

عنوان مقاله:

Isolation and purification of ESAT6/CFP10 complex antigen secreted by Mycobacterium tuberculosis strains C

محل انتشار:

بیستمین کنگره بین المللی میکروب شناسی ایران (سال: 1398)

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خلاصه مقاله:

Introduction and Objectives: Tuberculosis (TB), caused by *Mycobacterium tuberculosis*, is one of the most prevalent infectious diseases. The pathogenesis of *Mycobacterium tuberculosis* is related to its proteins; the early secretory antigenic target (ESAT6) and culture filtrate protein-10 (CFP10), expressed from the region of deletion-1 (RD1). These proteins are highly specific and potentially useful for the diagnosis of tuberculosis. This research focused on purification of low molecular weight proteins from *M. tuberculosis* C vaccinal strain by ammonium sulphate precipitation. **Materials and Methods:** *M. tuberculosis* C vaccinal strain grown in Löwenstein-Jensen medium and Dorset Henley liquid medium were inactivated by subjecting to 68°C for 90 min, filtered, and the proteins in the supernatant fluid precipitated with ammonium sulphate at 4°C. The obtained precipitates were dialyzed and subjected to gel chromatography (G-50) and the obtained fractions analyzed for protein concentrations by Lowry method. The approximate molecular weight in the purified fractions were determined by SDS-PAGE. Finally, ESAT6 and CFP10 protein complex in the purified fractions were confirmed by western blot analysis using ESAT6 specific antibody. **Results:** Maximum amount of the proteins were precipitated in the presence of 20 % ammonium sulphate. During SDS-PAGE analysis, protein bands of approximately 10-15 kDa were observed. Purity of the proteins preparations was $\geq 95\%$, as judged by sodium dodecyl sulfate (SDS)-PAGE and subsequent staining with Coomassie blue. The presence of ESAT-6/CFP10 complex in the partially purified fractions were confirmed by Western blot analysis. **Conclusion:** Sufficient amounts of ESAT-6/CFP-10 purified by the mentioned method might be suitable for the development of diagnostic kit for *Mycobacterium tuberculosis* in future.

کلمات کلیدی:

Mycobacterium tuberculosis, ESAT-6/CFP10, purification, chromatography, SDS-PAGE, Western blotting

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