

Cloning, Expression and Characterization of *Leptospira interrogans* serovar Copenhageni Gene LIC13435

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Leptospirosis is a re-emergent zoonosis characterized by an acute febrile and systemic illness in humans caused by pathogenic spirochetes belonging to the genus *Leptospira*. In Brazil, leptospirosis is an important economic and public health problem. The complete genomic sequence of *Leptospira interrogans* offered a new strategy for the identification of new proteins that could be vaccine candidates, since environmental control measures are difficult to implement and there is not an available vaccine for human use. Secreted and surface exposed molecules are potential targets for inducing immune responses in the host. Thus, we selected the gene LIC13435, predicted to code for a putative outer membrane protein, to be characterized biochemically. The sequence was selected from the genome of *Leptospira interrogans* serovar Copenhageni using bioinformatics tools. The sequence was cloned by PCR and the expression of the recombinant protein was tested in *Escherichia coli* strains. Purification of the recombinant proteins was done by metal affinity chromatography due to the presence of a 6Xhis tag introduced at the N-terminus of LIC13435. Circular dichroism was performed to characterize the secondary structure, being mainly composed of a helix. The antisera were produced by intraperitoneal immunization of BALB/C mice. ELISA and western blot were done to confirm the titers and specificity of the antiserum. Preliminary western blot test indicates that this protein is expressed by virulent low-passage forms of pathogenic *Leptospira* serovar Copenhageni, while it is decreased during stationary phase. Further characterizations are underway.

Key words: *Leptospira*, protein characterization, vaccine

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