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Slac2-a siRNA (h): sc-44754

BACKGROUND

Slac2-a (for synaptotagmin-like protein (Slp) homolog lacking C2 domains-a) links Rab27A on melanosomes with Myosin VA in melanocytes. Slac2-a binds Myosin VA through a C-terminal region and GTP-bound Rab27A through its synaptotagmin-like protein homology domain (SHD), which is located near the N-terminus. The transport of pigment and cytotoxic granules in melanocytes requires the stable formation of this complex, and thus mutations in the binding domains of the three protein components may cause albinism and/or severe immune disorders.

REFERENCES

1. Fukuda, M., et al. 2002. Slac2-a/Melanophilin, the missing link between Rab27 and Myosin VA: implications of a tripartite protein complex for melanosome transport. *J. Biol. Chem.* 277: 12432-12436.
2. Fukuda, M. 2002. Synaptotagmin-like protein (Slp) homology domain 1 of Slac2-a/Melanophilin is a critical determinant of GTP-dependent specific binding to Rab27A. *J. Biol. Chem.* 277: 40118-40124.
3. Fukuda, M., et al. 2002. Slac2-c (synaptotagmin-like protein homologue lacking C2 domains-c), a novel linker protein that interacts with Rab27, Myosin VA/VIIA and Actin. *J. Biol. Chem.* 277: 43096-43103.
4. Kuroda, T.S., et al. 2003. The Actin-binding domain of Slac2-a/Melanophilin is required for melanosome distribution in melanocytes. *Mol. Cell Biol.* 23: 5245-5255.
5. Fukuda, M. 2003. Distinct Rab binding specificity of Rim1, Rim2, rabphilin and Noc2. Identification of a critical determinant of Rab3A/Rab27A recognition by Rim2. *J. Biol. Chem.* 278: 15373-15380.
6. Kuroda, T.S., et al. 2004. Rab27A-binding protein Slp2-a is required for peripheral melanosome distribution and elongated cell shape in melanocytes. *Nat. Cell Biol.* 6: 1195-1203.

CHROMOSOMAL LOCATION

Genetic locus: MLPH (human) mapping to 2q37.3.

PRODUCT

Slac2-a 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 Slac2-a shRNA Plasmid (h): sc-44754-SH and Slac2-a shRNA (h) Lentiviral Particles: sc-44754-V as alternate gene silencing products.

For independent verification of Slac2-a (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-44754A, sc-44754B and sc-44754C.

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 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

Slac2-a siRNA (h) is recommended for the inhibition of Slac2-a 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

Slac2-a (G-10): sc-515894 is recommended as a control antibody for monitoring of Slac2-a 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 Slac2-a gene expression knockdown using RT-PCR Primer: Slac2-a (h)-PR: sc-44754-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.