



Developing a method for T7 RNA polymerase production for applications in mRNA synthesis by in vitro transcription

mRNA has aroused in the last years as one of the most promising biologicals for the prophylaxis and treatment of many unmet medical needs, such as Covid-19 vaccines. In order to use mRNA as a pharmaceutical, it is required to produce it by in vitro transcription. Nevertheless, in vitro transcription has as main issue the costs of the enzymes required, namely T7 RNA polymerase (T7 RNAP). In this context, we propose this TFM as a multidisciplinary project focused on the expression and purification of a recombinant T7 RNAP enzyme in *E.coli* host, for applications in *in vitro* transcription. The objectives of this project are: 1) Cloning of an epitope-tagged T7-RNAP gene for expression *in E.coli*. 2) Expression, purification, and storage of the recombinant protein. 3) Functional characterization of produced T7 RNA polymerase enzyme and optimization of the in vitro transcription reactions to produce therapeutic mRNAs.

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