Expression, Purification, and Refolding of Recombinant Fusion Protein hIL-2/mGM-CSF[1]

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Objective To study the activities of interleukin (IL)-2 and granulocyte-macrophage colony-stimulating factor (GM-CSF) (hIL-2/mGM-CSF). **Methods** SOE PCR was used to change the linker of the fusion protein for higher activities. The fusion protein was expressed in *Escherichia coli* (*E. coli*) BL21 (DE3) in inclusion body (IB) form. After IB was extracted and clarified, it was denatured and purified by affinity chromatography. The protein was refolded by dilution in a L-arginine refolding buffer and refined by anion chromatography. The protein activity was detected by cytokine-dependent cell proliferation assay. **Results** The expression of hIL-2/mGM-CSF in *E. coli* yielded approximately 20 mg protein /L culture and the purity was about 90%. The specific activities of IL-2 and GM-CSF were 5.4×10^6 IU/mg and 7.1×10^6 IU/mg, respectively. **Conclusion** This research provides important information about the anti-tumor activity of hIL-2/mGM-CSF *in vivo*, thus facilitating future clinical research on hIL-2/mGM-CSF used in immune therapy.

Key words: HIL-2/mGM-CSF; Fusion protein; Purification; Refolding

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