SANTA CRUZ BIOTECHNOLOGY, INC.

PSMD4 siRNA (m): sc-41386

BACKGROUND

In eukaryotic cells, selective breakdown of cellular proteins is ensured by two distinct pathways. First, appropriate proteins are tagged for degradation by ubiquitination. Second, these multiubiquitinated proteins are degraded by the highly selective 26S proteasome protein-destroying machinery. At specific stages of development, embryo- and tissue-specific components of the 26S proteasome are formed, which are termed Rpn10a through Rpn10e. All members of this family can be generated by a single PSMD4 gene by developmentally regulated alternative splicing. PSMD4, originally identified as S5a (also designated antisecretory factor and multiubiquitin chain binding protein) is ubiquitously expressed and may perform proteolysis constitutively in a wide variety of cells. Rpn10D and Rpn10E may have embryo- or tissue-specific expression and may play specialized roles in early embryonic development.

REFERENCES

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2. Johansson, E., Lonnroth, I., Lange, S., Jonson, I., Jennische, E. and Lonnroth, C. 1995. Molecular cloning and expression of a pituitary gland protein modulating intestinal fluid secretion. J. Biol. Chem. 270: 20615-20620.
3. Coux, O., Tanaka, K. and Goldberg, A.L. 1996. Structure and functions of the 20S and 26S proteasomes. Annu. Rev. Biochem. 65: 801-847.
4. Voges, D., Zwickl, P. and Baumeister, W. 1999. The 26S proteasome: a molecular machine designed for controlled proteolysis. Annu. Rev. Biochem. 68: 1015-1068.
5. Kawahara, H., Kasahara, M., Nishiyama, A., Ohsumi, K., Goto, T., Kishimoto, T., Saeki, Y., Yokosawa, H., Shimbara, N., Murata, S., Chiba, T., Suzuki, K. and Tanaka, K. 2000. Developmentally regulated, alternative splicing of the Rpn10 gene generates multiple forms of 26S proteasomes. EMBO J. 19: 4144-4153.

CHROMOSOMAL LOCATION

Genetic locus: Psmd4 (mouse) mapping to 3 F2.1.

PRODUCT

PSMD4 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 PSMD4 shRNA Plasmid (m): sc-41386-SH and PSMD4 shRNA (m) Lentiviral Particles: sc-41386-V as alternate gene silencing products.

For independent verification of PSMD4 (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-41386A, sc-41386B and sc-41386C.

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

PSMD4 siRNA (m) is recommended for the inhibition of PSMD4 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

PSMD4 (F-6): sc-398033 is recommended as a control antibody for monitoring of PSMD4 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 PSMD4 gene expression knockdown using RT-PCR Primer: PSMD4 (m)-PR: sc-41386-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.