Cloning, Expression and Purification of Predicted Lipoproteins of *Leptospira interrogans*

Oliveira, R.[1,2], Oliveira, T.R.[1], Atzingen, M.V.[1,2] and Nascimento, A.L.T.O.[1,2]

[1] Centro de Biotecnologia, Instituto Butantan, São Paulo, Brazil;
[2] Interunidades em Biotecnologia, Instituto de Ciências Biomédicas, Universidade de São Paulo, São Paulo, Brazil.

Introduction: Leptospirosis is worldwide zoonotic disease caused by pathogenic spirochaetes of the genus *Leptospira*. In the urban settings, rodents are the most important carriers of the disease because they continuously shed live leptospires in their urine. Humans can be infected through contact with, soil or water contaminated with urine containing leptospires. Since the control of the rodents and sanitation measures are not easily implemented, the development of reliable vaccine is necessary to combat the leptospirosis. **Objectives:** The aim of this project is to study three genes that encode for predicted lipoproteins selected from the genome sequences of *Leptospira interrogans* serovar Copenhageni and to evaluate their immune reactivity in animal model and in sera from patients diagnosed with leptospirosis. **Results and Conclusion:** The gene sequences of LIC10258, LIC12880 and LIC12238 were amplified by PCR methodology from genomic DNA of *L. interrogans* serovar Copenhageni and the DNA inserts cloned into the *E. coli* expression vector pAE. The pAE constructions were employed to transform BL21 SI *E. coli* strain. Protein expression was achieved by the addition of NaCl to the medium. The expected protein bands of ~65.7, 30.6 and 17.6 kDa, corresponding to rLIC10258, rLIC12880 and rLIC12238, respectively, were visualized by Coomassie blue stained. The rLIC12238 was expressed in soluble form while rLIC10258 and rLIC12880 were expressed in their insoluble form. The purification of the proteins through metal-chelating chromatography rendered, in each case, a major protein band, suitable for biological assays.

Key words: lipoprotein, characterization, *Leptospira interrogans*
Supported by FAPESP, CNPq and Fundação Butantan