

Rho T1 siRNA (m): sc-152856

BACKGROUND

The Rho subfamily of Ras-related GTPases controls multiple aspects of cell function, including cytoskeletal rearrangement, nuclear signaling and cell growth. Rho T1 (ras homolog gene family, member T1), also known as ARHT1 or MIRO-1 (mitochondrial Rho GTPase 1), is an evolutionarily conserved member of the mitochondrial Rho GTPase family of proteins. Localizing to the mitochondrion, Rho T1 is widely expressed with predominant expression in skeletal muscle and heart. Rho T1 is a single-pass type IV membrane protein with two EF-hand domains and two GTPase domains (one at the N-terminus and one at the C-terminus). It is believed to play a role in the regulation of mitochondrial homeostasis and specifically binds to the kinesin-interacting proteins GRIF-1 and OIP106. Mutations in the gene encoding Rho T1 result in aggregation of the mitochondria. This suggests a potential role for Rho T1 in mitochondrial trafficking.

REFERENCES

1. Fransson, A., et al. 2003. Atypical Rho GTPases have roles in mitochondrial homeostasis and apoptosis. *J. Biol. Chem.* 278: 6495-6502.
2. Aspenström, P., et al. 2004. Rho GTPases have diverse effects on the organization of the actin filament system. *Biochem. J.* 377: 327-337.
3. Shan, Y., et al. 2004. Cloning and characterization of the mouse Arht2 gene which encodes a putative atypical GTPase. *Cytogenet. Genome Res.* 106: 91-97.
4. Guo, X., et al. 2005. The GTPase dMiro is required for axonal transport of mitochondria to *Drosophila* synapses. *Neuron* 47: 379-393.
5. Fransson, S., et al. 2006. The atypical Rho GTPases Miro-1 and Miro-2 have essential roles in mitochondrial trafficking. *Biochem. Biophys. Res. Commun.* 344: 500-510.
6. Rice, S.E., et al. 2006. Paradigm lost: milton connects kinesin heavy chain to miro on mitochondria. *J. Cell Biol.* 173: 459-461.
7. Glater, E.E., et al. 2006. Axonal transport of mitochondria requires milton to recruit kinesin heavy chain and is light chain independent. *J. Cell Biol.* 173: 545-557.
8. Frederick, R.L., et al. 2007. Moving mitochondria: establishing distribution of an essential organelle. *Traffic* 8: 1668-1675.

CHROMOSOMAL LOCATION

Genetic locus: Rhot1 (mouse) mapping to 11 B5.

PRODUCT

Rho T1 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Rho T1 shRNA Plasmid (m): sc-152856-SH and Rho T1 shRNA (m) Lentiviral Particles: sc-152856-V as alternate gene silencing products.

For independent verification of Rho T1 (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-152856A, sc-152856B and sc-152856C.

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

Rho T1 siRNA (m) is recommended for the inhibition of Rho T1 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 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

Rho T1 (A-8): sc-398520 is recommended as a control antibody for monitoring of Rho T1 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 Rho T1 gene expression knockdown using RT-PCR Primer: Rho T1 (m)-PR: sc-152856-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.