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# Slac2-a shRNA (m) Lentiviral Particles: sc-44755-V

## 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. Kuroda, T.S., et al. 2002. The Slp homology domain of synaptotagmin-like proteins 1-4 and Slac2 functions as a novel Rab27A binding domain. *J. Biol. Chem.* 277: 9212-9218.
2. 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.
3. 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.
4. 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.
5. 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.
6. 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.
7. 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 (mouse) mapping to 1 D.

## PRODUCT

Slac2-a shRNA (m) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200  $\mu$ l frozen stock containing  $1.0 \times 10^6$  infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see Slac2-a siRNA (m): sc-44755 and Slac2-a shRNA Plasmid (m): sc-44755-SH as alternate gene silencing products.

## STORAGE

Store lentiviral particles at  $-80^\circ$  C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at  $4^\circ$  C for up to one week. Avoid repeated freeze thaw cycles.

## APPLICATIONS

Slac2-a shRNA (m) Lentiviral Particles is recommended for the inhibition of Slac2-a expression in mouse cells.

## SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200  $\mu$ l frozen viral stock containing  $1.0 \times 10^6$  infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

## GENE EXPRESSION MONITORING

Slac2-a (C-3): sc-365735 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Slac2-a gene expression knockdown using RT-PCR Primer: Slac2-a (m)-PR: sc-44755-PR (20  $\mu$ l). Annealing temperature for the primers should be  $55-60^\circ$  C and the extension temperature should be  $68-72^\circ$  C.

## BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

## RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

## PROTOCOLS

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