

Tim22 siRNA (m): sc-154273

BACKGROUND

Tim22, also known as TIMM22 (mitochondrial import inner membrane translocase subunit Tim22) or TEX4 (testis-expressed sequence 4), is a 194 amino acid multi-pass membrane protein that belongs to the Tim17/Tim22/Tim23 family. Tim22 is an essential core component of the Tim22 complex, a complex that mediates the import and insertion of multi-pass transmembrane proteins into the mitochondrial inner membrane. In the complex, Tim22 constitutes the voltage-activated and signal-gated channel, and forms a twin-pore translocase that uses the membrane potential as external driving force in two voltage-dependent steps. As a member of the Tim22 complex, Tim22 also associates with peripheral protein Tim9B and the 70 kDa heterohexamers, made up of Tim9A and Tim10. The gene that encodes Tim22 contains more than 5,000 bases and maps to human chromosome 17p13.

REFERENCES

1. Bauer, M.F., Rothbauer, U., Mühlenbein, N., Smith, R.J., Gerbitz, K., Neupert, W., Brunner, M. and Hofmann, S. 1999. The mitochondrial TIM22 preprotein translocase is highly conserved throughout the eukaryotic kingdom. *FEBS Lett.* 464: 41-47.
2. Koehler, C.M., Murphy, M.P., Bally, N.A., Leuenberger, D., Oppliger, W., Dolfini, L., Junne, T., Schatz, G. and Or, E. 2000. Tim18p, a new subunit of the TIM22 complex that mediates insertion of imported proteins into the yeast mitochondrial inner membrane. *Mol. Cell. Biol.* 20: 1187-1193.
3. Bauer, M.F. and Neupert, W. 2001. Import of proteins into mitochondria: a novel pathomechanism for progressive neurodegeneration. *J. Inher. Metab. Dis.* 24: 166-180.
4. Kovermann, P., Truscott, K.N., Guiard, B., Rehling, P., Sepuri, N.B., Müller, H., Jensen, R.E., Wagner, R. and Pfanner, N. 2002. Tim22, the essential core of the mitochondrial protein insertion complex, forms a voltage-activated and signal-gated channel. *Mol. Cell* 9: 363-373.
5. Online Mendelian Inheritance in Man, OMIM[™]. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 607251. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
6. Rehling, P., Model, K., Brandner, K., Kovermann, P., Sickmann, A., Meyer, H.E., Kühlbrandt, W., Wagner, R., Truscott, K.N. and Pfanner, N. 2003. Protein insertion into the mitochondrial inner membrane by a twin-pore translocase. *Science* 299: 1747-1751.
7. Mühlenbein, N., Hofmann, S., Rothbauer, U. and Bauer, M.F. 2004. Organization and function of the small Tim complexes acting along the import pathway of metabolite carriers into mammalian mitochondria. *J. Biol. Chem.* 279: 13540-13546.
8. Curran, S.P., Leuenberger, D., Leverich, E.P., Hwang, D.K., Beverly, K.N. and Koehler, C.M. 2004. The role of Hot13p and redox chemistry in the mitochondrial TIM22 import pathway. *J. Biol. Chem.* 279: 43744-43751.
9. Peixoto, P.M., Graña, F., Roy, T.J., Dunn, C.D., Flores, M., Jensen, R.E. and Campo, M.L. 2007. Awakening TIM22, a dynamic ligand-gated channel for protein insertion in the mitochondrial inner membrane. *J. Biol. Chem.* 282: 18694-18701.

CHROMOSOMAL LOCATION

Genetic locus: Timm22 (mouse) mapping to 11 B5.

PRODUCT

Tim22 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 Tim22 shRNA Plasmid (m): sc-154273-SH and Tim22 shRNA (m) Lentiviral Particles: sc-154273-V as alternate gene silencing products.

For independent verification of Tim22 (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-154273A, sc-154273B and sc-154273C.

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

Tim22 siRNA (m) is recommended for the inhibition of Tim22 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Tim22 gene expression knockdown using RT-PCR Primer: Tim22 (m)-PR: sc-154273-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.