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High level expression of a functional humanized single-chain variable fragment (scFv) antibody against CD22 marker in *Pichia pastoris*

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ScFvs have recently emerged as attractive candidates in targeted immunotherapy of various malignancies. The anti-CD22 scFv is able to target CD22, on B-cell surface and is being considered as a promising molecule in targeted immunotherapy of B-cell malignancies. The recombinant anti-CD22 scFv has been successfully expressed in *Escherichia coli*; however, the insufficient production yield has been a major bottleneck for its therapeutic application. *Pichia pastoris* has become a highly popular expression host for the production of antibody fragments. In this study we used the *Pichia* expression system to express a model scFv against CD22. The full-length humanized scFv gene was introduced into the expression vector pPICZαA under the control of the AOX1 promoter and expressed in GS115 strain. The maximum production level of the scFv were achieved at methanol concentration, 1%; pH, 6.0; inoculum density, $OD_{600}=3$ and the induction time of 72 hours. Using affinity chromatography, 72% of the recombinant protein was recovered with a purity and final yield of about 93% and 25 mg/L, respectively. The correlation between scFv gene dosage and expression level was also investigated by real time PCR and the results confirmed the presence of such correlation up to 5 gene copies. Flowcytometry and biacore analysis demonstrated binding to CD22 on the surface of human lymphoid cell line Raji and recombinant CD22, respectively. Taken together, the presented data suggest that the *Pichia pastoris* can be considered as an efficient host for the large-scale production of anti-CD22 scFv as a possible promising carrier for targeted drug delivery.

Biography

Najmeh Zarei has completed her PharmD degree and PhD in the field of Pharmaceutical Biotechnology from Pasteur Institute of Iran. She is currently working as an Assistant Professor of the Pasteur Institute of Iran and has published more than 10 papers in reputed journals.

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