PI31 siRNA (h): sc-76129



The Power to Question

BACKGROUND

Proteasomes are the catalytic sites of intracellular protein degradation via proteolysis. The most common form of proteasome, the 26S Proteasome, is composed of a cylindrical-shaped 20S core particle accompanied by two 19S regulatory subunits. Pl31 (proteasome inhibitor Pl31 subunit) is a 271 amino acid protein that is a potent inhibitor of proteasome activity. Pl31 is a proline-rich protein, particularly within its C-terminal domain, where nearly one-quarter of the amino acids are proline. Working within the ubiquitin-dependent pathway, Pl31 inhibits the hydrolysis of protein and peptide substrates by the 20S proteasome and also inhibits proteasome activation by PA700 and PA28. Pl31 functions as a monomer and localizes to the nuclear envelope and endoplasmic reticulum membrane.

REFERENCES

- 1. Zaiss, D.M., et al. 1999. The proteasome inhibitor Pl31 competes with PA28 for binding to 20S proteasomes. FEBS Lett. 457: 333-338.
- McCutchen-Maloney, S.L., et al. 2000. cDNA cloning, expression, and functional characterization of Pl31, a proline-rich inhibitor of the proteasome.
 J. Biol. Chem. 275: 18557-18565.
- 3. Zaiss, D.M., et al. 2002. Pl31 is a modulator of proteasome formation and antigen processing. Proc. Natl. Acad. Sci. USA 99: 14344-14349.
- 4. Dahlmann, B. 2005. Proteasomes. Essays Biochem. 41: 31-48.
- Gharbi, S.I., et al. 2007. Exploring the specificity of the PI3K family inhibitor LY294002. Biochem. J. 404: 15-21.
- 6. Kuznetsova, E.B., et al. 2007. Novel methylation and expression markers associated with breast cancer. Mol. Biol. 41: 624-633.
- 7. Cui, Y., et al. 2008. Proteomic analysis of testis biopsies in men treated with injectable testosterone undecanoate alone or in combination with oral levonorgestrel as potential male contraceptive. J. Proteome Res. 7: 3984-3993.

CHROMOSOMAL LOCATION

Genetic locus: PSMF1 (human) mapping to 20p13.

PRODUCT

Pl31 siRNA (h) is a pool of 3 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 Pl31 shRNA Plasmid (h): sc-76129-SH and Pl31 shRNA (h) Lentiviral Particles: sc-76129-V as alternate gene silencing products.

For independent verification of Pl31 (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-76129A, sc-76129B and sc-76129C.

PROTOCOLS

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

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 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

PI31 siRNA (h) is recommended for the inhibition of PI31 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

Pl31 (26): sc-136416 is recommended as a control antibody for monitoring of Pl31 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 Pl31 gene expression knockdown using RT-PCR Primer: Pl31 (h)-PR: sc-76129-PR (20 μ l). 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.

Santa Cruz Biotechnology, Inc. 1.800.457.3801 831.457.3801 Fax 831.457.3801 Europe +00800 4573 8000 49 6221 4503 0 www.scbt.com