

Molecular Analysis of the OmpL1 Gene of *L.interrogans* Vaccinal Serovars in Iran

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Background & Objectives: Leptospirosis, caused by infection with pathogenic *Leptospira* species is the most prevalent zoonotic disease in the world. The leptospiral vaccines used currently are mainly multivalent dead whole-cell mixtures made of several local dominant serovars. These vaccines, however, do not confer cross-protective immunity and may lead to incomplete, short-term immunity as well as serious side effects. Thereupon design and construction of an efficient recombinant vaccine for leptospirosis control is very important. OmpL1 is an immunogenic porin protein i.e. expressed only in pathogenic *Leptospira* spp. Highly conserved OmpL1 antigen is of special significance in vaccination and serodiagnosis for leptospirosis. In order to homological (polymorphism) analysis we sequenced and compared ompL1 genes cloned from standard pathogenic serovars of leptospires prevalent in Iran.

Methods: Three pathogenic vaccinal serovars and one saprophytic species (*L.biflexa*) were used to inoculate into the selective culture medium and extraction of the genomic DNA by standard Phenol-Chloroform methods. The specific primers for proliferation of ompL1 gene were designed. The PCR products of pathogenic serovars were ligated in pJET1.2/blunt vector and transformed in competent *E.coli* Top10 cells. The extracted recombinant plasmids were sequenced.

Results: PCR amplification of the ompL1 gene using the designed primers resulted in a 963 bp ompL1 gene product in all three pathogenic vaccinal serovars tested. No PCR products were amplified from the non-pathogenic *L.biflexa*. Our results showed that sequence identity of the ompL1 gene between *L.Canicola* (RTCC2805) and *L.Sejroe hardjo* (RTCC2821) was 100% while *L. grippityphosa* (RTCC2808) had less identity (88.5 %) with other two serovars named.

Conclusion: According to the results of this study and other researches, OmpL1 gene nucleotide sequence is slightly different within some of leptospires. However, the predicted secondary structure of the OmpL1 proteins revealed that there is little difference among them. Thus, the differences in nucleotide sequences in the ompL1 gene types may not affect the immunogenicity and OmpL1 proteins, identifying OmpL1 as a genus-specific protein antigen, hence the cloned gene could be further used for expression and recombinant OmpL1 may be a useful vaccine candidate against leptospirosis.

Keywords: Leptospirosis; Sequencing; OmpL1; Molecular Analysis