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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

SANTA CRUZ BIOTECHNOLOGY, INC.

MECL-1 siRNA (m): sc-42907

BACKGROUND

The 20S proteasome is a protease complex that is responsible for cytosolic protein degradation and generation of peptide ligands for major histocompatabilty complex (MHC) class I molecules, either in their final form or in the form of amino-terminally extended precursors. Upon IFN-γ stimulation of cells, three constitutively expressed subunits of the 20S proteasome are replaced by inducible subunits LMP2 (low-molecular mass polypeptide 2), LMP7, and MECL-1 (multicatalytic endopeptidase complex-like-1, LMP10). LMP2, LMP7, and MECL-1 subunits form immunoproteasomes, which are associated with more efficient class I antigen processing and presentation. Independent assortment of LMP-2, LMP-7, and MECL-1 into different proteasome complexes can lead to 36 unique proteasome subsets, which may mediate differences in the cleavage specificities/cleavage motifs of proteins subject to constitutive- and immuno-proteasomes.

REFERENCES

1. Nandi, D., et al. 1996. Identification of MECL-1 (LMP-10) as the third IFN-γ-inducible proteasome subunit. J. Immunol. 156: 2361-2364.
2. Frisan, T., et al. 1998. Phenotype-dependent differences in proteasome subunit composition and cleavage specificity in B cell lines. J. Immunol. 160: 3281-3289.
3. Sijts, A.J., et al. 2000. Efficient generation of a hepatitis B virus cytotoxic T lymphocyte epitope requires the structural features of immunoproteasomes. J. Exp. Med. 191: 503-514.
4. Schwarz, K., et al. 2000. Overexpression of the proteasome subunits LMP2, LMP7, and MECL-1, but not PA28 α/β, enhances the presentation of an immunodominant lymphocytic choriomeningitis virus T cell epitope. J. Immunol. 165: 768-778.
5. Toes, R.E., et al. 2001. Discrete cleavage motifs of constitutive and immunoproteasomes revealed by quantitative analysis of cleavage products. J. Exp. Med. 194: 1-12.

CHROMOSOMAL LOCATION

Genetic locus: Psmb10 (mouse) mapping to 8 D3.

PRODUCT

MECL-1 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see MECL-1 shRNA Plasmid (m): sc-42907-SH and MECL-1 shRNA (m) Lentiviral Particles: sc-42907-V as alternate gene silencing products.

For independent verification of MECL-1 (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-42907A, sc-42907B and sc-42907C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

MECL-1 siRNA (m) is recommended for the inhibition of MECL-1 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 µM in 66 µl. 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 MECL-1 gene expression knockdown using RT-PCR Primer: MECL-1 (m)-PR: sc-42907-PR (20 µl). 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.