**INDIAN INSTITUTE OF TECHNOLOGY GUWAHATI**
**SHORT ABSTRACT OF THESIS**


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Thesis Title: **Studies on identification and *in vivo* function of novel drug target enzymes of *Leishmania donovani* using biomolecular approaches**


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## SHORT ABSTRACT


Leishmaniasis is a neglected parasitic disease caused by several species of *Leishmania* parasite. The current drug scenario against this disease is not very good, due to poor efficacy, high cost and major side effects. Moreover, there are several reports about resistance against available drugs. There is an eminent requirement for drugs with higher efficacy and lower side effects. Discovery and validation of novel drug targets are prerequisite for any drug discovery effort. In our study, we have chosen two proteins of the pathogen *Leishmania*, for evaluation as potential drug targets, *viz.* a conserved hypothetical protein and a functionally annotated protein, CAAX prenyl protease II. The conserved hypothetical protein, LdBPK_070020, was conserved throughout the other species of *Leishmania* hinting towards important role of the protein. The other possible target, functionally annotated protein CAAX prenyl protease II, has key role in Ras proteins maturation and localization. The sequence similarity of both the proteins with human genome was significantly low.

The expression of LdBPK_070020 was completely removed from the parasite by employing gene knockout strategy based upon homologous recombination. The data suggests that LdBPK_070020 knock out results in impaired mitochondrial function. In order to identify whether the role of LdBPK_070020 is directly related to mitochondrial function or the protein is indirectly affecting it, we went forward with localization studies. The results illustrated that the protein was present in the nucleus and kinetoplast of the parasite. This suggested the indirect role of LdBPK_070020, possibly by regulating expression of other proteins necessary for mitochondrial function. Complementation of the gene (LdBPK_070020) by episomal expression resulted in partial recovery of the effects. Likewise, complete removal of CAAX prenyl protease II expression did not kill the parasite but the cell growth rate and macrophage infectivity was significantly lower compared to the wild type. Improper localization of Ras proteins (signal proteins) in CAAX prenyl protease II knock out parasite increased G1 to S phase transition in cell cycle. The data support that the removal of CAAX prenyl protease II was affecting the parasite growth and infectively significantly. In short both the proteins are potential drug targets, with LdBPK_070020 being a significantly better target than CAAX preny protease II.