SPECIFIC– AND *IN VIVO* EXPRESSED *LEPTOSPIRA* ANTIGENS FOR A BROAD SPECTRUM LEPTOSPIROSIS VACCINE DEVELOPMENT

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ABSTRACT

Leptospirosis, a zoonosis caused by bacteria of the genus *Leptospira*, is an important emerging infectious disease worldwide. Vaccines to prevent against the leptospirosis prepared from inactivated whole bacterial cells or outer membrane components of pathogenic *Leptospira* spp. elicit immunity which is limited to the homologous infection. Also the vaccines failed to induce long-lasting immunity. Thus, there is a need of more effective vaccine that not only elicits immunity across the heterologous *Leptospira* spp. serovars but also induces long-lasting immunological memory, for both human and veterinary uses. Recently, the genomes of three *Leptospira* spp. serovars, *i.e.*, Copenhageni, Lai, and Hardjo have been completely sequenced and the whole-genome (*in silico*) analysis has been used for identifying the broad spectrum *Leptospira* vaccine candidate genes. However, while such approach, *i.e.*, reverse vaccinology, may be relatively convenient, it is required that several genes should be studied concurrently as many of the selected candidates or their expressed proteins counterparts may turned out to be either poorly immunogenic or confer only limited immunity.

In this study, the difference of proteins expressed by non-pathogenic and pathogenic *Leptospira* spp., the epitopes, antigenic components, immunome and *in vivo* expressed protein antigens of pathogenic *Leptospira* spp. were revealed using 2D-DIGE, 2DE-immunoblotting, proteomics, protein identification by mass spectrometry and bioinformatics. It was found that there are several common

immunogenic proteins which could be used for diagnostic targets and vaccine candidates.

In this study, a plasmid DNA vaccine was constructed from cloning gene encoding a transmembrane porin protein, OmpL1, of pathogenic *Leptospira interrogans*, serogroup Icterohaemorrhagiae, serovar Copenhageni into a mammalian expression vector pcDNA3.1(+). The protective efficacy of the *ompL1*-pcDNA3.1(+) plasmid DNA vaccine was studied by immunizing hamsters intramuscularly with three doses of the vaccine at two week intervals. The empty pcDNA3.1(+) and PBS were used as mock as negative vaccine controls, respectively. All animals were challenged with the heterologous *Leptospira interrogans,* serogroup Pomona, serovar Pomona (10 LD50), at one week after the last vaccine booster. The *ompL1*-pcDNA3.1(+) plasmid DNA vaccine rescued some vaccinated animals from the lethal challenge and delayed death time, reduce morbidity. While the results are encouraging, further studies are needed to optimize the immunization schedule, vaccine dosage and formulation in order to maximize the efficacy of the vaccine.

KEYWORDS: LEPTOSPIROSIS / LEPTOSPIROSIS VACCINE / VIRULENCE FACTORS / PROTEOMICS / IMMUNOMICS

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