SANTA CRUZ BIOTECHNOLOGY, INC.

NPEPL1 siRNA (h): sc-75953

BACKGROUND

Aminopeptidases are widely distributed in eukaryotes and prokaryotes. These enzymes catalyze the removal of amino acids from the N-termini of proteins or peptide substrates. Aminopeptidases are involved in regulating hormone levels and are essential for digestive and intracellular protein metabolism. NPEPL1, also referred to as aminopeptidase-like 1, is a 523 amino acid protein that belongs to the peptidase M17 family and is ubiquitously expressed. NPEPL1 may be involved in the processing, catabolism and degradation of intracellular proteins by catalyzing the removal of unsubstituted N-terminal amino acids from various peptides. NPEPL1 contains several zinc binding sites and and is expressed as three isoforms due to alternative splicing events.

REFERENCES

1. Taylor, A. 1993. Aminopeptidases: structure and function. FASEB J. 7: 290-298.
2. Taylor, A. 1993. Aminopeptidases: towards a mechanism of action. Trends Biochem. Sci. 18: 167-171.
3. Meinnel, T., Serero, A. and Giglione, C. 2006. Impact of the N-terminal amino acid on targeted protein degradation. Biol. Chem. 387: 839-851.
4. Herrera-Camacho, I., Rosas-Murrieta, N.H., Rojo-Domínguez, A., Millán, L., Reyes-Leyva, J., Santos-López, G. and Suárez-Rendueles, P. 2007. Biochemical characterization and structural prediction of a novel cytosolic leucyl aminopeptidase of the M17 family from *Schizosaccharomyces pombe*. FEBS J. 274: 6228-6240.
5. Chen, S.L., Marino, T., Fang, W.H., Russo, N. and Himo, F. 2008. Peptide hydrolysis by the binuclear zinc enzyme aminopeptidase from *Aeromonas proteolytica:* a density functional theory study. J. Phys. Chem. B 112: 2494-2500.
6. SWISS-PROT/TrEMBL (Q8NDH3). World Wide Web URL: http://www.expasy.ch/sprot/sprot-top.html

CHROMOSOMAL LOCATION

Genetic locus: NPEPL1 (human) mapping to 20q13.32.

PRODUCT

NPEPL1 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 NPEPL1 shRNA Plasmid (h): sc-75953-SH and NPEPL1 shRNA (h) Lentiviral Particles: sc-75953-V as alternate gene silencing products.

For independent verification of NPEPL1 (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-75953A, sc-75953B and sc-75953C.

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 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

NPEPL1 siRNA (h) is recommended for the inhibition of NPEPL1 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

NPEPL1 (E-10): sc-515463 is recommended as a control antibody for monitoring of NPEPL1 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 NPEPL1 gene expression knockdown using RT-PCR Primer: NPEPL1 (h)-PR: sc-75953-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.