Purification and Bioassay of pertussis toxin from *Bordetella pertussis* (vaccinal strain 509) based on CHO-cell test

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Abstract

Pertussis toxin (PT), the main virulence factor of *Bordetella pertussis* is a protein-based AB5-type exotoxin. The methods for purification of pertussis toxin are not available exactly because of economic considerations by vaccine companies. The aim of this study was to setup and modify an in-house method for the PT purification based on affinity chromatography to develop acellular pertussis vaccine in future. *B. pertussis* and CHO cells were provided from Razi Institute (Karaj, Iran). The bacteria were grown in a 300 L fermenter (44 h, 35 °C, in B2 medium). The fermentation broth was clarified and concentrated by 0.45 µm membrane filter and 10 KDa molecular weight cut-off membrane, respectively. The isolation of pertussis toxin was performed based on affinity chromatography by Fetuin Sepharose column. Immune dot blot test showed significant amounts of pertussis toxin qualitatively. The clustering of CHO- cells mono-layer were observed after first hour of applying the purified pertussis toxin and stopped after the 12th hour. The average amount of extracted PT was 2.53 IU/ml± 0.43. Among the production procedure of whole cell pertussis vaccine, culture broth was discarded, whereas, the results showed that it was a suitable source for extraction of pertussis toxin. Finally examine other strains and bacterial culture methods to obtain desired pertussis toxin are recommended.

Key words: Whooping cough, Pertussis toxin, Bordetella pertussis, Affinity chromatography, CHO-cell test.