

Lethbridge iGEM 2017

NEB PURExpress Kit

The following protocol was obtained from the NEB Purexpress *In Vitro* Protein Synthesis Instruction Manual

- 1. Thaw Solution A, Solution B and template DNA on ice
- 2. Assemble reaction on ice in new tube in the following order for a 25 µL reaction:

 $\begin{array}{ccc} \text{Solution A} & & 10 \; \mu\text{L} \\ \text{Solution B} & & 7.5 \; \mu\text{L} \\ \text{Supplements} & & x \; \mu\text{L} \end{array}$

(RNase Inhibitor, etc.)

Nuclease-free H2O $\qquad \qquad x \ \mu L$ Template DNA $\qquad \qquad x \ \mu L$

(recommended starting concentration of 250ng)

- 3. Mix gently and pulse-spin in microfuge to collect mixture at the bottom of the tube
- 4. Incubate at 37°C for 2 hours
- 5. Stop the reaction by placing the tube on ice
- 6. Use samples for analysis (SDS-PAGE) or purification or freeze at -20°C for use at later time