

Expression of a functional human Tumor Necrosis Factor- α in yeast *Saccharomyces cerevisiae*

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Abstract

Tumor necrosis factor- α (TNF- α), a 17.4 kDa protein produced by macrophage and other cells, is a potent lymphoid factor which exerts cytotoxic effects on a wide range of tumor cells and certain other target cells.⁽¹⁾ The recombinant human Tumor necrosis factor-alpha (rhTNF- α) was cloned and expressed in yeast *Saccharomyces cerevisiae*. Two different promoters for heterologous expression of hTNF- α were tested: glyceraldehyde-3-phosphate dehydrogenase (*GPD*) promoter and a yeast hybrid *ADH2-GPD* promoter consisting of alcohol dehydrogenase II (*ADH2*) and *GPD* promoter.^(2,3) Northern blot analysis revealed that, although variation in the expression level of hTNF- α existed among transformants, the highest expression was obtained by the *GPD* promoter. Expressed hTNF- α protein (rhTNF- α) was successfully secreted into culture medium due to the presence of the signal peptide of rice amylase 1A. It was possible to produce 1.8 mg of rhTNF- α protein per liter of culture filtrate without any changes in cell growth. The secreted rhTNF- α had an estimated molecular mass of 55 kDa, which is considered that the rhTNF- α in culture filtrate exists mainly as trimer.

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References

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