SANTA CRUZ BIOTECHNOLOGY, INC.

VPS41 siRNA (h): sc-76907

BACKGROUND

Vacuolar sorting proteins (VPSs) are required for proper trafficking of endocytic and biosynthetic proteins to the vacuole and play an important role in the budding process of cells. VPS41 (vacuolar protein sorting 41), also known as HVPS41, is an 854 amino acid protein that contains one clathrin repeat and one RING-type zinc finger. Existing as two alternatively spliced isoforms, designated short and long, VPS41 is required for proper vacuolar assembly and vacuolar traffic, playing a role in the formation and fusion of transport vesicles from the Golgi. The gene encoding VPS41 maps to human chromosome 7, which houses over 1,000 genes and comprises nearly 5% of the human genome. Defects in some of the genes localized to chromosome 7 have been linked to osteogenesis imperfecta, Williams-Beuren syndrome, Pendred syndrome, lissencephaly, citrullinemia and Shwachman-Diamond syndrome.

REFERENCES

1. Radisky, D.C., et al. 1997. Characterization of VPS41, a gene required for vacuolar trafficking and high-affinity iron transport in yeast. Proc. Natl. Acad. Sci. USA 94: 5662-5666.
2. Rehling, P., et al. 1999. Formation of AP-3 transport intermediates requires VPS41 function. Nat. Cell Biol. 1: 346-353.
3. McVey Ward, D., et al. 2001. hVPS41 is expressed in multiple isoforms and can associate with vesicles through a RING-H2 finger motif. Exp. Cell Res. 267: 126-134.
4. Online Mendelian Inheritance in Man, OMIM™. 2001. Johns Hopkins University, Baltimore, MD. MIM Number: 605485. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
5. Takeda, K., et al. 2008. The vacuolar V_1/V_0-ATPase is involved in the release of the HOPS subunit VPS41 from vacuoles, vacuole fragmentation and fusion. FEBS Lett. 582: 1558-1563.
6. Brett, C.L., et al. 2008. Efficient termination of vacuolar Rab GTPase signaling requires coordinated action by a GAP and a protein kinase. J. Cell Biol. 182: 1141-1151.

CHROMOSOMAL LOCATION

Genetic locus: VPS41 (human) mapping to 7p14.1.

PRODUCT

VPS41 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 VPS41 shRNA Plasmid (h): sc-76907-SH and VPS41 shRNA (h) Lentiviral Particles: sc-76907-V as alternate gene silencing products.

For independent verification of VPS41 (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-76907A, sc-76907B and sc-76907C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

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

VPS41 siRNA (h) is recommended for the inhibition of VPS41 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

VPS41 (D-12): sc-377118 is recommended as a control antibody for monitoring of VPS41 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 VPS41 gene expression knockdown using RT-PCR Primer: VPS41 (h)-PR: sc-76907-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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