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PRODUCTION OF A HUMAN RECOMBINANT ANTIBODY AGAINST SEROTYPE A CANDIDA ALBICANS

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Abstract:

After using 3 different generations of antibodies including human and non-human hyperimmune sera, monoclonal antibodies and chimeric antibodies, more recently a newer approach has been developed in which the antibody genes are cloned directly from a patient peripheral B-lymphocytes and expressed in a host like E. coli. In this study the Candida albicans serotype A (NCTC 3153) mannan was purified using a modified Fehling method and used for selection of human recombinant antibody from a C. albicans phage antibody library. After four rounds of affinity selecting (panning), 2 predominant clones were chosen by DNA fingerprinting and ELISA. A 248 amino acid DNA fragment coding for anti-C. albicans mannan scFv was sequenced and cloned in a pBAD-TOPO cloning vector to produce a soluble and phage free antibody. The analysis of antibody sequences by V base Index (DNAPLOT) confirmed the human antibody origin with the VH4 family in V segment of heavy variable chain and VL3 (Lambda 3) in J segment of the light variable chain. This antibody fragment was purified using immobilized metal affinity chromatography and inmmunoblotted as a 31kDa recombinant protein.

Keywords:

human recombinant antibody ، scFv ، protein purification

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