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Isocitrate Lyase Homologue of *Mycobacterium fortuitum* Plays Role in *in-vitro* Survival and Stress Response

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Abstract—*M. fortuitum is a versatile pathogen capable of infecting with variable clinical manifestations to normal as well as immunocompromised individuals with varying degree; accounting from asymptomatic to severe presentation. Isocitrate lyase (icl) is an important gene which plays an imperative role in Mycobacterium tuberculosis persistent infection. icl is the initial enzyme of the glyoxylate shunt, a secondary metabolic pathway which allows the bacilli to utilize fatty acids as carbon and energy sources during limited availability of primary carbon sources. Literature regarding Mycobacterium tuberculosis persistence was searched on Google scholar, Go-Pubmed and Science direct for genes involved in persistence. To study role of icl in regulation of persistence, stress response and pathogenesis of M. fortuitum, multiple sets of primers were designed from flanking region of icl conserved domain sequence (CDS) and were PCR amplified on M. fortuitum genome. The different PCR products obtained were sequenced and checked for homology with the icl of M. tuberculosis. The longest sequence of M. fortuitum ATCC 6841, having maximum homology with M. tuberculosis was submitted to NCBI (GenBANK ID - KM275229). Further to confirm its role the gene was cloned in pMV261 vector in sense and antisense orientations. Presence of full length gene and its orientation was confirmed by restriction digestion and PCR using hsp60 primer (promoter). Obtained sense and antisense mutants were electroporated in M. fortuitum. Sense and antisense mutants of M. fortuitum icl were then studied for their role under different stress conditions including granuloma specific stress conditions with wild type M. fortuitum as control. icl played an important role in survival and proliferation under different stress conditions, thus can be a potential drug target for M. fortuitum infections.*