

pBABEpuro-Ras V12S35 Retroviral Vector

CATALOG NUMBER: RTV-102

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 are 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).

Ras genes encode 21 kDa guanine nucleotide-binding proteins, including H-, K- and N-Ras. H-Ras was first identified as an oncogene, and mutated Ras genes have been found in many human tumors. Like all GTPases, Ras acts as a molecular switch to control downstream cellular events. The interconversion of the inactive GDP-bound form into the active GTP-bound form is regulated by guanine nucleotide exchange factors, whereas inactivation of the GTP-bound form is stimulated by GTPase-activating proteins (GAPs). Ras in its active GTP bound form binds to Raf, resulting in activation of the MAP kinase cascade. In mammals, Ras functions through a multiplicity of effectors. The use of effector loop mutants of Ras has shown that three effectors account for many, if not all, Ras functions: phosphatidylinositol 3-kinase (PI3K; a preferred effector of the Ras Y40C allele), Raf (a preferred effector of the Ras T35S allele), and Ral guanine nucleotide exchange factors (Ral GEFs; preferred effectors of the Ras E37G allele). A constitutively active form of human H-Ras (V12S35) is cloned into the retroviral vector pBABEpuro at the *Sna*B I site.

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. Marshall C. J., Lloyd A. C., Morris J. D., Paterson H., Price B and Hall A. (1989) *Int J Cancer Suppl.* 4:29-31.

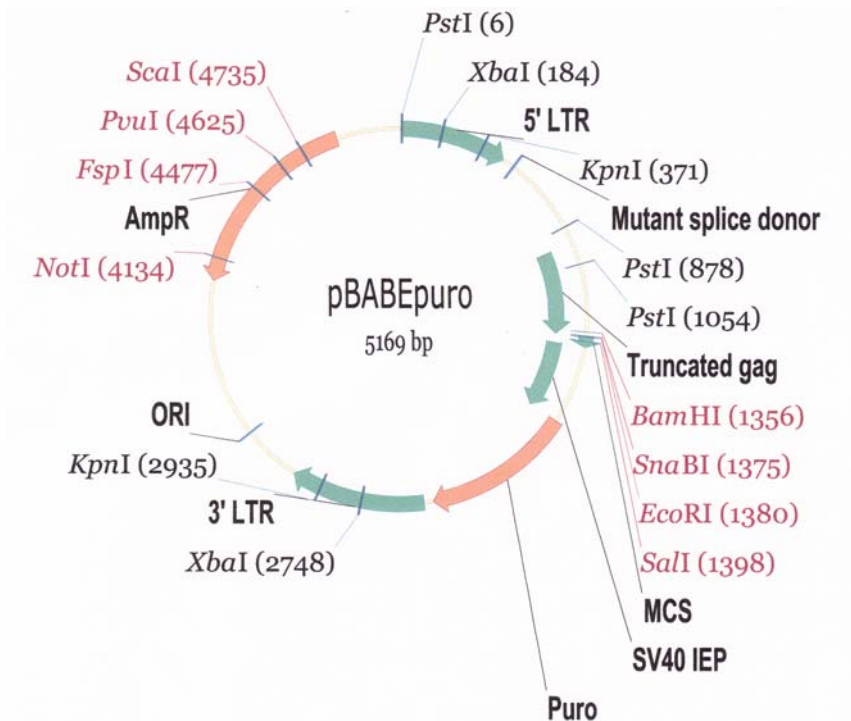


Figure 1. Retroviral Vector Map

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