

8<sup>th</sup> Annual Congress on

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# & VIROLOGY, INFECTIONS AND OUTBREAKS

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## Characterization of Rv2037c, a phospholipase of *Mycobacterium tuberculosis*

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*Mycobacterium tuberculosis* is the most successful pathogen humans encountered ever. Genome sequence analysis of *Mtb* genome has revealed that out of 4000 genes, 40–50% genes have been identified as hypothetical. Few hypothetical mycobacterium proteins were reported to actively participate in the complex physiology of *M. tuberculosis*. Rv2037c has been annotated as hypothetical protein conserved in pathogenic strains of mycobacteria and involved in cell wall and cell processes. In this study, we have cloned rv2037c in pET28a vector, expressed in *E.coli* BL21(DE3) strain. The recombinant protein was purified by Ni-NTA chromatography. Lipase activity was determined spectrophotometrically using pNP-esters. The enzyme was characterized by biochemical and biophysical methods. Also, the active site mutants were created using site-directed mutagenesis technique. The expression pattern rv2037c in *M. tuberculosis* H37Ra was studied under various stresses *in vitro* culture conditions. Based on biochemical characterization, Rv2037c possess lipase activity toward mid-carbon chain length having pNP-decanoate as its optimal substrate. Its optimum temperature and pH were 40°C and 8, respectively, stable up to 50°C and pH 7-10. Biophysical characterization confirms that it belongs to the  $\alpha/\beta$  hydrolase family. It was also confirmed to be phospholipase category of lipases by activity analysis of mutants and by phospholipase assay. The expression of Rv2037c was upregulated 26 fold in iron, 3 fold in nutritive and 2 fold acidic stress conditions. Our study demonstrated that Rv2037c, a phospholipase, was significantly upregulated under stress condition and might be helpful in the survival of the bacterium.

### Biography

Bandana Kumari is PhD scholar in the Department of Biotechnology, Chandigarh, India. She has been working on mycobacterial lipases since last two years under the supervision of Prof Jagdeep Kaur. Her expertise is in Molecular biology, Protein chemistry and animal tissue culture.

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