## Cloning, Sequencing and Expression of *helicobacter pylori hpaA* gene in *E. coli* host

<u>Neda Soleimani</u><sup>\*1</sup>, Ashraf Mohabati Mobarez <sup>2</sup>

- 1. PhD student, Dept of Bacteriology, Faqulty of Medical sciences, Tarbiat Modares University.
- 2. Associated professor, Department of Bacteriology, School of Medical Sciences, Tarbiat Modares

University, Tehran, Iran.

n.soleimani@modares.ac.ir

**Introduction and objective**: *Helicobacter pylori* is a widely distributed gram negative bacterium that infects the human stomach and duodenum. *HpaA* is a *Helicobacter pylori* specific lipoprotein that has been shown to be an effective protective antigen for mucosal vaccination against *H. pylori* infection in mice. *hpaA* of *helicobacter pylori* as a vaccine antigen is fully competent for stimulation of immune responses and here we report the expression of the protein in a recombinant form.

**Materials and methods:** The *hpaA* gene was inserted into pET28a (+) as cloning and expression vectors respectively. The recombinant plasmid (pET-hpaA) was subjected to sequencing other than PCR and digestion analysis. Protein expression was induced by adding 1mM isopropyl- $\beta$ -D-thiogalactoside to cultures of *E. coli* strain BL21 transformed with pET-hpaA. Protein expression assessed with SDS-PAGE analysis.

**Results:** The restriction endonuclease digestion, PCR amplification analysis showed that the *hpaA* gene of 730 bp was amplified from *helicobacter pylori* DNA and Sequencing analysis of the pET-hpaA confirmed the cloning accuracy and in frame insertion of *hpaA* fragment. SDS-PAGE analysis showed the expression of an approximately 29000 Dalton protein.

**Conclusions:** sequencing results along with SDS-PAGE analysis confirms the expression of recombinant hpaA in the heterologous *E.coli* BL21.

Key words: recombinant hpaA, helicobacter pylori, E.coli BL21, SDS-PAGE.