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SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

mail@szabo-scandic.com

www.szabo-scandic.com

[linkedin.com/company/szaboscandic](https://www.linkedin.com/company/szaboscandic) 

Tom1 siRNA (m): sc-154550

BACKGROUND

Tom1 (target of Myb 1) is a 492 amino acid cytoplasmic protein that belongs to the Tom1 family. Widely expressed with highest expression in heart, liver, placenta and skeletal muscle, Tom1 is thought to be involved in intracellular trafficking and may be associated with the translocation of ubiquitinated proteins to early endosomes for degradation. Tom1 contains one GAT domain and one VHS domain through which it interacts and complexes with proteins such as Tollip and endofin (also known as SARA). These interactions recruit Tom1 to the endosome, allowing it to participate in the endosomal trafficking of ubiquitin-tagged proteins. Human Tom1 shares 76% similarity with its chicken counterpart and 89% similarity with its mouse counterpart, suggesting a conserved function between species.

REFERENCES

1. Seroussi, E., et al. 1999. TOM1 genes map to human chromosome 22q13.1 and mouse chromosome 8C1 and encode proteins similar to the endosomal proteins HGS and STAM. *Genomics* 57: 380-388.
2. Misra, S., Beach, B.M. and Hurley, J.H. 2000. Structure of the VHS domain of human Tom1 (target of Myb 1): insights into interactions with proteins and membranes. *Biochemistry* 39: 11282-11290.
3. Yamakami, M., et al. 2003. Tom1, a VHS domain-containing protein, interacts with tollip, ubiquitin, and clathrin. *J. Biol. Chem.* 278: 52865-52872.
4. Seet, L.F., et al. 2004. Endofin recruits TOM1 to endosomes. *J. Biol. Chem.* 279: 4670-4679.
5. Katoh, Y., et al. 2004. Tollip and Tom1 form a complex and recruit ubiquitin-conjugated proteins onto early endosomes. *J. Biol. Chem.* 279: 24435-24443.
6. Akutsu, M., et al. 2005. Structural basis for recognition of ubiquitinated cargo by Tom1-GAT domain. *FEBS Lett.* 579: 5385-5391.
7. Seet, L.F. and Hong, W. 2005. Endofin recruits clathrin to early endosomes via TOM1. *J. Cell Sci.* 118: 575-587.
8. Katoh, Y., et al. 2006. Recruitment of clathrin onto endosomes by the Tom1-Tollip complex. *Biochem. Biophys. Res. Commun.* 341: 143-149.

CHROMOSOMAL LOCATION

Genetic locus: Tom1 (mouse) mapping to 8 C1.

PRODUCT

Tom1 siRNA (m) is a target-specific 19-25 nt siRNA 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 Tom1 shRNA Plasmid (m): sc-154550-SH and Tom1 shRNA (m) Lentiviral Particles: sc-154550-V as alternate gene silencing products.

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

Tom1 siRNA (m) is recommended for the inhibition of Tom1 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

Tom1 (H-5): sc-514430 is recommended as a control antibody for monitoring of Tom1 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 Tom1 gene expression knockdown using RT-PCR Primer: Tom1 (m)-PR: sc-154550-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.