Pim-1 siRNA (r): sc-270539



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BACKGROUND

Pim-1 is a serine/threonine kinase that cooperates with c-Myc in lymphoid cell transformation. The expression of Pim-1 increases during the progression from early to late G_1 , remaining high at the G_1 /S boundary and G_2 phases of the cell cycle. Pim-1 is regulated at both the transcriptional and translational level, and it has been shown to be induced by IL-2 stimulation. Pim-1 also plays a role in T cell differentiation, and it has been shown to stimulate c-Myc-mediated apoptosis upstream of caspase-3-like proteases.

REFERENCES

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- Rohwer, F., et al. 1996. The effect of IL-2 treatment on transcriptional attenuation in proto-oncogenes Pim-1 and c-Myb in human thymic blast cells. J. Immunol. 157: 643-649.
- 3. Hoover, D.S., et al. 1997. Pim-1 protein expression is regulated by its 5'-untranslated region and traslation initiation factor eIF-4E. Cell Growth Differ. 8: 1371-1380.
- Mochizuki, T., et al. 1997. Pim-1 kinase stimulates c-Myc-mediated death signaling upstream of caspase-3 (CPP32)-like protease activation. Oncogene 15: 1471-1480
- Schmidt, T., et al. 1998. Evidence implicating Gfi-1 and Pim-1 in pre-T cell differentiation steps associated with β-selection. EMBO J. 17: 5349-5359.
- 6. Leverson, J.D., et al. 1998. Pim-1 kinase and p100 cooperate to enhance c-Myb activity. Mol. Cell 2: 417-425.

CHROMOSOMAL LOCATION

Genetic locus: Pim1 (rat) mapping to 20p12.

PRODUCT

Pim-1 siRNA (r) 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Pim-1 shRNA Plasmid (r): sc-270539-SH and Pim-1 shRNA (r) Lentiviral Particles: sc-270539-V as alternate gene silencing products.

For independent verification of Pim-1 (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-270539A and sc-270539B.

STORAGE AND RESUSPENSION

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

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

APPLICATIONS

 $\operatorname{Pim-1}$ siRNA (r) is recommended for the inhibition of $\operatorname{Pim-1}$ expression in rat 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

Pim-1 (G-11): sc-374116 is recommended as a control antibody for monitoring of Pim-1 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-lgG κ BP-HRP: sc-516102 or m-lgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-lgG κ BP-FITC: sc-516140 or m-lgG κ 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 Pim-1 gene expression knockdown using RT-PCR Primer: Pim-1 (r)-PR: sc-270539-PR (20 μ I, 584 bp). 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.

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