Generation of recombinant *Helicobacter pylori* ghosts by PhiX174 E-mediated mechanism as a putative vaccine candidate

Thesis
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Abstract
High rate of *Helicobacter pylori* (Hp) infection in developing countries is a major risk factor for gastrointestinal disorders which due to its several associated complications accurate population screening, preventive and therapeutic vaccination approaches are critical.

Methods: After electro-transformation of lysis vector, Hp ghosts were generated by the induction of lysis gene E expression from bacteriophage PhiX174. Recombinant Omp18 protein was expressed in E. coli not only to investigate its role in therapeutic vaccination of Hp infected mice when loaded in Hp bacterial ghosts but also for its application in serological screening approaches. In brief, *omp18* gene was amplified from Iranian Hp strain and fused to IPTG inducible promoter by soeing PCR. The amplified fragment was cloned into pHel2 shuttle expression vector and its identity was confirmed. rOmp18 was expressed under IPTG induction and confirmed by SDS-PAGE and immuno-blotting with specific Hp positive and negative pooled sera.

For therapeutic vaccination trial, rOmp18 loaded HPBGs were orally administered to Hp-infected C57BL/6 mice. RUT and antigen-specific serologic assays were used to assess the efficacy of immunization.

Results: The efficiency of the lysis procedure was confirmed through several tests. Loading of HPBGs with rOmp18 was also confirmed by immunoblotting.

Comparative analysis of immuno-blotting assays revealed that rOmp18 has the highest criteria in comparison with tested recombinant proteins. Following oral immunization, rOmp18 loaded-HPBG plus cholera toxin stimulated significant production of serum specific antibodies in the immunized mice, which coincided with a significant reduction of bacterial colonization of the resected stomachs relative to controls (P<0.05).

Conclusion: This study recommends rOmp18 loaded HPBGs as suitable vaccine candidates in reducing Hp colonization rate. On the other hand, Omp18 protein can be recommended as a reliable serologic marker for accurate detection of Hp infection.

Keywords: Bacterial ghost, cloning, outer membrane protein18, immuno-blotting, therapeutic vaccine.