Functional analysis of *Rv1900c* gene product from *Mycobacterium tuberculosis*

Bandana Kumari and **Jagdeep Kaur**
Panjab University, India

Tuberculosis is still a global threatened disease caused by intracellular pathogen *Mycobacterium tuberculosis (Mtb)*. India accounts for 60% new cases worldwide. The bacterium can outperform the host defense mechanism and can persist inside the body for decades in dormant stage. There are many factors that are known to play role during this stage of bacterium; storage of triacylglycerol (TAG) is one of them. The literature on *Mtb* is replete with strong evidence that TAGs play a critical role in the intracellular/ intraphagosomal survival of this pathogen in the host. *Mtb* genome contains about 4000 genes and 26 (LipA- lipZ) of them are annotated as putative lipases/esterases. *Rv1900c* of *Mtb* Lip family annotated as LipJ; conserved in all *Mycobacterium* species. It contains two domains, N- terminal α/β hydrolase domain and C- terminal cyclase homology domain. In this study we have cloned LipJ full length and N- Terminal lipolytic domain in pET28a vector, expressed in *E. coli* BL21 (DE3) host cells. Recombinant protein was purified using Ni-NTA affinity chromatography. Biochemical characterization of holoenzyme and its N-terminal revealed esterase activity towards pNP- caprate as their optimal substrate. Both the holoenzyme and N-terminal has 40°C as optimal temperature and is stable up to 60 °C, and their pH optima was found to be pH9 and stable from pH6 to 9. Hence, it is confirmed that the esterase activity of holoenzyme was just because of its N- terminal domain. Biophysical characterization confirms that it belongs to α/β hydrolase family.

Speaker Biography

Bandana Kumari is a PhD Scholar in 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.

e: vandanadhiman21@gmail.com

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