

Designing, Optimization and Construction of Myelin Basic Protein Coding Sequence Binding to the Immunogenic Subunit of Cholera Toxin

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Abstract

Background and Objectives: Multiple sclerosis (MS) is a chronic inflammatory autoimmune disease. Mucosal feeding of myelin basic protein binding to the cholera toxin B subunit can reduce the intensity of the immune response in MS patients. Expression system, the domain composition of the fusion protein, accessibility of two domains, codon adaptation index (CAI) and GC contents are very important for the large scale production of fusion protein.

Material and Methods: we used DNA2, PSIPRED and ProtParam softwares for designing the best form to produce fusion protein. Moreover, the correct open reading frame of myelin basic protein was also considered. First the coding sequence was verified and then synthesized. For confirmation of the recombinant vector, PCR test was carried out using T7 primers. Finally it was inserted into the cloning site of pET28 expression vector.

Results: After coding optimization, the CAI rate was increased from 64 % to 80% and GC content from 41 % to 49%. The presence of a band near 700bp resulted from PCR amplification test demonstrates the correct cloning of recombinant vectors in the cloning site of pET28 expression vector.

Conclusion: According to software and experimental analysis, the designed sequence probably in the best form could be used for production of recombinant protein.

Keywords: Multiple Sclerosis, Cholera Toxin, Myelin Basic Protein