Research in Pharmaceutical Sciences, 2012;7(5)
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Expression of reteplase in *Escherichia coli* top10 using arabinose promoter

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Background and Aims: Production of tissue plasminogen activator protein (t-PA) in prokaryotes systems has many problems such as lack of active protein production, multiple purification steps, and refolding process which has been shown to be costly and time consuming. In the present study TOP 10 *E. c*oli bacteria was utilized for the production of reteplase which is the nonglycosylated active domain of t-PA. Retplase gene was ligated into pBAD/gIII plasmid which allows secretion of this protein in periplasmic space which would allow the correct formation of disulfide bonds in protein structure.
Methods: Recombinant pET15b/reteplase plasmid was digested by NcoI and BamHI restriction enzymes while pBAD/gIII vector was digested by NcoI and BglII. Then the insert and vector were ligated and used for transformation *E.coli* Top10 cells by heat shock method. Overnight culture of transformed bacteria was induced by L-Arabinose in various concentrations (0/2, 0/02, 0/002 and 0/0002%) at various temperatures.
Results: The obtained recombinant plasmid was sequenced to confirm the presence and correct framing of reteplase gene regarding the expression of reteplase. Maximum production of this enzyme was obtained under the following condition: 0/0002% L-Arabinose (as inducer), at 37°C (tempreture of incubation). The production of reteplase was confirmed by Western blotting using Anti-TpA antibody.
Conclusions: Reteplase was expressed in *E. coli* after activation of pBAD/gIII promoter region by arabinose.

Keywords: Reteplase; Expression; *Escherichia coli*; L-Arabinose.