

Prostasin siRNA (h): sc-63314

BACKGROUND

Prostasin, a serine protease first identified in prostate tissue, activates epithelial sodium channels in a variety of tissues. Though typically a membrane-anchored protein, free Prostasin is also found in physiologic fluids and tissue culture media, indicating a mechanism for secretion from the cells as well. Aprotinin and other protease inhibitors suppress the channel-activating capacity of Prostasin, while Aldosterone increases Prostasin expression and stimulates sodium uptake. In addition, DNA methylation negatively correlates with Prostasin expression in cancer cells, while enforced reexpression decreases invasiveness as well as metastatic potential, implying that Prostasin activity reflects epithelial cell physiology.

REFERENCES

1. Liu, L., et al. 2002. Serine protease activity in M-1 cortical collecting duct cells. *Hypertension* 39: 860-864.
2. Nariaki, T., et al. 2002. Regulation of Prostasin by Aldosterone in the kidney. *J. Clin. Invest.* 109: 401-408.
3. Chen, L.M., et al. 2002. Prostasin serine protease inhibits breast cancer invasiveness and is transcriptionally regulated by promoter DNA methylation. *Int. J. Cancer* 97: 323-329.
4. Wang, C., et al. 2003. Adenovirus-mediated human Prostasin gene delivery is linked to increased aldosterone production and hypertension in rats. *Am. J. Physiol. Regul. Integr. Comp. Physiol.* 284: 1031-1036.
5. Iwashita, K., et al. 2003. Inhibition of Prostasin secretion by serine protease inhibitors in the kidney. *J. Am. Soc. Nephrol.* 14: 11-16.

CHROMOSOMAL LOCATION

Genetic locus: PRSS8 (human) mapping to 16p11.2.

PRODUCT

Prostasin 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Prostasin shRNA Plasmid (h): sc-63314-SH and Prostasin shRNA (h) Lentiviral Particles: sc-63314-V as alternate gene silencing products.

For independent verification of Prostasin (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-63314A, sc-63314B and sc-63314C.

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

Prostasin siRNA (h) is recommended for the inhibition of Prostasin 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 μ M in 66 μ l. 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

Prostasin (C-10): sc-514983 is recommended as a control antibody for monitoring of Prostasin 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-IgG κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG 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 Prostasin gene expression knockdown using RT-PCR Primer: Prostasin (h)-PR: sc-63314-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.

PROTOCOLS

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