

Title: IDENTIFICATION OF OUTER MEMBRANE PROTEINS OF *LEPTOSPIRA* THAT INTERACT WITH HOST'S EXTRACELLULAR MATRIX PROTEINS

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Resumo:

Leptospirosis is caused by pathogenic species of the spirochete *Leptospira*. It is one of the most widespread zoonosis worldwide and represents a major public health problem in tropical and developing countries. During the infection process, pathogenic leptospire are able to survive, multiply and trigger a specific immune response. This is attributed to their ability to adhere to eukaryotic cells and extracellular matrix proteins and to escape the mechanisms of innate host's defenses. In this project, we aim to identify surface proteins of *Leptospira* that interact with host's molecules. *Leptospira* outer membrane proteins, obtained by sarkosyl extraction, were incubated with fibronectin, fibrinogen and laminin pre-immobilized on magnetic beads. Bound ligands were analyzed by LTQ Orbitrap Velos Mass Spectrometer (Thermo Scientific) and identified using the Mascot engine version 2.4.0 (Matrix Science, London, UK) with uniprot *Leptospira interrogans* database. Selection of proteins of interest was performed using the Scaffold software (version_4.4.4, Proteome Software Inc., Portland, OR), Mascot version 2.4.1 as search engine, and the gene_association.goa_uniprot and NCBI as GO annotation sources. The following parameters were adopted: 95% protein threshold, 80% peptide threshold and minimum number of peptides 1. Interestingly, some of the proteins identified have been already described as potential leptospiral adhesins, such as LipL32, OmpL1, OmpA family protein and Elongation Factor Tu. In addition, a number of unknown proteins was also identified, and might be considered as promising candidates for further analysis. The selected proteins will be produced in *Escherichia coli* and a number of functional assays will be performed to better characterize the interactions observed. It is believed that this approach will bring a wider knowledge about the ligands of *Leptospira* involved in the interactions with host molecules. Given the need to develop preventive strategies that can interfere with the invasion of this bacterium, the identification and characterization of proteins that may be involved in these processes is of fundamental importance.

Key words: *Leptospira*, outer membrane proteins, interaction, fibronectin, fibrinogen, laminin.

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