Porphyromonas gingivalis FimA Type II - PVXCP Fusion DNA Vaccine Expression in Mammalian Cells

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Abstract

Objective: It has been shown that *Porphyromonas gingivalis (P. gingivalis)* FimA is able to induce antibody production and protection against bone loss in animal model. *P. gingivalis* FimA is therefore the candidate antigen in periodontitis vaccine production. In this study, DNA vaccine was produced by fusion sequences of FimA type II, the most prevalent type associated with periodontitis, and potato virus X coat protein (PVXCP), the immune-enhancing molecule. The ability of this DNA vaccine to be expressed in mammalian cells was investigated.

Methods: The DNA vaccine was constructed in a fusion form of FimA and PVXCP (FimA -PVXCP) DNA vaccine, in which the mammalian expression plasmid pcDNA3 was used as a backbone plasmid. FimA type II gene was amplified from genomic DNA of P.gingivalis FimA type II by PCR. The FimA PCR product was inserted into predigested pcDNA3 containing PVXCP sequence; and consequently FimA was fused to the N-terminal side of PVXCP. The resulting plasmid was transfected into human embryonic kidney (HEK293) cells. FimA-PVXCP RNA expression in transfected cells was detected by RT-PCR. The fusion protein inside the cells and the protein secreted into medium were analyzed by Western Blot analysis using anti-PVXCP antibody.

Results: RT-PCR of RNA extracted from pcDNA3. FimA-PVXCP transfected HEK293 cells showed the expected band size of 1.9 kb of FimA-PVXCP sequence. FimA-PVXCP protein was detected by Western blot analysis both in cell lysate of the transfected cells and in the medium. However, the secreted protein appeared to be larger than the protein remained inside the cells.

Conclusion: FimA-PVXCP DNA vaccine was able to be expressed in HEK293 cells as confirmed by RT-PCR and Western blot analysis.

Keywords *: P.gingivalis* vaccine, Periodontitis vaccine, DNA vaccine, Protein expression in mammalian cells

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