Separation and Characterization of Micobacterium bovis Cell Surface Lysate Antigen

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Abstract : Improving the early diagnosis of tuberculosis and solving a number of problems associated with the differential diagnosis of Mycobacterium bovis infection, nonspecific tuberculin reactions caused by sensitization of the body by non-tuberculosis mycobacteria, is urgent. The filtrates and extracts of M. bovis cell surface components are promising antigens with diagnostic potential. The purpose of this study was to isolate and characterize antigenic proteins and determine the dominant M. bovis antigens recognized by the humoral immune system. The mycobacterial cells were homogenized on FastPrep-24. Gel-filtration chromatography was used to fractionate the lysates of cell surface component extracts and proteins isolated from M. bovis culture supernatant. The separated fractions were analyzed using two-dimensional gel electrophoresis followed by determination of antigen serological activity using immunoblot with specific hyperimmune rabbit blood serum. As a result of electrophoretic separation of components by molecular weight, 23 antigen fractions were obtained. Analysis of densitograms showed that the fractions contained two zones of antigens with pronounced serological activity, corresponding to molecular weights of 28 and 21 kDa. The high serological activity of the 28 kDa antigen was established by immunoblot using hyperimmune blood sera. Separated and characterized by M. bovis specific antigen with a molecular weight of 28 kDa was added to the collection of specific marker antigens for M. bovis.

Keywords : antigen, gel-filtration chromatography, immunoblot, Mycobacterium bovis

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