Combinatorial Modulation of Translation Rates in *Escherichia coli*

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The *Escherichia coli* has been the most popular host for recombinant protein expression due to its rapid growth, availability for high density fermentation and well-characterized genetics. It is, however, not unusual that heterologous proteins overexpressed in *E. coli* often lead to inclusion bodies. A traditional approach to alleviate the protein misfolding involves lowering the cultivation temperature to reduce the transcription and translation rates. Since the polypeptides being synthesized from the translating ribosomes are exposed to the cytosol vectorially and co-translationally assisted by the molecular chaperones to assume the native conformation, the kinetics of protein folding is closely related to the translation initiation and elongation rate.

Thus, to regulate the rate of protein synthesis here we constructed a mutant library with the various translation rates in *E. coli* by the direct switching of mutant genes with the original target gene in the chromosome. By combination with error-prone PCR, the functions of initiation factor 2 and elongation factor G could be easily modulated. The mutant library allows the various combinations of translation rates to provide more folding environments.