SANTA CRUZ BIOTECHNOLOGY, INC.

PREPL siRNA (h): sc-94770

BACKGROUND

PREPL (Prolyl endopeptidase-like) is a 727 amino acid cytosolic protein that belongs to the prolyl oligopeptidase subfamily of serine peptidases. PREPL functions as a homodimer and is widely expressed, with highest levels found in heart, brain, kidney and skeletal muscle. Unlike its family members PREP and oligopeptidase B which require both amino and carboxy-terminal sequences for activity, PREPL activity appears to be dependent on only the carboxy-terminal domain. Defects in the gene encoding PREPL results in hypotonia-cystinuria syndrome, also known as 2p21 deletion syndrome, which is characterized by hypotonia at birth, growth hormone deficiency and failure to thrive. The disease is caused by homozygous deletion resulting in disruption of both the SLC3A1 and PREPL genes on chromosome 2p21. There are four isoforms of PREPL that are produced as a result of alternative splicing events.

REFERENCES

1. Szeltner, Z., et al. 2005. The PREPL A protein, a new member of the prolyl oligopeptidase family, lacking catalytic activity. Cell. Mol. Life Sci. 62: 2376-2381.
2. Parvari, R., et al. 2005. The 2p21 deletion syndrome: characterization of the transcription content. Genomics 86: 195-211.
3. Jaeken, J., et al. 2006. Deletion of PREPL, a gene encoding a putative serine oligopeptidase, in patients with hypotonia-cystinuria syndrome. Am. J. Hum. Genet. 78: 38-51.
4. Martens, K., et al. 2006. PREPL: a putative novel oligopeptidase propelled into the limelight. Biol. Chem. 387: 879-883.
5. Online Mendelian Inheritance in Man, OMIM™. 2007. Johns Hopkins University, Baltimore, MD. MIM Number: 609557. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
6. Martens, K., et al. 2008. Multi-system disorder syndromes associated with cystinuria type I. Curr. Mol. Med. 8: 544-550.
7. Chabrol, B., et al. 2008. Deletion of C2orf34, PREPL and SLC3A1 causes atypical hypotonia-cystinuria syndrome. J. Med. Genet. 45: 314-318.

CHROMOSOMAL LOCATION

Genetic locus: PREPL (human) mapping to 2p21.

PRODUCT

PREPL 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 PREPL shRNA Plasmid (h): sc-94770-SH and PREPL shRNA (h) Lentiviral Particles: sc-94770-V as alternate gene silencing products.

For independent verification of PREPL (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-94770A, sc-94770B and sc-94770C.

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

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

PREPL (E-9): sc-393321 is recommended as a control antibody for monitoring of PREPL 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 PREPL gene expression knockdown using RT-PCR Primer: PREPL (h)-PR: sc-94770-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.