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Codon optimization, cloning and expression of Crimean-Congo hemorrhagic fever virus Gn glycoprotein by baculovirus expression system

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Crimean-Congo Hemorrhagic Fever Virus (CCHFV) is a member nairovirus genus from Bunyaviridae family, which causes life threatening disease in human with mortality rates of approximately 30%. CCHFV has been reported in more than 30 countries in Asia, Europe and Africa and is endemic in Iran that is considered as an important health problem by health authorities. The CCHFV was detected in Iran for the first time in 1970. The presence of antibodies against CCHFV in Iran peoples was confirmed in 1975. The disease has been reported from many provinces of the country but Sistan-Baluchestan province can be considered as a hot spot for the disease. Many people are infected with the virus through different routes of transmission and unfortunately some of them lose their life annually. Currently, there is no licensed vaccine against CCHFV. CCHFV encodes two glycoproteins known as Gn and Gc. The surface proteins of the virus are important in vaccine designing studies, since they are involved in early steps of interactions between the pathogen and the host cell receptors and also are targets for neutralizing antibodies. Studies show that antibodies against Gn can be protective in mice. To express the Gn protein in insect cells, that can be used as antigen in animal model vaccine studies, CCHFV Gn coding region was codon optimized for Sf9 cells, sub cloned and expressed in Sf9 cells.

Materials and methods: The M segment sequence of CCHFV (accession no: DQ446216.1) was retrieved from NCBI database, then coding sequence of Gn was selected and used for further studies. V5- tag sequence was added to construct to detect recombinant protein in western blot analysis. The Sf9 cells were transfected by constructed recombinant bacmid using cellfectin. Expression of Gn was performed by infection of approximately 8×10⁵ Sf9 cells using P4 stock of viruses. Total protein of the cell lysates was visualized on a 12% PAGE and Western blot analysis with anti-v5 antibody demonstrated expression and production of recombinant protein with 37 kDa.

Conclusion: Currently there are no specific treatments or licensed vaccine available for CCHFV and because CCHFV is a health problem in some countries of the world and also in our country, any attempt on developing a vaccine candidate for this virus will be extremely important. CCHFV recombinant Gn protein that was expressed in Sf9 cells could be used as a peptide vaccine in animal model studies also as diagnostic tools in research laboratories.

Biography

Mehdi Rahpeyma has completed his undergraduate degree in Microbiology. Because of his interest to viruses, he continued MS studies in medical Virology in Ahvaz University School of Medicine. Then his carrier was as scientific board at Yazd University School of Medicine for 3 years. He participated in government health ministry PhD grant exam and passed the exam successfully and currently is PhD student at Ahvaz Medical sciences University, Iran. Now he is working on viral glycoproteins and protein purification.

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