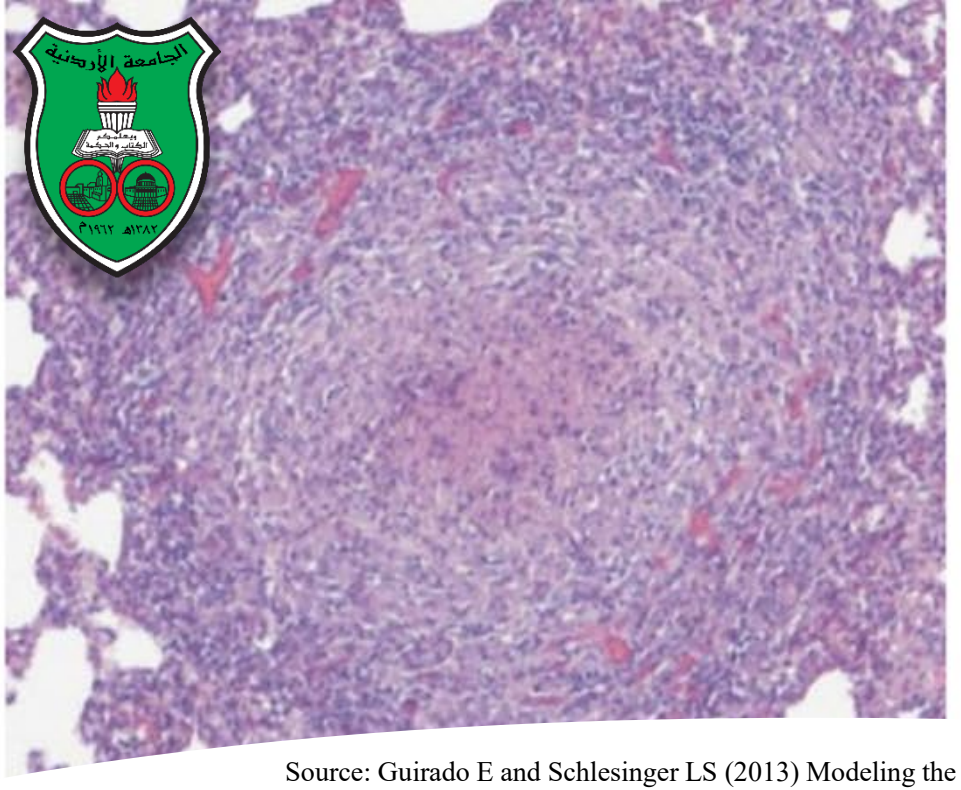
- Cell wall component lipoarabinomannan (LAM): Suppresses IL-1β production, reduces TNF-α signaling, and impairs macrophage activation. This delays effective adaptive immune response (2-4 weeks)
- Slow intracellular replication within macrophages and formation of a primary pulmonary lesion (Ghon focus). This is followed by spread to regional lymph nodes forming Ghon complex (pulmonary + draining LN)

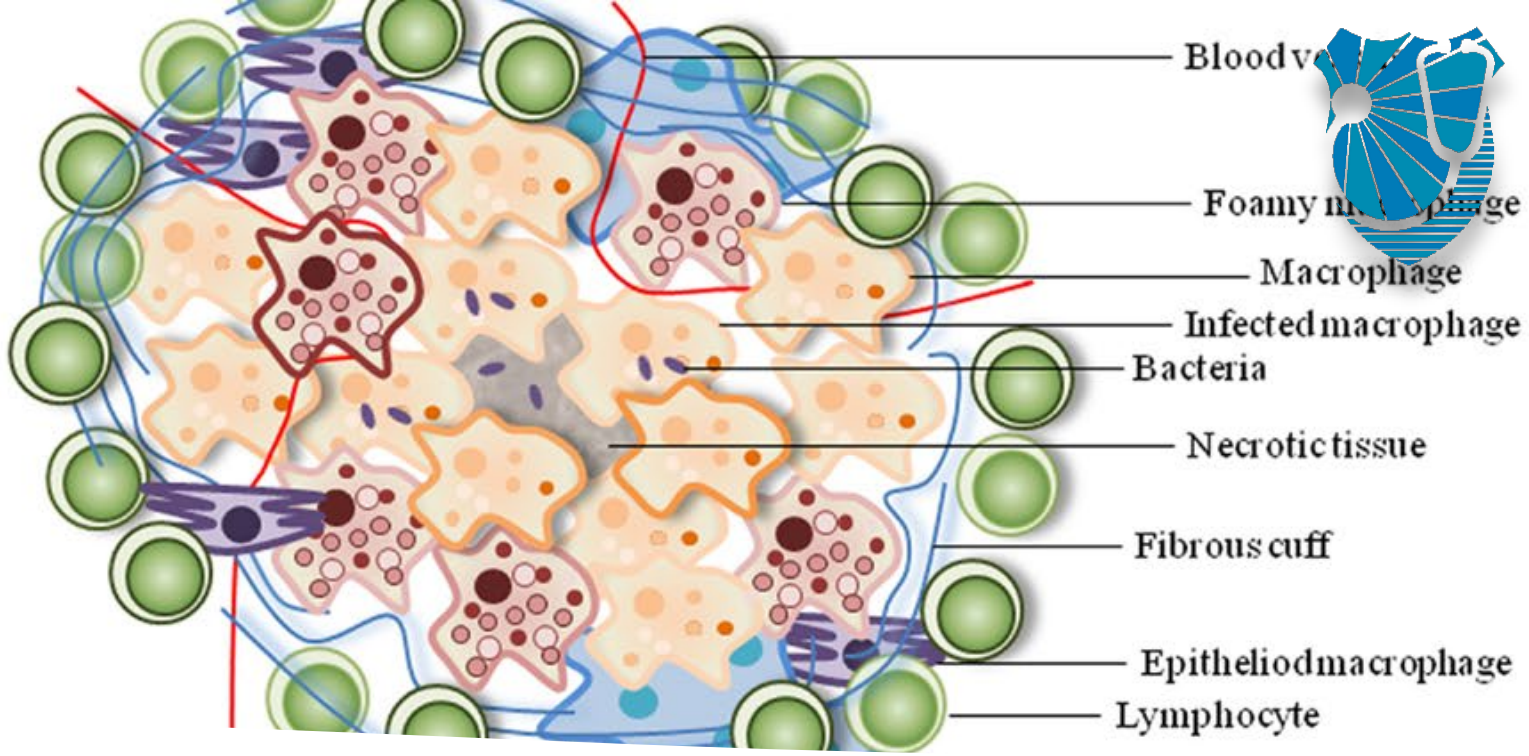



Source: Behr MA, Waters WR. Is tuberculosis a lymphatic disease with a pulmonary portal? Lancet Infect Dis. 2014 Mar;14(3):250-5.




Source: Chandra, P., Grigsby, S.J. & Philips, J.A. Immune evasion and provocation by Mycobacterium tuberculosis. Nat Rev Microbiol 20, 750–766 (2022).






Source: Guirado E and Schlesinger LS (2013) Modeling the Mycobacterium tuberculosis granuloma – the critical battlefield in host immunity and disease. Front. Immunol. 4:98.

## MTBC Determinants of Clinical Outcome

- **Effective Immune Response = Latent TB**
- **Robust Th1-mediated cellular immunity**. IFN-γ → activates macrophage resulting in killing of intracellular MTB; TNF-α → essential for granuloma formation and maintenance. This results in containment of infection, formation of stable granulomas, and latent tuberculosis infection (LTBI)
- **Ineffective Immune Response = Active TB.** Weak, delayed, or dysregulated immune response caused by immunosuppression (e.g., HIV, malnutrition, diabetes), extremes of age. Failure of granuloma integrity, uncontrolled bacterial replication, and progression to active TB

# MTBC Immunopathogenesis
# Determinants of Clinical Outcome

| State | How the patient looks | Immune control status | Terminology |
|---|---|---|---|
| 0 | Completely well | Innate immunity clears MTB | Uninfected, Innate clearance, Early elimination, Resistance to infection |
| I | Well, immune memory present | Adaptive immunity clears MTB | LTBI reverter, Resister, Transient infection, Adaptive clearance |
| II | Well, asymptomatic | Stable immune containment | Latent TB infection (LTBI), Quiescent infection, Immunological equilibrium |
| III | Well or vague symptoms | Early loss of immune control | Incipient TB, Progressor, Subclinical active TB |
| IV | Asymptomatic but test-positive | Bacillary replication without symptoms | Subclinical TB, Asymptomatic bacteriologically positive TB |
| V | Symptomatic (cough, fever, weight loss) | Failure of immune containment | Active TB, Clinical TB, Pulmonary or extrapulmonary TB |
| VI | Very ill, highly infectious | Advanced immune breakdown | Cavitary TB, Disseminated TB, Severe TB |
| VII | Well, prior TB history | Post-treatment immune state | Past TB, Treated TB, Old TB |

Modified from: Zaidi SMA, Coussens AK, Seddon JA, Kredo T, Warner D, Houben RMGJ, Esmail H. Beyond latent and active tuberculosis: a scoping review of conceptual frameworks. EClinicalMedicine. 2023 Nov 17;66:102332.

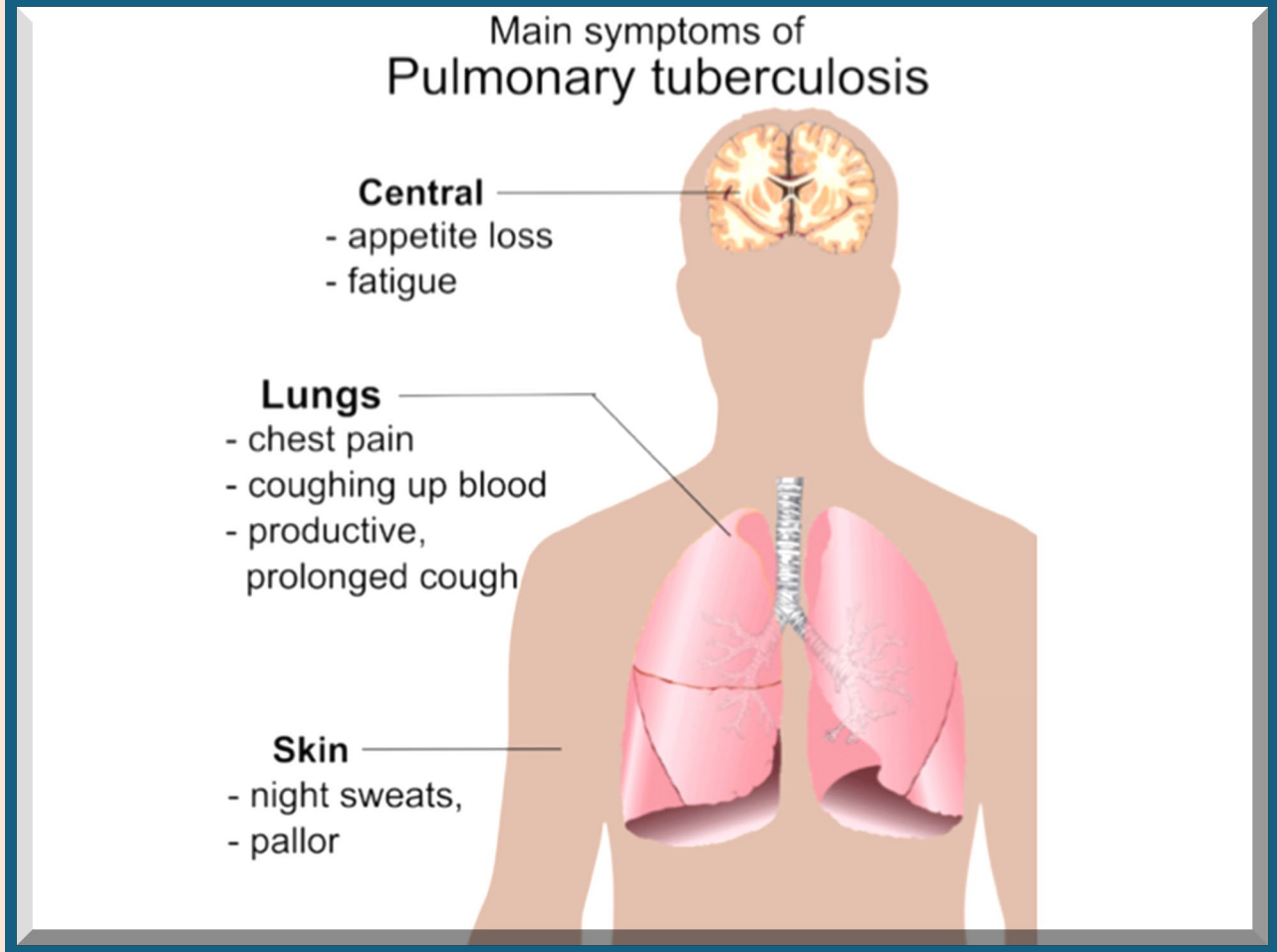
# MTBC Immunopathogenesis
# Determinants of Clinical Outcome

| State | Should it be treated? | How it is diagnosed? |
|---|---|---|
| 0 | No | No tests positive; TST/IGRA negative |
| I | No | Prior exposure history; TST/IGRA may revert to negative |
| II | Yes (preventive therapy in selected patients) | TST or IGRA positive, normal CXR, asymptomatic |
| III | Yes (early intervention) | IGRA/TST positive, subtle CXR changes, biomarkers, early microbiology often negative |
| IV | Yes (full TB treatment) | Sputum NAAT/culture positive, asymptomatic, CXR abnormal |
| V | Yes (mandatory) | Symptoms + abnormal CXR + microbiology (smear/NAAT/culture) |
| VI | Yes (urgent, intensive) | Smear-positive, cavitary disease, disseminated findings |
| VII | No (unless relapse/reinfection) | Past TB history, residual CXR changes, inactive disease |

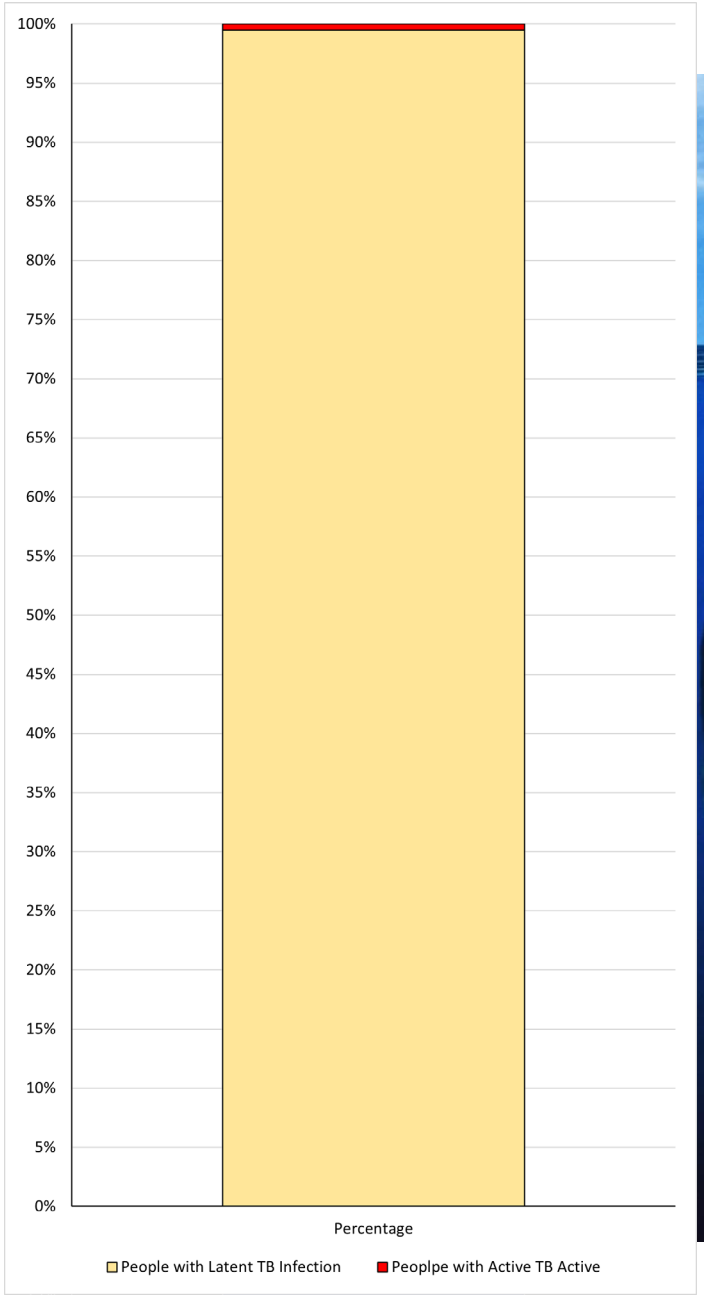
Modified from: Zaidi SMA, Coussens AK, Seddon JA, Kredo T, Warner D, Houben RMGJ, Esmail H. Beyond latent and active tuberculosis: a scoping review of conceptual frameworks. EClinicalMedicine. 2023 Nov 17;66:102332.

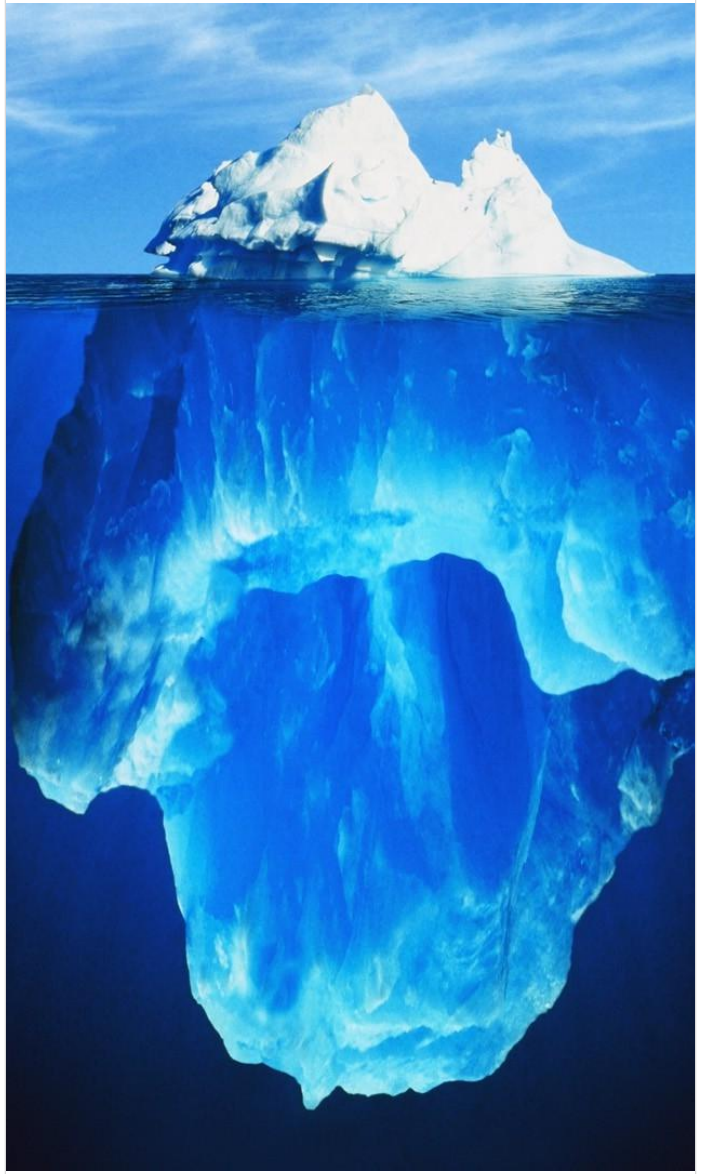
# Respiratory TB infection features

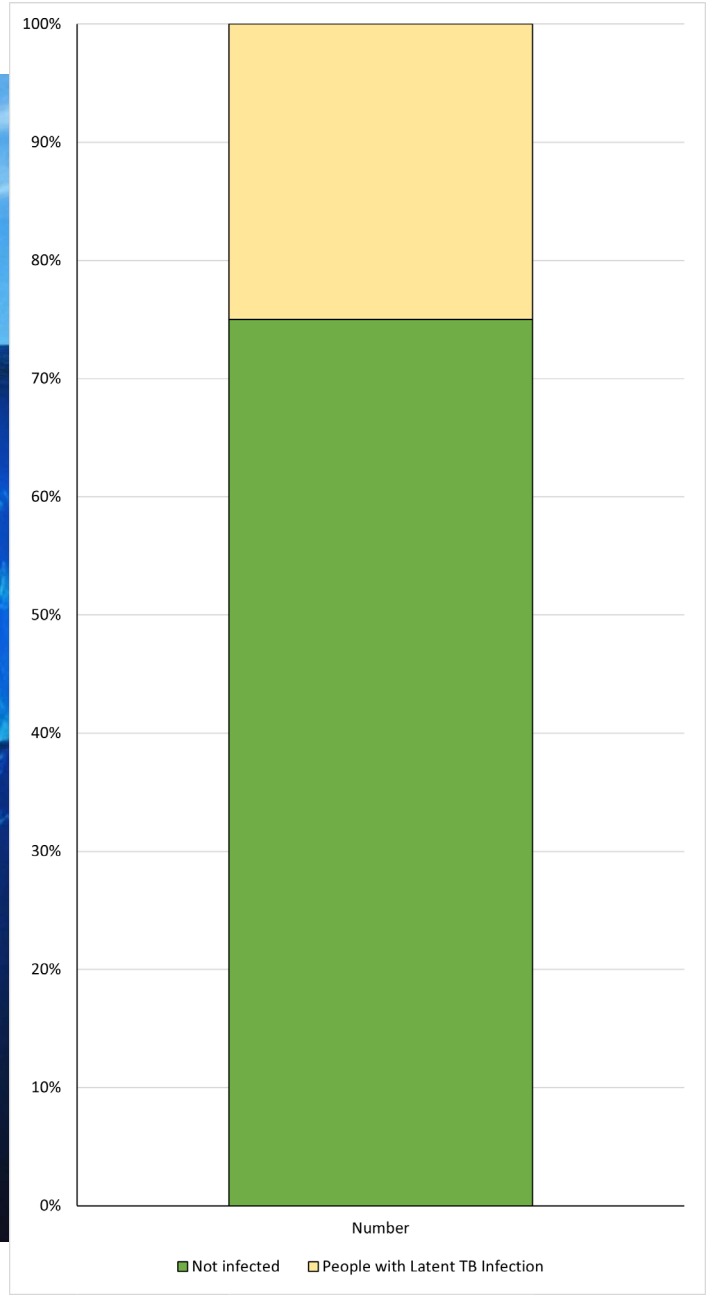
- Onset is gradual and insidious (weeks-months). Can be more acute in young children and immunocompromised patients (e.g., HIV)
- Dominant symptoms include persistent, non-remitting cough (in 90-95% of cases). May be dry cough initially followed by productive cough with or without **hemoptysis**. Systemic symptoms include **fever** (75%), **night sweats** (50%), **weight loss** (60%). Fatigue, anorexia are common. Chest pain, dyspnea in advanced disease.
- 70-75% pulmonary TB. About 20% exclusively extrapulmonary TB (EPTB). 5-10% combined pulmonary + extrapulmonary disease



# Latent vs Active TB

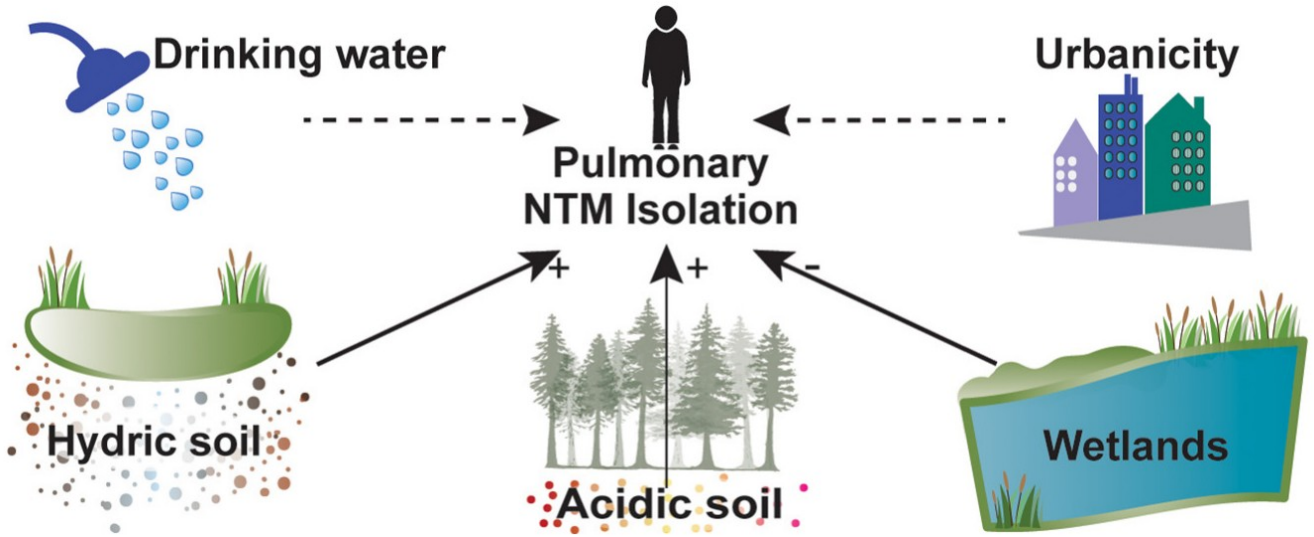




# NTM Immunopathogenesis

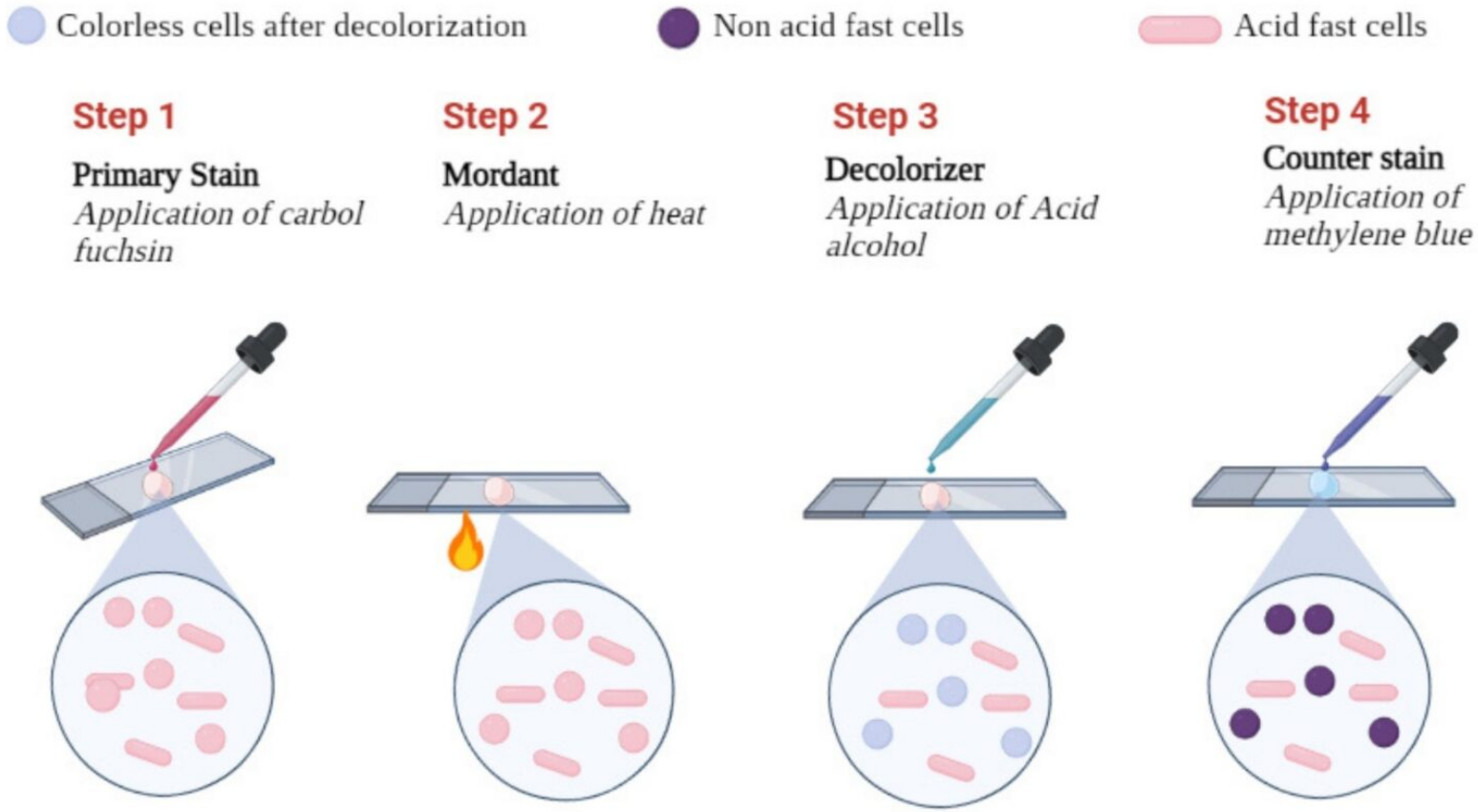
- Non-tuberculous *mycobacteria* (NTM) are environmental present in soil, water, etc. They are rarely transmitted person-to-person.
- Predisposing factors: COPD, cystic fibrosis, immunodeficiency (severe AIDS)
- Control depends on cell-mediated immunity: IFN-γ → macrophage activation. Defects → poor intracellular killing → dissemination
- Chronic, indolent respiratory infection. Cough, sputum, fatigue, weight loss.




Source: DeFlorio-Barker S, Egorov A, Smith GS, Murphy MS, Stout JE, Ghio AJ, Hudgens EE, Messier KP, Maillard JM, Hilborn ED. Environmental risk factors associated with pulmonary isolation of nontuberculous mycobacteria, a population-based study in the southeastern United States. Sci Total Environ. 2021 Apr 1;763:144552.

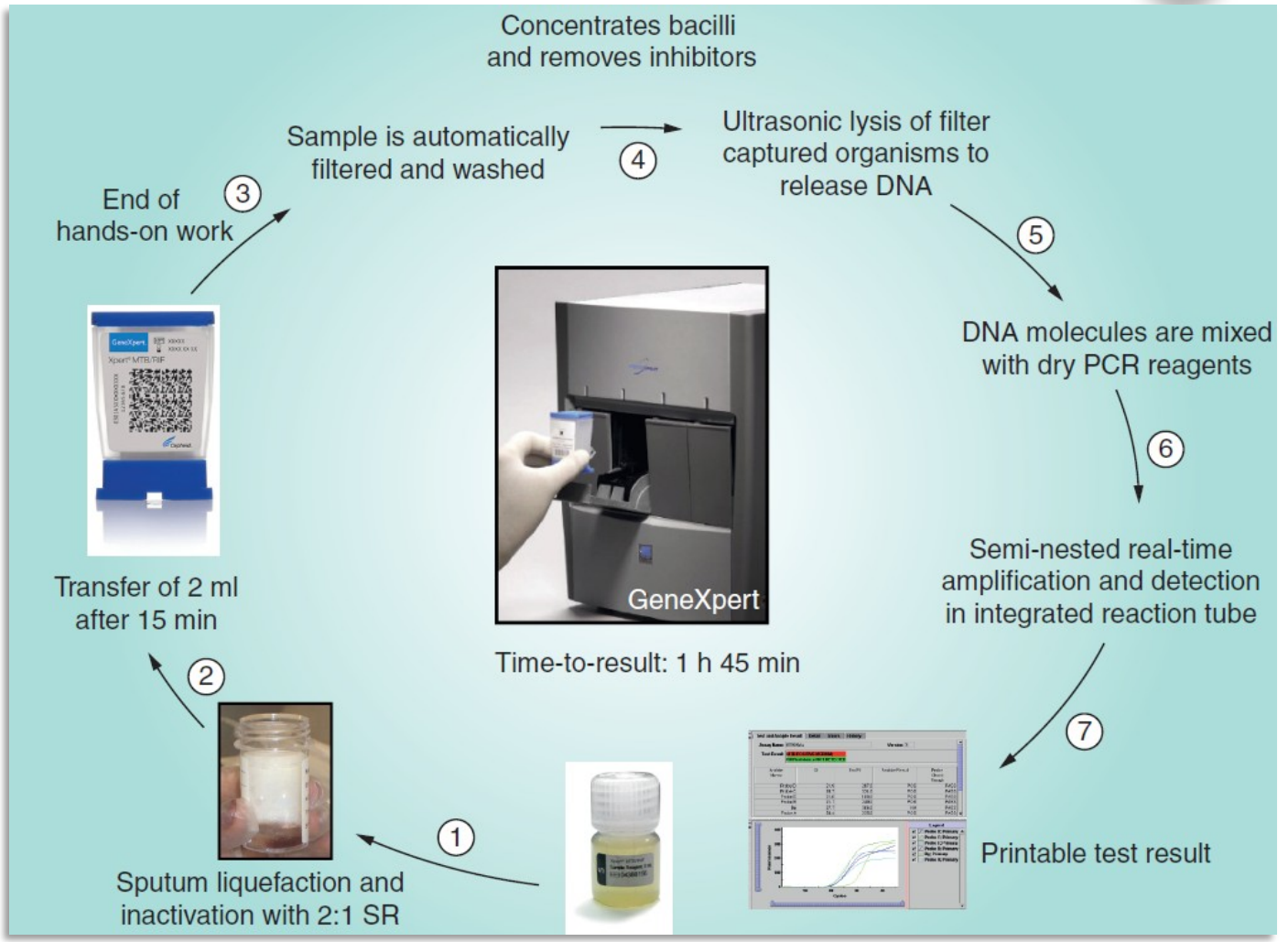
# Diagnosis of TB

- AFB Smear Microscopy:
- **Ziehl-Neelsen** or **Kinyoun** stain: Acid-fast bacilli (pink/red) against blue background
- Advantages: rapid, inexpensive
- Limitations: Cannot distinguish MTB from NTM. Requires ≥10,000 organisms/mL
- Useful for determining infectiousness, not species.



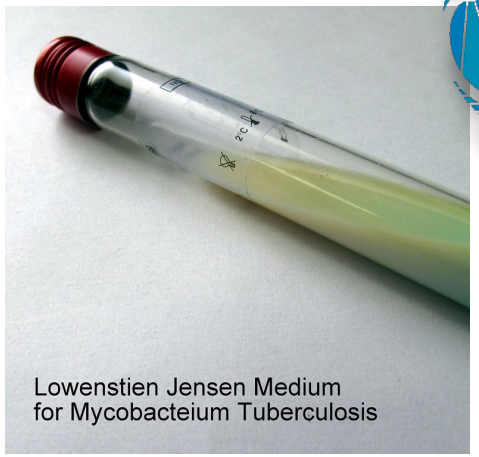
# Diagnosis of TB

- GeneXpert MTB/RIF (PCR-based): Cartridge-based automated PCR. Detects MTB DNA, rifampicin resistance (*rpoB* gene mutation). Turnaround: about 2 hours. High sensitivity in smear-positive disease.
- Does not detect all resistance types (performs best for rifampicin).




Source: Lawn, S. D., & Nicol, M. P. (2011). Xpert® MTB/RIF Assay: Development, Evaluation and Implementation of a New Rapid Molecular Diagnostic for Tuberculosis and Rifampicin Resistance. Future Microbiology, 6(9), 1067–1082.

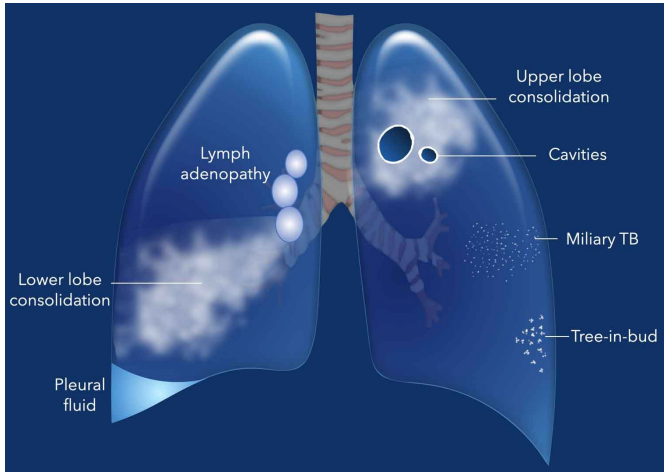
# Diagnosis of TB




- **Culture is the gold standard for TB diagnosis and resistance profiling.**
- Culture methods: Solid media (Lowenstein-Jensen): slow growth in 3-8 weeks.
- Liquid culture (MGIT): faster detection in about 7-14 days.
- Advantages: Confirms viable MTB, allows species identification (MTBC vs. NTM). Enables phenotypic drug susceptibility testing.
- Limitations: Slow turnaround

# Diagnosis of TB

- **IGRA (Interferon-γ Release Assay):** Detects TB infection (latent or active), not affected by BCG vaccination, does NOT distinguish latent from active TB, limited sensitivity in severe immunosuppression
- Tuberculin Skin Test (TST): Detects TB infection, false positives with BCG vaccination and some NTM.
- Imaging (CXR/CT): upper-lobe infiltrates, cavitary lesions, hilar/mediastinal lymphadenopathy with or without consolidation

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**Classification of the Tuberculin Skin Test (PPD) Reaction**

**≥ 5 mm**

- HIV positive
- Recent contact with an active TB patient
- Nodular or fibrotic changes on chest X-ray
- Organ transplant

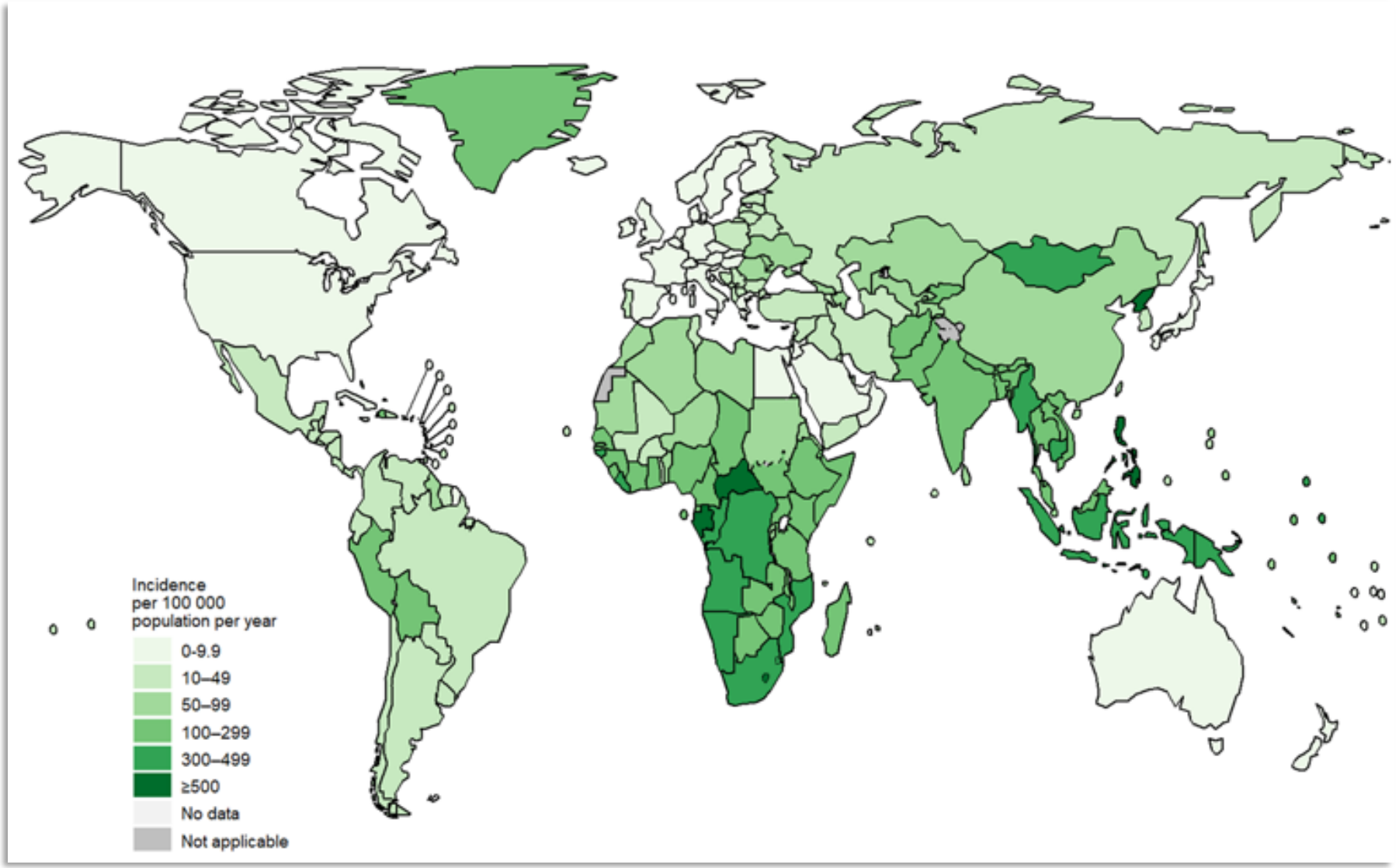
**≥ 10 mm**

- Recent arrivals (< 5 yrs) from high-prevalence countries
- IV drug users
- Resident/employee of high-risk congregate settings
- Mycobacteriology lab personnel
- Comorbid conditions
- Children < 4 yrs old
- Infants, children, & adolescents exposed to high risk categories

**≥ 15 mm**

- Persons with no known risk factors for TB

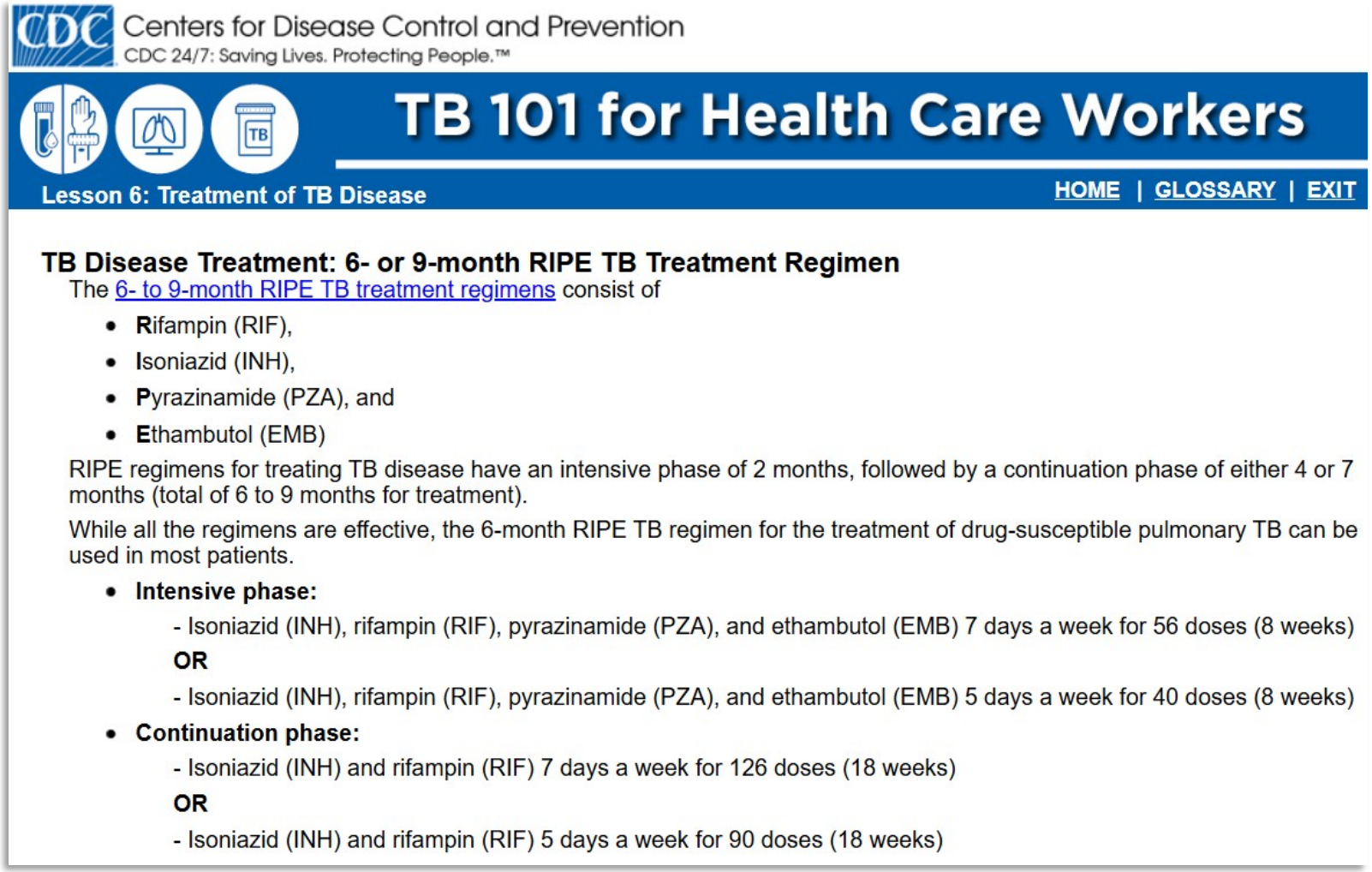
# TB Global Epidemiology




- Based on the WHO statistics, a total of 1.23 million people died from TB in 2024.
- In 2024, an estimated 10.7 million people fell ill with TB worldwide.

# Treatment of TB

- Standard therapy (**RIPE regimen**):
- Intensive phase (first 2 months): Rifampicin + INH + Pyrazinamide + Ethambutol.
- Continuation phase (next 4 months): INH + Rifampicin.
- Total duration: 6 months for drug-susceptible pulmonary TB
- *M. tuberculosis* develops resistance via spontaneous chromosomal mutations. Inadequate treatment selects resistant mutants, especially with monotherapy, poor adherence, incorrect dosing or duration



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TB 101 for Health Care Workers

Lesson 6: Treatment of TB Disease

HOME | GLOSSARY | EXIT

**TB Disease Treatment: 6- or 9-month RIPE TB Treatment Regimen**

The 6- to 9-month RIPE TB treatment regimens consist of

- **R**ifampin (RIF),
- **I**soniazid (INH),
- **P**yrazinamide (PZA), and
- **E**thambutol (EMB)

RIPE regimens for treating TB disease have an intensive phase of 2 months, followed by a continuation phase of either 4 or 7 months (total of 6 to 9 months for treatment).

While all the regimens are effective, the 6-month RIPE TB regimen for the treatment of drug-susceptible pulmonary TB can be used in most patients.

- **Intensive phase:**
  - Isoniazid (INH), rifampin (RIF), pyrazinamide (PZA), and ethambutol (EMB) 7 days a week for 56 doses (8 weeks)

  **OR**

  - Isoniazid (INH), rifampin (RIF), pyrazinamide (PZA), and ethambutol (EMB) 5 days a week for 40 doses (8 weeks)
- **Continuation phase:**
  - Isoniazid (INH) and rifampin (RIF) 7 days a week for 126 doses (18 weeks)

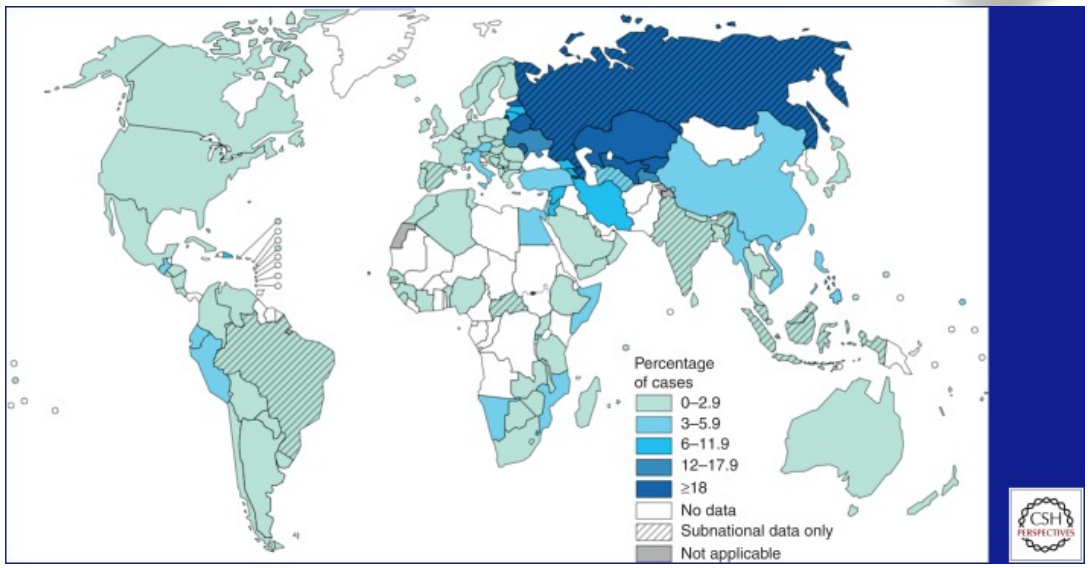
  **OR**

  - Isoniazid (INH) and rifampin (RIF) 5 days a week for 90 doses (18 weeks)

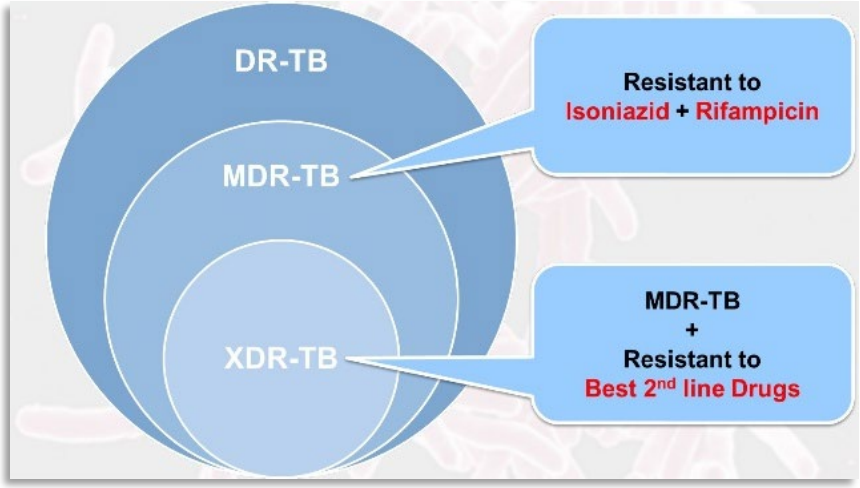
# Treatment of TB

- **Major Resistance Patterns**
- INH resistance: most common first-step resistance. Rifampicin resistance: Surrogate marker for multidrug-resistant TB.
- Multidrug-resistant TB (**MDR-TB**): Resistance to both isoniazid and rifampicin.
- Extensively drug-resistant TB (**XDR-TB**): resistant to rifampicin (and may also be resistant to INH), and that is also resistant to at least one fluoroquinolone (levofloxacin or moxifloxacin) and to at least one other Group A drug (bedaquiline or linezolid).

- Group A = levofloxacin or moxifloxacin, bedaquiline, and linezolid; Group B = clofazimine, and cycloserine or terizidone; and Group C = ethambutol, delamanid, pyrazinamide, imipenem–cilastatin or meropenem, amikacin (or streptomycin), ethionamide or prothionamide, and *p*-aminosalicylic acid.

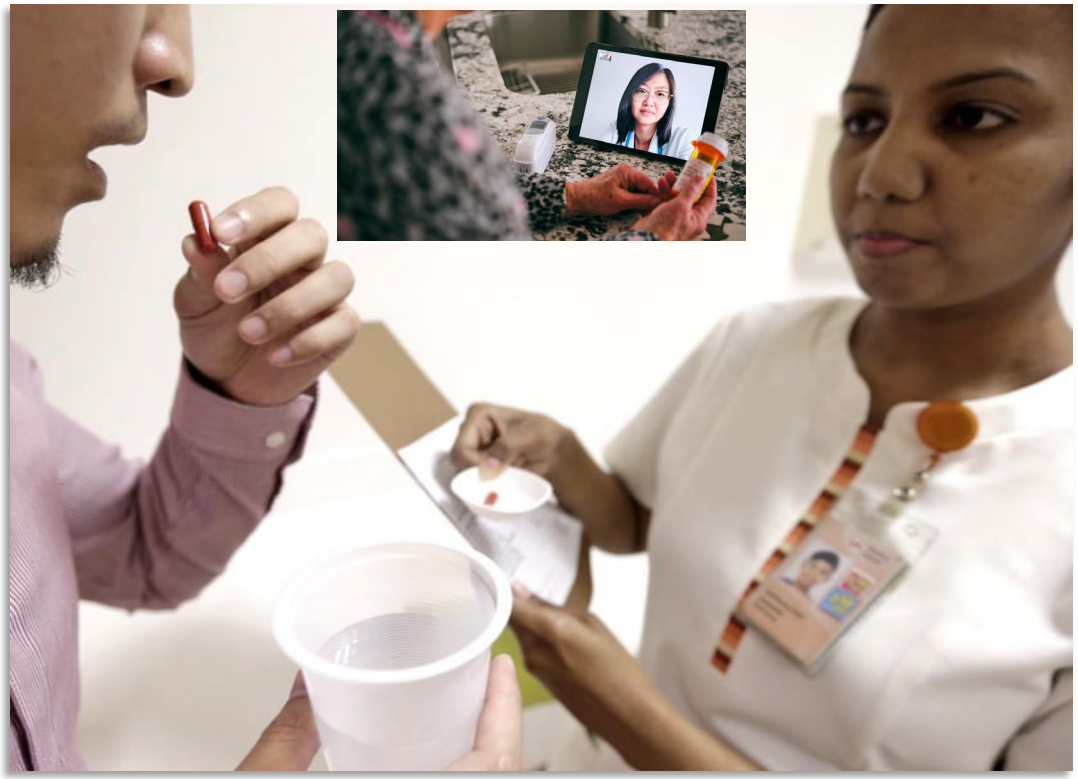



Source: Seung, K. J., Keshavjee, S., & Rich, M. L. (2015). Multidrug-Resistant Tuberculosis and Extensively Drug-Resistant Tuberculosis. Cold Spring Harbor perspectives in medicine, 5(9), a017863.



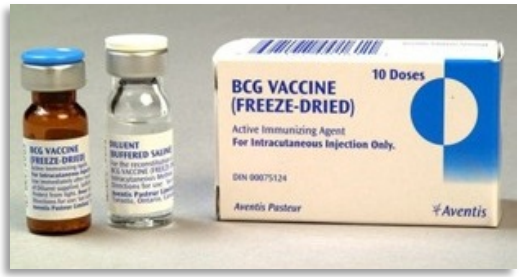

Source: 2025 Epomedicine . All rights reserved; https://epomedicine.com/medical-students/mdr-xdr-tb/

# Treatment of TB

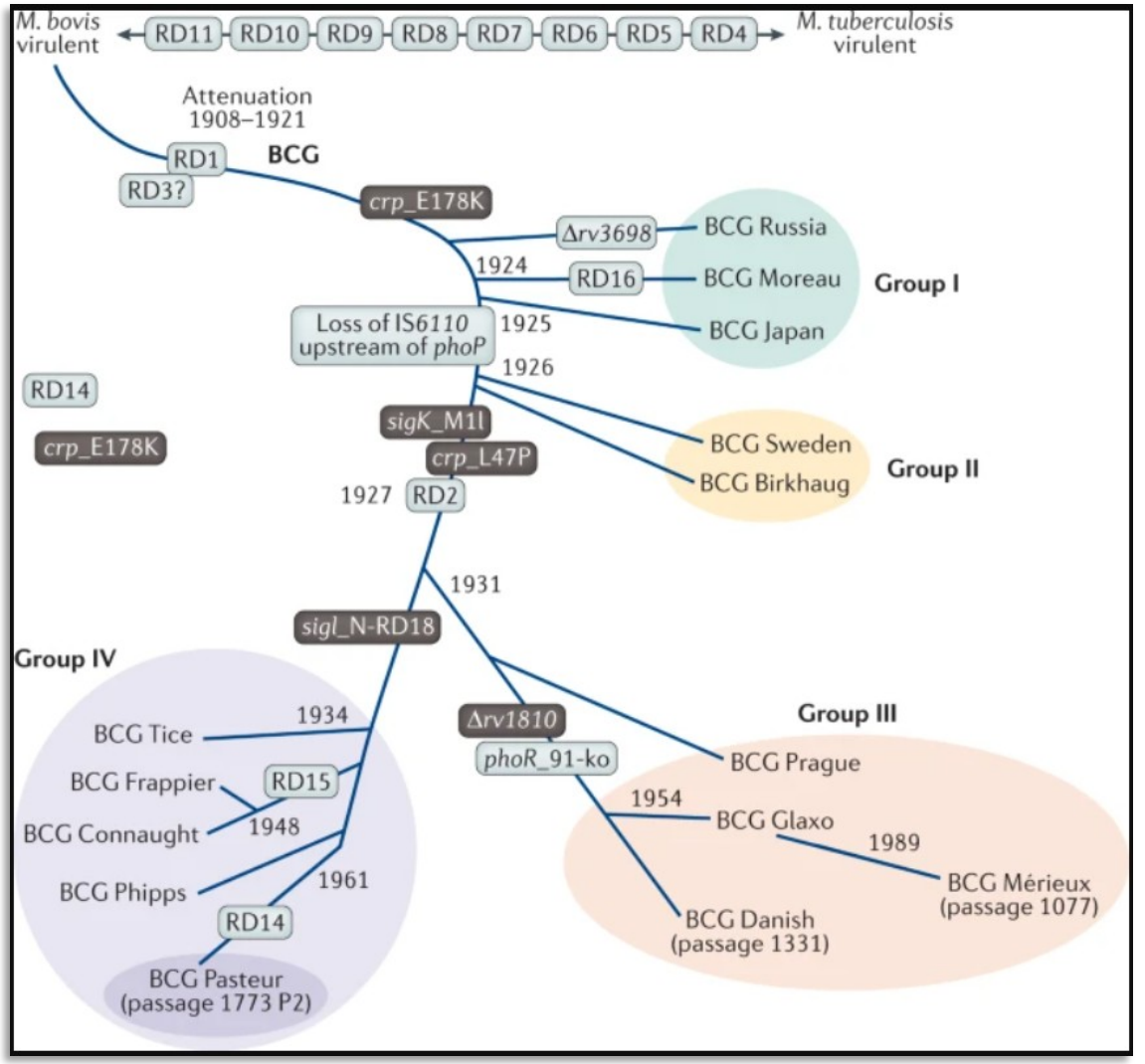


- **Adherence & Resistance Prevention**
- Poor adherence is a primary risk factor for TB resistance
- **Directly Observed Therapy (DOT):** Ensures full course completion. Reduces emergence of MDR-TB. The core strategy recommended by World Health Organization
- **Clinical Consequences of Drug Resistance:**
- Longer treatment duration (≥18–24 months, use of less effective, more toxic second-line drugs, higher mortality and transmission risk

# TB Prevention




- **BCG (Bacillus Calmette-Guérin) vaccination:**
- Live attenuated strain of *Mycobacterium bovis*. Administered intradermally, usually at birth in TB-endemic countries. BCG induces Th1 response. Highly effective in infants and young children against TB meningitis and miliary TB.
- **It reduces severity rather than acquisition of infection.**
- Variable and limited efficacy against adult pulmonary TB. Protection wanes over time and it does not prevent latent infection. It causes false-positive TST

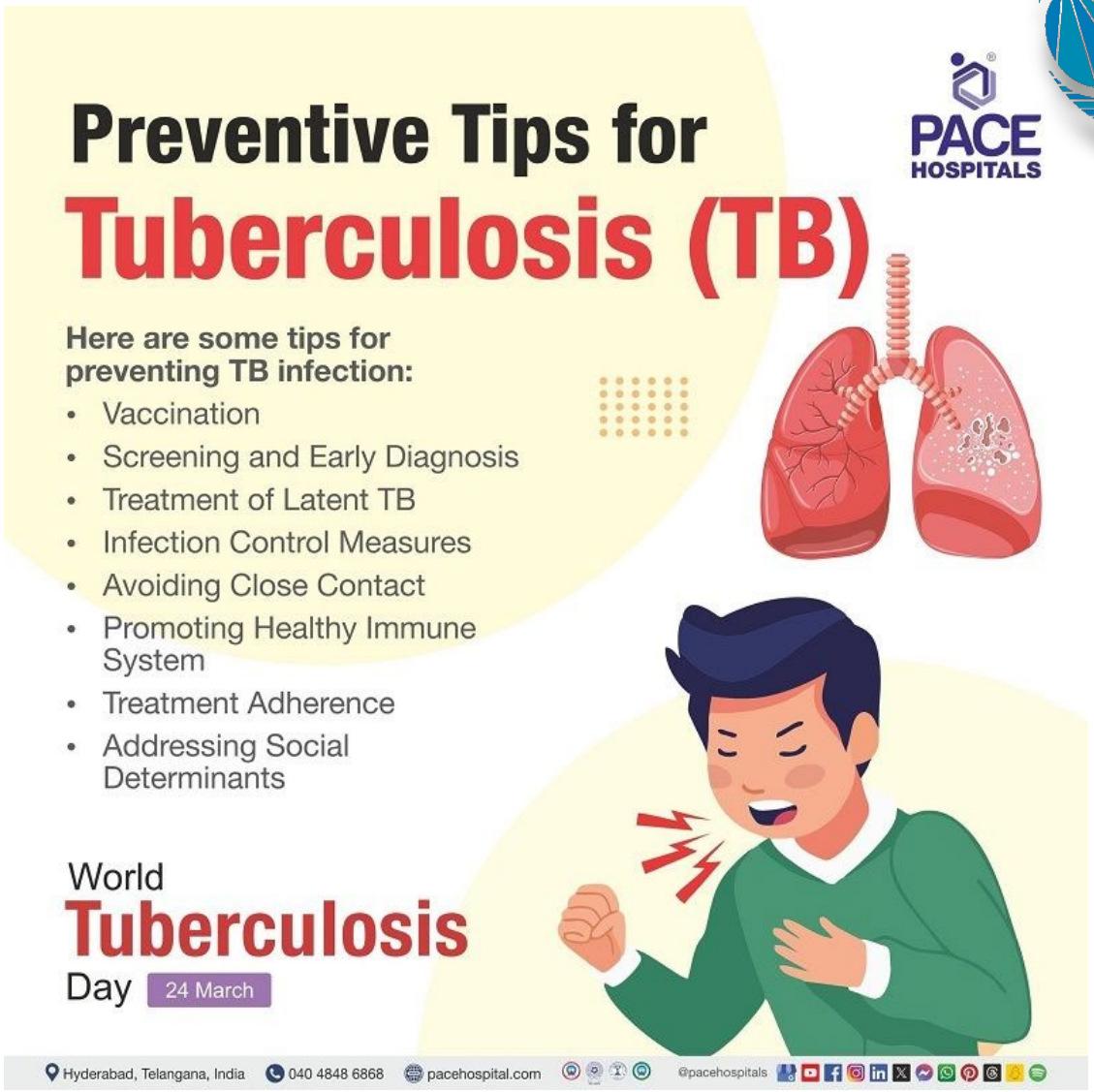



Source: Lobo, N., Brooks, N.A., Zlotta, A.R. et al. 100 years of Bacillus Calmette–Guérin immunotherapy: from cattle to COVID-19. Nat Rev Urol 18, 611–622 (2021).

# TB Prevention

- **TB is preventable and curable.**
- **Early case detection and prompt treatment**
- **Active TB case finding**
- **Treatment of latent TB infection**

- Addressing social determinants of health (overcrowding, poor housing, malnutrition)
- Infection control measures (BCG, adequate ventilation, respiratory masks in high-risk settings, isolation of infectious cases when needed)



# Thank You!
# Wishing you all the best!