

TAP1 siRNA (h): sc-42981

BACKGROUND

The transporter associated with antigen processing (TAP) is a member of the ATP binding cassette (ABC) family of transmembrane transporters and is an essential component of the major histocompatibility complex (MHC) class I antigen-presenting pathway. TAP consists of two structurally related subunits, TAP1 and TAP2, that associate into stable dimers and together they form a translocation pore for peptides in the endoplasmic reticulum (ER) membranes. The functional TAP transporter facilitates the translocation of peptides from the cytosol into the ER lumen for presentation to MHC class I molecules. Structurally, TAP1 and TAP2 contain an N-terminal transmembrane (TM) region, which together forms the TM pore, and a cytoplasmic peptide-binding pocket. In addition, the TAP transporter also contains several C-terminal nucleotide-binding domains (NBD), which bind and hydrolyze ATP and, in turn, induce structural changes at the membrane to allow the passage of substrates into the ER.

REFERENCES

1. Androlewicz, M.J., et al. 1993. Evidence that transporters associated with antigen processing translocate a major histocompatibility complex class I-binding peptide into the endoplasmic reticulum in an ATP-dependent manner. *Proc. Natl. Acad. Sci. USA* 90: 9130-9134.
2. Androlewicz, M.J., et al. 1994. Characteristics of peptide and major histocompatibility complex class I/ β -2-Microglobulin binding to the transporters associated with antigen processing (TAP1 and TAP2). *Proc. Natl. Acad. Sci. USA* 91: 12716-12720.
3. Nijenhuis, M., et al. 1996. Multiple regions of the transporter associated with antigen processing (TAP) contribute to its peptide binding site. *J. Immunol.* 157: 5467-5477.
4. Powis, S.J. 1997. Major histocompatibility complex class I molecules interact with both subunits of the transporter associated with antigen processing, TAP1 and TAP2. *Eur. J. Immunol.* 27: 2744-2747.
5. Knittler, M.R., et al. 1999. Nucleotide binding by TAP mediates association with peptide and release of assembled MHC class I molecules. *Curr. Biol.* 9: 999-1008.

CHROMOSOMAL LOCATION

Genetic locus: TAP1 (human) mapping to 6p21.32.

PRODUCT

TAP1 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 TAP1 shRNA Plasmid (h): sc-42981-SH and TAP1 shRNA (h) Lentiviral Particles: sc-42981-V as alternate gene silencing products.

For independent verification of TAP1 (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-42981A, sc-42981B and sc-42981C.

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

TAP1 siRNA (h) is recommended for the inhibition of TAP1 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

TAP1 (D-11): sc-518133 is recommended as a control antibody for monitoring of TAP1 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 TAP1 gene expression knockdown using RT-PCR Primer: TAP1 (h)-PR: sc-42981-PR (20 μ l, 579 bp). 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.