SANTA CRUZ BIOTECHNOLOGY, INC.

Furin siRNA (m): sc-40596

BACKGROUND

Furin (FUR, PACE, PCSK3, SPC1, Kex2p) is a calcium-dependent serine endoprotease that belongs to the subtilisin-like proprotein convertase family. The members of this family process latent precursor proteins into their biologically active products. Furin cleaves at paired basic amino acid processing sites within proparathyroid hormone, transforming growth factor β1 precursor, proalbumin, pro-β-secretase, membrane type-1 matrix metalloproteinase, β subunit of pro-nerve growth factor and von Willebrand factor. Furin can directly cleave proMMP-2 within the *trans*-Golgi network leading to an inactive form of matrix metalloproteinase-2 (MMP-2). Furin is synthesized as an inactive zymogen that may minimize the occurrence of premature enzymatic activity that would lead to alternative protein activation or degradation. The inhibitory mechanism is based on the presence of an inactivating prosegment at the NH_2 terminal of the Furin. After initial autocatalytic cleavage, the prosegment remains tightly associated until it reaches the *trans*-Golgi network where the dissociation of the prosegment and activation of Furin occurs.

REFERENCES

1. Hatsuzawa, K., et al. 1990. Structure and expression of mouse Furin, a yeast Kex2-related protease. Lack of processing of coexpressed prorenin in GH4C1 cells. J. Biol. Chem. 265: 22075-22078.
2. Nakayama, K. 1997. Furin: a mammalian subtilisin/Kex2p-like endoprotease involved in processing of a wide variety of precursor proteins. Biochem. J. 327: 625-635.
3. Rozan, L., et al. 2004. Plasticity of extended subsites facilitates divergent substrate recognition by Kex2 and Furin. J. Biol. Chem. 279: 35656-35663.
4. Podsiadlo, P., et al. 2004. Furin inhibition by compounds of copper and zinc. J. Biol. Chem. 279: 36219-36227.
5. Wickham, L., et al. 2005. β-Amyloid protein converting enzyme 1 and brain-specific type II membrane protein BRI3: binding partners processed by furin. J. Neurochem. 92: 93-102.
6. Cao, J., et al. 2005. Furin directly cleaves proMMP-2 in the *trans*-Golgi network resulting in a nonfunctioning proteinase. J. Biol. Chem. 280: 10974-10980.

CHROMOSOMAL LOCATION

Genetic locus: Furin (mouse) mapping to 7 D3.

PRODUCT

Furin siRNA (m) 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 Furin shRNA Plasmid (m): sc-40596-SH and Furin shRNA (m) Lentiviral Particles: sc-40596-V as alternate gene silencing products.

For independent verification of Furin (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-40596A, sc-40596B and sc-40596C.

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

Furin siRNA (m) is recommended for the inhibition of Furin 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

Furin (B-6): sc-133142 is recommended as a control antibody for monitoring of Furin 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 Furin gene expression knockdown using RT-PCR Primer: Furin (m)-PR: sc-40596-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.