Recombinant production of Soluble Tumor Necrosis Factor-Related Apoptosis-Inducing Ligand (sTRAIL) as a therapeutic protein

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Abstract: Successfully cancer therapies aim to induce apoptosis in cancer cell lines. Recent advances in cancer therapy based on the use of some recombinant proteins such as tumor necrosis factor-related apoptosis-inducing ligand (TRAIL). TRAIL is a new member of the TNF superfamily. In this paper, we report the expression, purification, and preparation of a recombinant form of the extracelluar domain of the TRAIL (sTRAIL) in *Escherichia coli* rosetta gami under the control of T7 promoter; which may selectively induce apoptosis of tumor cells in vitro. To obtain recombinant sTRAIL protein, the encoding region for sTRAIL was cloned between *Xho*1 and *Bam*HI in pET28a expression vector. The results showed that the recombinant sTRAIL was efficiently produced in *Ecoli* rosetta gami strain.

Introduction: Apoptosis is an evolutionarily conserved and essential for maintenance of tissue homeostasis and removal of unwanted cells. TRAIL belongs to the group of therapeutic agents selectively targeting a wide variety of cancer cells without affecting the normal cells .The therapeutic potential of TRAIL is attributed to its receptor expression in a variety of tissues; which initiates apoptosis in cancer cells through interaction with the death receptors DR4 and DR5. Due to its selective nature, it is considered as a significant therapeutic agent in cancer therapy. The purpose of this study was to produce recombinant human sTRAIL in Rosetta Gami2 *E.coli* strain and its functions on cancerous cells in vitro.

Methods and results: we optimized the coding sequence of this protein. The recombinant plasmid was transformed into Rosetta Gami2 *E.coli* strain for expression. The transformed bacteria which contain recombinant plasmid were cultured in 37ºC with 250 rpm in LB and in 20ºC in TB medium for 18 hours. TRAIL was purified by Ni sepharose column, and the presence of the recombinant protein was confirmed by SDS-PAGE. The concentration of purified protein was measured by Bradford assay. Our finding showed that the recombinant protein (34kD) has been successfully produced for next experiments, the purified protein was desalted and applied toward cancerous cells.

Conclusions: In summary, TRAIL can be considered as a promising therapeutic agent for effective, targeted and less toxic agents for treatment of cancers.

Key words: TRAIL, Codon optimization, Rosetta gami2, Cancer therapy