6. SUMMARY

The main theme of the thesis was to study the host-pathogen interaction mechanism between the macrophage nuclear receptors and the pathogen, *M. tuberculosis*. We found few candidate NRs which were involved in the crosstalk with the bacilli and also revisited the function of vitamin D, which has anti-TB properties.

M. tuberculosis–macrophage interactions are key to pathogenesis and clearance of these bacteria. Although interactions between *M. tuberculosis*-associated lipids and TLRs, non-TLRs, and opsonic receptors have been investigated, interactions of these lipids and infected macrophage lipid repertoire with LSNRs expressed in macrophages have not been addressed. In the first objective (Chapter 3) (Mahajan et al., 2012), using loss of function strategy we reported that *M. tuberculosis*–macrophage lipids can interact with host PPARγ and TR4 to ensure survival of the pathogen by modulating macrophage function. These two LSNRs were modulated by *M. tuberculosis* lipids to create a foamy niche by alternative polarization of the macrophages and induction of IL-10, which leads to survival of *M. tuberculosis*. These results suggest *M. tuberculosis* lipids as possible heterologous ligands for PPARγ and TR4 and are suggestive of adaptive or co-evolution of the host and pathogen. These observations expose a novel paradigm in the pathogenesis of *M. tuberculosis* amenable for pharmacological modulation.

The cell wall of *M. tuberculosis* is configured of bioactive lipid classes that are essential for virulence and potentially involved in the formation of FMs and granulomas. In the second objective (chapter 4) (Dkhar et al., 2014), we have characterized, identified, and adopted a heterologous ligand "keto-MA" from amongst *M. tuberculosis* lipid repertoire for the host orphan NR TR4. Crosstalk between cell wall lipids and TR4 was analyzed by luciferase promoter reporter assays.MA was found to significantly transactivate TR4 compared to other cell wall lipids. Among the MA, the oxygenated form, keto-MA, was responsible for transactivation, and the identity was validated by TR4 binding assays followed by TLC and NMR. This keto-MA-TR4 axis seems to be essential to this oxygenated MA induction of FMs and granuloma formation as evaluated by *in vivo* model of granuloma formation. TR4 binding with keto-MA features a unique association of host NR with a bacterial lipid and adds to the presently known ligand repertoire beyond dietary lipids.

Summary

Pharmacological modulation of this heterologous axis may hold promise as an adjunct therapy to frontline TB drugs. Vitamin D or $1,25(OH)_2D_3$ has been a widely used supplement against TB well before the discovery of the drug regimen. Barring the known classical functions of $1,25(OH)_2D_3$- VDR axis, it has been implicated in impairing the growth of *M. tuberculosis* and rescuing the host defenses blocked by the bacilli. As an anti-*M. tuberculosis* host factor, VDR has been reported to co-ordinate with PRRs like the TLRs triggering the CAMP pathway leading to autophagy. In the third objective (Chapter 5), we reported anti-*M. tuberculosis* role of VDR independent of CAMP pathway via blocking of *M. tuberculosis* induced FMs and by preventing of trafficking of lipids in bacilli laden phagosomes by gene repression of CD63. FMs are nutrient laden depots essential for the survival of *M. tuberculosis* within the macrophages. $1,25(OH)_2D_3$-VDR inhibition of FMs is causatum to suppression of oxidized LDL uptake by gene repression of CD36. Our bioinformatics prediction software revealed a negative VDRE confirmed by the presence of a negative half site (non- consensus) in proximal gene promoter of gene CD63 as confirmed by DNA binding experiment. This reported negative regulation of gene CD63 seems to prevent the trafficking of lipid nutrient and perhaps LBs–phagosome interaction.