pMCs-Puro Retroviral Vector

CATALOG NUMBER: RTV-041 STORAGE: -20°C

QUANTITY AND CONCENTRATION: 10 μ g at 0.25 μ g/ μ L in TE

Background

Retroviruses are efficient tools for delivering heritable genes into the genome of dividing cells. Most retrovirus vectors including pBABE and pMXs are based on Moloney murine leukemia virus (MMLV). MMLV-based vectors usually are silenced in immature cells including embryonic carcinoma (EC) cells and embryonic stem (ES) cells, and possibly hematopoietic stem cells. PCC4-cell-passaged myeloproliferative sarcoma virus (PCMV) are mutants of MMLV and can stably express genes in immature cells including ES cells.

Cell Biolabs' pMCs-Puro retroviral vector includes hybrid LTRs containing elements from both MMLV and PCMV and is capable of expressing genes in both EC and ES cells. The vector provides the viral package signal, transcription and processing elements, and MCS for cloning of a target gene. The viral *env* gene, produced by the package cell line, encodes the envelope protein, which determines the viral infectivity range. Transfection into a package cell line produces high-titer, replication-incompetent viruses. In addition to transfer and expression of exogenous genes in mammalian cells, recently, retroviruses have been used to express silencing RNAs (siRNA) to decrease the expression of target genes both *in vitro* and *in vivo*.

The vector contains the ampicillin-resistance gene, LTRs, package signal and MCS for cloning of your gene of interest (Figure 1).

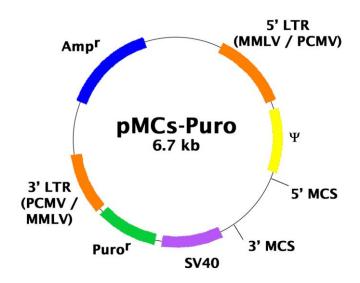


Figure 1. Schematic representation of pMCs-Puro retroviral vector.



5'-MCS:

- Enzyme Sites: 5'-PacI, BamHI, EcoRI-3'
- MCS Sequence: TTAATTAA<u>GGATCC</u>CAGTGTGGTGGTACGG<u>GAATTC</u>AAGCTTGATC

3'-MCS:

- Enzyme Sites: 5'-EcoRI, XhoI, NotI-3'
- MCS Sequence: GGCG<u>GAATTC</u>CAGCTGAGCGCCGGTCGCTACCATTACCAGTTGGTCTGGTGTCAAAAA TAATAATAACCGGGCAGGCCATGTCTGCCCGTATTTCGCGTAAGGAAATCCATTATGT ACTATTTAAA<u>CTCGAGCGGCCGC</u>CAGCACAGTG<u>GTCGAC</u>---SV40---puro-<u>GTCGAC</u>---

Note: For optimal expression, both 5' MCS and 3' MCS should be used to clone gene of interest and replace the stuffer sequence (partial LacZ) between them.

Safety Consideration

Remember that you will be working with samples containing infectious virus. Follow the recommended NIH guidelines for all materials containing BSL-2 organisms. Always wear gloves, use filtered tips and work under a biosafety hood.

References

1. Kitamura T., et al., (2003) Exp. Hematol. **31**, 1007-1014.

<u>Recent Production Citations</u>

- Handorf, A. et al. (2014). Endogenously Produced Indian Hedgehog Regulates TGFβ-Driven Chondrogenesis of Human Bone Marrow Stromal/Stem Cells. *Stem Cells Dev.* doi:10.1089/scd.2014.0266.
- 2. Tang, Y. et al. (2014). Differential effects of Akt isoforms on somatic cell reprogramming. *J Cell Sci.* **127**:3998-4008.
- 3. Mochizunki, Y. et al. (2013). Phosphatidylinositol 3-phosphatase myotubularin-related protein 6 (MTMR6) Is regulated by small GTPase Rab1B in the early secretory and autophagic pathways. *J. Biol. Chem.* **288**:1009-1021.

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