SANTA CRUZ BIOTECHNOLOGY, INC.

RIP/Rab siRNA (h): sc-40913

BACKGROUND

HIV-1 Rev is the prototype of a class of retroviral regulatory proteins that control the sequence-specific nuclear export and translation of a class of incompletely spliced HIV-1 mRNAs that encode viral structural proteins. In the absence of Rev, these late viral RNAs remain sequestered in the nucleus until they are either spliced or degraded. The protein designated Rev interacting protein (RIP) or Rev/Rex activation domain-binding protein (Rab) contains 562 amino acids. RIP/Rab has been identified as a cellular cofactor that binds not only to the HIV-1 Rev activation domain, but also to equivalent domains of other Rev and Rex proteins. On the basis of these findings it has been speculated that RIP/Rab is required for the Rev response and thus for HIV-1 replication.

REFERENCES

1. Bogerd, H.P., et al. 1995. Identification of a novel cellular cofactor for the Rev/Rex class of retroviral regulatory proteins. Cell 82: 485-494.
2. Fritz, C.C., et al. 1995. A human nucleoporin-like protein that specifically interacts with HIV Rev. Nature 376: 530-533.
3. Ragheb, J.A., et al. 1995. Analysis of trans-dominant mutants of the HIV type 1 Rev protein for their ability to inhibit Rev function, HIV type 1 replication, and their use as anti-HIV gene therapeutics. Aids Res. Human Retro. 11: 1343-1353.
4. Wu, B.Y., et al. 1995. Regulation of human retroviral latency by the NFκB/IκB family: inhibition of human immunodeficiency virus replication by IκB through a Rev-dependent mechanism. Proc. Natl. Acad. Sci. USA 92: 1480-1484.
5. Fischer, U., et al. 1995. The HIV-1 Rev activation domain is a nuclear export signal that accesses an export pathway used by specific cellular RNAs. Cell 82: 475-483.
6. Kubota, S., et al. 1996. Nuclear preservation and cytoplasmic degradation of human immunodeficiency virus type 1 Rev protein. J. Virol. 70: 1282-1287.
7. Bevec, D., et al. 1996. Inhibition of HIV-1 replication in lymphocytes by mutants of the Rev cofactor eIF-5A. Science 271: 1858-1860.

CHROMOSOMAL LOCATION

Genetic locus: AGFG1 (human) mapping to 2q36.3.

PRODUCT

RIP/Rab siRNA (h) 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 RIP/Rab shRNA Plasmid (h): sc-40913-SH and RIP/Rab shRNA (h) Lentiviral Particles: sc-40913-V as alternate gene silencing products.

For independent verification of RIP/Rab (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-40913A and sc-40913B.

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

RIP/Rab siRNA (h) is recommended for the inhibition of RIP/Rab expression in human 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

RIP/Rab (H-2): sc-166651 is recommended as a control antibody for monitoring of RIP/Rab 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 RIP/Rab gene expression knockdown using RT-PCR Primer: RIP/Rab (h)-PR: sc-40913-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.