**1mL 5X In-Fusion Master Mix**

PEG 8000 25%

0.5M Tris-Hcl pH 7.5

50mM MgCl2

50mM DTT

1ul 10U/ul T5 exonuclease

1ul Pfu polymerase

1mM dNTP

Reference：

T5 exonuclease-dependent assembly offers a low-cost method for efficient cloning and site-directed mutagenesis，Nucleic Acids Res，2019 Feb 20;47(3):e15. doi: 10.1093/nar/gky1169.

Seamless Insert-Plasmid Assembly at High Efficiency and Low Cost， Roger M. Benoit ,Christian Ostermeier,Martin Geiser,Julia Su Zhou Li,Hans Widmer,Manfred Auer