



Description:

pLKO_AS1014 (TRC2 vector) was derived from pLKO_TRC014 with the following modification: PAC-F2A-tRFP ORF on TRC014 (the expression of tRFP is not sufficient) was replaced by an IRES-directed bicistronic cassette with the following configuration: tRFP-IRES-PAC. This results in the expression of tRFP and puromycin selection marker in transduced cells simultaneously. In addition, WPRE element was also added.

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

- Human U6 promoter: nt8380-8631.
- Stuffer : nt8632-10500.
- shRNA cloning sites: AgeI(nt8627)-EcoRI(nt10502)
- PGKp-tRFP-IRES-PAC cassette: nt4-2505.

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.