The Gene and Regulatory Network Involved in Ethambutol Resistance in Mycobacterium tuberculosis

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Background: Tuberculosis (TB) remains one of the top ten causes of death worldwide, aggravated by the emergence of drug-resistant Mycobacterium tuberculosis. Ethambutol (EMB), a first-line anti-TB drug, targets the mycobacterial cell wall by inhibiting arabinan biosynthesis (1). However, increasing resistance to EMB threatens TB control strategies (2). Understanding the genetic and regulatory mechanisms underlying EMB resistance is crucial for improving diagnosis and developing novel therapeutic targets.

Objectives: to provide a comprehensive analysis of the genes implicated in Ethambutol resistance within Mycobacterium tuberculosis, while also detailing the underlying regulatory networks and the pharmacoproteomic effects of the drug.
Methods: an extensive review and synthesis of existing literature to compile data on genetic mutations, specifically focusing on the embCAB operon and its associated protein alterations using bioinformatic techniques, such as protein sequence alignment, to identify highly conserved residues across various mycobacterial species and employed chord plots to visualize the complex interactions between Ethambutol-related genes and essential cellular pathways

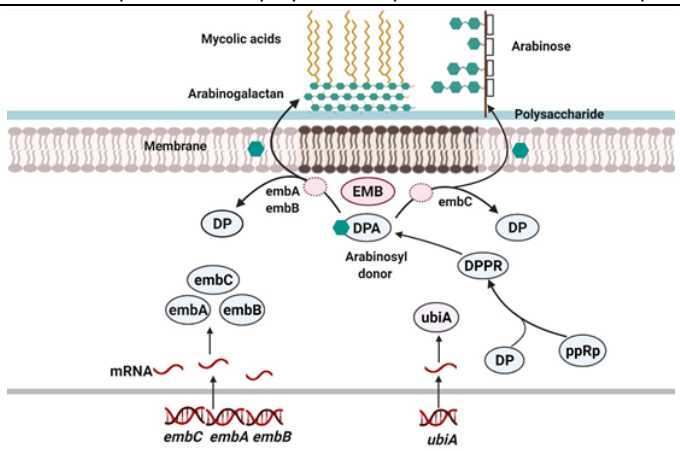

Figure A. Illustration of the pathway regulated by emb genes in the cell wall of M. tuberculosis.

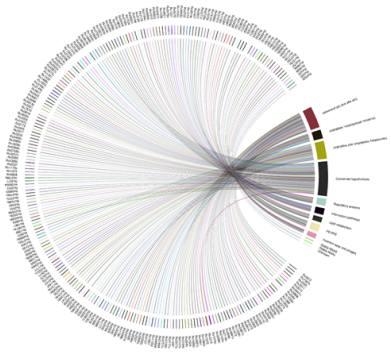

Figure B. Data visualization of EMB-specific genes and cellular pathway.

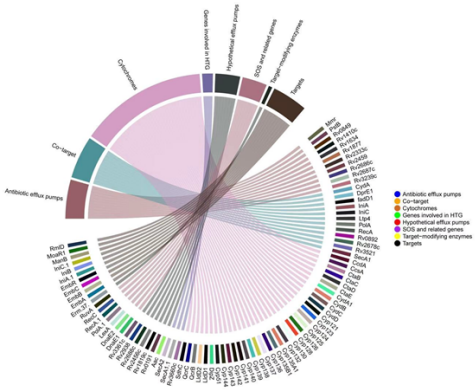

Figure C. Data visualization of the Protein-protein interaction networks and genes involved in different resistance mechanisms

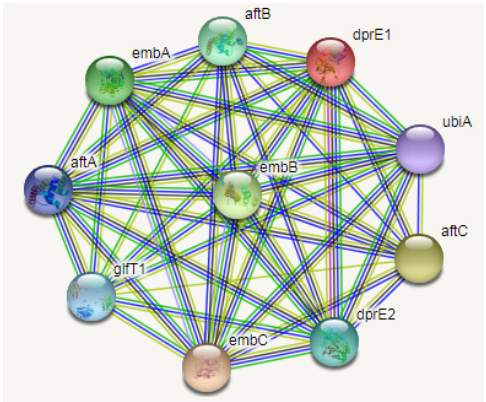

Figure D. EMB interaction network in the cell wall biosynthesis of M. tuberculosis.

Discussion: Ethambutol targets the arabinan component of the *Mycobacterium tuberculosis* cell wall by inhibiting the *EmbB* protein at codon 306. The *Met306Leu* or *Met306Val* mutations occur in 69% of resistant strains and increase the MIC to 40 µg/ml, critically altering drug-protein interaction. This dominant mutation in the embCAB operon represents the primary resistance biomarker detectable by targeted PCR.
Ethambutol resistance results from a complex multifactorial network involving five main pathways. Membrane genes (*embCAB)* cooperate with efflux pumps (Rv1258c, tap, drrA) to actively expel the drug. Lipid metabolism (fadD, acpM) and ribosomal proteins (rpsL, rpoB) contribute to resistant strain fitness. Finally, detoxification systems (katG, ahpC) protect against EMB-induced oxidative stress. An alarming cross-resistance exists between EMB and rifampicin. rpoB/C mutations co-select EMB resistance in 30% of MDR-TB cases, exacerbating the global multidrug-resistant tuberculosis crisis.
Pharmacoproteomic analysis reveals a marked differential response. Sensitive strains suffer catastrophic arabinan depletion causing cell lysis. Conversely, resistant strains activate the PhoP-PhoR network and upregulate efflux pumps, ensuring intracellular survival despite EMB exposure.
These findings carry major therapeutic implications. Priority surveillance of *embB306* mutations enables early detection in MDR-TB programs. Efflux inhibitors like verapamil could restore EMB sensitivity. Finally, developing specific arabinosyltransferase analogs offers a promising perspective.

Conclusion: Ethambutol resistance in M. tuberculosis is driven by a complex gene and regulatory network involving cell wall biosynthesis, efflux systems, metabolism, and stress responses. Integrating genomic, proteomic, and interaction network data provides valuable insight for improved diagnostics and the rational design of next-generation anti-TB therapies.

References : 1. Organisation, W.H. 2020. Global tuberculosis report 2018.
2. https://pubmed.ncbi.nlm.nih.gov/35939307/