

A simple and dual expression plasmid system in prokaryotic (*E. coli*) and mammalian cells.

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We introduce a simple and universal cloning plasmid system for gene expression in prokaryotic (*Escherichia coli*) and mammalian cells. This novel system has two expression modes: the (subcloning) prokaryotic and mammalian modes. This system streamlines the process of producing mammalian gene expression plasmids with desired genes. The plasmid (prokaryotic mode) has an efficient selection system for DNA insertion, multiple component genes with rare restriction sites at both ends (termed "units"), and a simple transformation to mammalian expression mode utilizing rare restriction enzymes and re-ligation (deletion step). This system is highly efficient for the subcloning of blunt-end fragments, including PCR products. After the insertion of the desired gene, protein encoded by the desired gene can be detected in *E. coli* with IPTG induction. Then, the lac promoter and operator are readily deleted with 8-nucleotide rare-cutter blunt-end enzymes (deletion step). Following re-ligation and transformation, the plasmid is ready for mammalian expression analysis (mammalian mode). This idea (conversion from prokaryotic to mammalian mode) can be widely adapted. With pgMAX system, we made epitope-library of the calcium channel alpha1 subunit (CaV1.2) and found a novel binding site to calcium channel beta2 subunit. The pgMAX system could be widely adopted for simple expression analyses.