

VPS16 siRNA (h): sc-76902

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. The VPS proteins are highly conserved in mammal, yeast and *Drosophila*. VPS16 (vacuolar protein sorting 16) is a 839 amino acid protein that localizes to the cytoplasmic side of membranes and is ubiquitously expressed. Existing as a component of the Class C VPS protein complex along with VPS11, VPS18 and VPS33, VPS16 is thought to play a role in membrane docking/fusion reactions of late endosomes/lysosomes and may also participate in vesicle-mediated protein trafficking to lysosomal compartments. Mutations in the gene encoding VPS16 may disrupt trafficking to lysosomes and lysosome-related organelles that can potentially cause multiple diseases, including Hermansky-Pudlak syndrome.

REFERENCES

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5. Kim, B.Y., Ueda, M., Nakamura, Y., Kohsaka, S. and Akazawa, C. 2004. Expression of the mammalian homologue of vacuolar protein sorting 16 (VPS16p) in the mouse and rat brain. *Neurosci. Lett.* 355: 217-220.
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CHROMOSOMAL LOCATION

Genetic locus: VPS16 (human) mapping to 20p13.

PRODUCT

VPS16 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see VPS16 shRNA Plasmid (h): sc-76902-SH and VPS16 shRNA (h) Lentiviral Particles: sc-76902-V as alternate gene silencing products.

For independent verification of VPS16 (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-76902A, sc-76902B and sc-76902C.

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 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

VPS16 siRNA (h) is recommended for the inhibition of VPS16 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 μ M in 66 μ l. 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

VPS16 (2F10): sc-293327 is recommended as a control antibody for monitoring of VPS16 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-IgG κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG κ 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 VPS16 gene expression knockdown using RT-PCR Primer: VPS16 (h)-PR: sc-76902-PR (20 μ l). 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.