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Expression and purification of rhIL-10-RGD from *Escherichia coli* as a potential wound healing agent

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Abstract：Various protocols for recombinant Interleukin-10 (IL-10) purification in wound healing have been reported previously. However, the therapeutic effect was not obvious. Thus, it is of great importance to find new and effective approaches for therapy. In this study, we propose that IL-10 and Arginine-Glycine-Aspartic (RGD) peptide would be a valuable therapeutic for wound healing. To explore a high-efficiency and cost-effective approach for the production of IL-10 and RGD peptide with bioactivity, a synthetic gene was cloned into a recombinant pTWIN1 vector. As a consequence, rhIL-10-RGD and the pH-induced self-cleavable *Ssp DnaB* mini-intein as a fusion protein was highly expressed by IPTG induction in *Escherichia coli* Rosetta without extra residues in a bioreactor. After Ni affinity chromatographic purification, rhIL-10-RGD was released by the *Ssp DnaB* intein-mediated self-cleavage that is triggered by pH shift. SDS-PAGE and silver staining showed a major band with an estimated molecular mass of 19.3 kDa. Cell proliferation assay confirmed its potent proliferation activity on MC/9 murine mast cells. In conclusion, we report a novel strategy to produce rhIL-10-RGD mediated by the pH-induced self-cleavable *Ssp DnaB* mini-intein, and show that rhIL-10-RGD could play an effective role in wound healing of BALB/c mice.

Keywords: rhIL-10-RGD; fusion protein; purification; wound healing