SANTA CRUZ BIOTECHNOLOGY, INC.

VPRBP siRNA (m): sc-76899

BACKGROUND

Infection by human immunodeficiency virus (HIV) is associated with an early immune dysfunction and progressive destruction of $CD4^+$T lymphocytes. The HIV-induced, premature destruction of lymphocytes is associated with the continuous production of HIV viral proteins, which modulate apoptotic pathways. The virion-associated protein (Vpr), an accessory protein of HIV, affects viral replication, as well as cell growth, differentiation and apoptosis. Involved in the pathogenesis of T cell depletion in HIV-infected people, Vpr has been shown to enhance the nuclear transport of the HIV-1 pre-integration complex, activate transcription of cellular and viral promoters and arrest the cell cycle at the G_2/M checkpoint. VPRBP (Vpr (HIV-1) binding protein), also known as DCAF1 or RIP, is a 1,507 amino acid cytoplasmic protein that contains one LisH domain and functions as a Vpr binding protein. Expressed ubiquitously, VPRBP is thought to act as a receptor for the CUL-4-DDB1 complex and, in response to HIV infection, interacts with Vpr and may cause cell cycle arrest at the G_2 phase. Multiple isoforms of VPRBP exist due to alternative splicing events.

REFERENCES

1. Zhao, L.J., et al. 1994. Biochemical mechanism of HIV-I Vpr function. Specific interaction with a cellular protein. J. Biol. Chem. 269: 15577-15582.
2. Zhang, S., et al. 2001. Cytoplasmic retention of HIV-1 regulatory protein Vpr by protein-protein interaction with a novel human cytoplasmic protein VPRBP. Gene 263: 131-140.
3. Le Rouzic, E., et al. 2007. HIV1 Vpr arrests the cell cycle by recruiting DCAF1/VPRBP, a receptor of the CUL-4-DDB1 ubiquitin ligase. Cell Cycle 6: 182-188.
4. Borgne-Sanchez, A., et al. 2007. Targeted Vpr-derived peptides reach mitochondria to induce apoptosis of aVb3-expressing endothelial cells. Cell Death Differ. 14: 422-435.
5. Tan, L., et al. 2007. DDB1 and CUL-4A are required for human immunodeficiency virus type 1 Vpr-induced G_2 arrest. J. Virol. 81: 10822-10830.
6. Wen, X., et al. 2007. The HIV1 protein Vpr acts to promote G_2 cell cycle arrest by engaging a DDB1 and Cullin4A-containing ubiquitin ligase complex using VPRBP/DCAF1 as an adaptor. J. Biol. Chem. 282: 27046-27057.

CHROMOSOMAL LOCATION

Genetic locus: Vprbp (mouse) mapping to 9 F1.

PRODUCT

VPRBP siRNA (m) is a pool of 2 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see VPRBP shRNA Plasmid (m): sc-76899-SH and VPRBP shRNA (m) Lentiviral Particles: sc-76899-V as alternate gene silencing products.

For independent verification of VPRBP (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-76899A and sc-76899B.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNAses and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

VPRBP siRNA (m) is recommended for the inhibition of VPRBP expression in mouse cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 µM in 66 µl. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

VPRBP (C-8): sc-376850 is recommended as a control antibody for monitoring of VPRBP gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgGκ BP-HRP: sc-516102 or m-IgGκ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgGκ BP-FITC: sc-516140 or m-IgGκ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor VPRBP gene expression knockdown using RT-PCR Primer: VPRBP (m)-PR: sc-76899-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.