



Description:

pLAS5w.PtRFP-I2-Puro was derived from pLAS5w.Ppuro by inserting an IRES-directed bicistronic cassette with the configuration of tRFP-IRES-Puro (from pLKO_AS1014) into the down stream of PGK promoter.

Location of Features (for other features, please refer to pLKO.1):

- Elongation factor 1 alpha promoter : nt10-1268
- Multiple cloning sites: nt1359-1478
- tRFP-I2-Puro expression cassette: nt1690-4188
- WPRE sequence: 4204-4792

Note:

1. Since lentiviral transfer vector may undergo sequence re-arrangement and/or deletion when its ligated products were transformed into DH5 α competent cells; to avoid that, Stbl3 (Invitrogen) is recommended for transformation.
2. If plasmid-transformed Stbl3 cells grow slowly, try to transform it into HB101 or GM2163 *E. coli* strain for large-prep.
3. Xba I (*) site is blocked by overlapping dam methylation.