

pBABEpuro-PRAK Retroviral Vector (Dominant Negative)

CATALOG NUMBER: RTV-123

STORAGE: -80°C

QUANTITY AND CONCENTRATION: 100 µL of bacterial glycerol stock

Background

Retroviruses are efficient tools for delivering heritable genes into the genome of dividing cells. Cell Biolabs' retrovirus vector is based on the pBABE vector system, which is derived from Moloney murine leukemia virus (MMLV). The vector provides the viral package signal, transcription and processing elements, and a target gene. The viral *env* gene, produced by the package cell line, encodes the envelop 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 bacterial origin of replication, ampicillin-resistance gene, and puromycin-resistance gene for the growth of infected mammalian cells to select stable cell lines (Figure 1).

Mitogen-activated protein kinases (MAPK), including ERK1/2, p38, and JNK1/2, are important regulators of cell function. The ERK MAPKs are most frequently activated by mitogens, whereas the JNK and p38 MAPKs are strongly responsive to stress and inflammatory signals. The p38 MAPK family includes the p38 α , β , δ , and γ isoforms. PRAK activity is regulated by p38 α and p38 β *in vitro* and *in vivo* through phosphorylation. T182 within the activation loop of PRAK has been determined to be the regulatory phosphorylation site. Small heat shock protein 27 (Hsp27) and the regulatory light chain of myosin II have been shown to be potential substrates of PRAK. PRAK may play a role in balancing other MAPK pathways because overactivation of PRAK can inhibit Ras mediated cell proliferation and gene activation. A dominant negative form of human PRAK (A) is cloned into the retroviral vector pBABEpuro at the *Sna*B I site. The p38 phosphorylation site T182 has been changed to Ala.

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

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2. Coffin, J. M. and H. E. Varmus, *Retroviruses*, Cold Spring Harbor Press, NY.
3. Schuck S, Manninen A, Honsho M, Fullekrug J and Simons K. (2004) *Proc Natl Acad Sci U S A.* 101, 4912-4917.
4. New L., Jiang Y., Zhao M., Liu K., Zhu W., Flood L.J., Kato Y., Parry G.C., and Han J. (1998). *EMBO J.* 17:3372-3384.

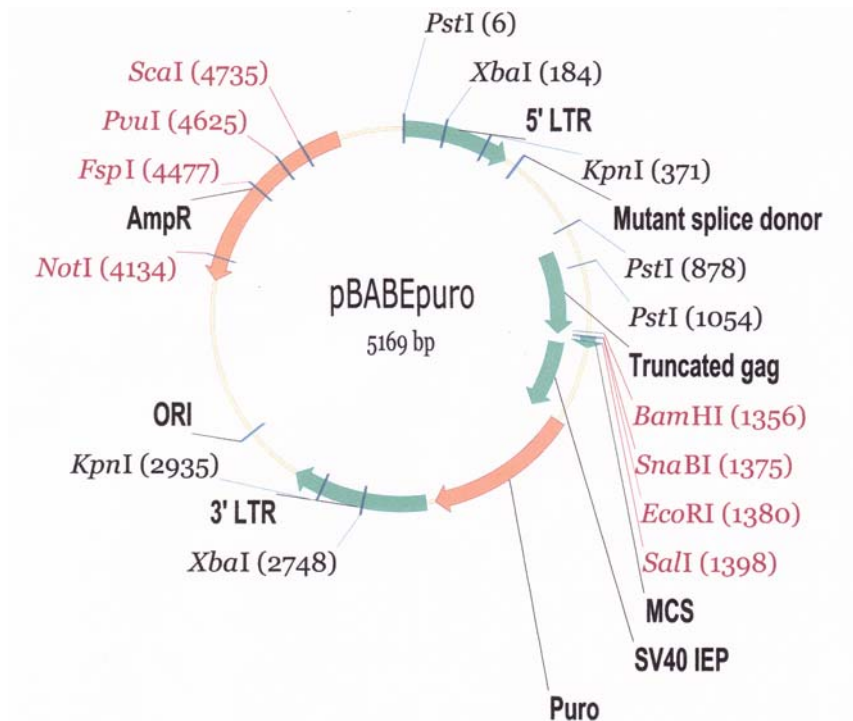


Figure 1. Retroviral Vector Map

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