

MRP-L28 siRNA (m): sc-62644

BACKGROUND

MRP-L28 (39S ribosomal protein L28, melanoma-associated antigen recognized by T lymphocytes) is a mitochondrial protein that belongs to the ribosomal protein L28P family. MRP-L28 potentially represents an important therapeutic reagent for HLA-A24 (A24) patients as this antigen is recognized by tumor-infiltrating lymphocyte (TIL) 1290, which targets the A24 serotype. HLA-A24 (A24) is an HLA-A serotype that identifies the more common HLA-A24 gene products. A24 is a split antigen that is also recognized by the A9 broad antigen type and the similar A23 types. A24 is common in Austronesia and has one of the highest A allele frequencies for a number of peoples, including Papua New Guineans, indigenous Taiwanese (eastern tribals), Yupik and Greenland Eskimos. MRP-L28 is found in a variety of normal tissues including spleen, testis, thymus, liver, kidney, brain, adrenal, lung and retinal tissue.

REFERENCES

1. Robbins, P.F., el-Gamil, M., Li, Y.F., Topalian, S.L., Rivoltini, L., Sakaguchi, K., Appella, E., Kawakami, Y. and Rosenberg, S.A. 1995. Cloning of a new gene encoding an antigen recognized by melanoma-specific HLA-A24-restricted tumor-infiltrating lymphocytes. *J. Immunol.* 154: 5944-5950.
2. Robbins, P.F., El-Gamil, M., Li, Y.F., Kawakami, Y., Loftus, D., Appella, E. and Rosenberg, S.A. 1996. A mutated β -catenin gene encodes a melanoma-specific antigen recognized by tumor infiltrating lymphocytes. *J. Exp. Med.* 183: 1185-1192.
3. Kawakami, Y., Dang, N., Wang, X., Tupesis, J., Robbins, P.F., Wang, R.F., Wunderlich, J.R., Yannelli, J.R. and Rosenberg, S.A. 2000. Recognition of shared melanoma antigens in association with major HLA-A alleles by tumor infiltrating T lymphocytes from 123 patients with melanoma. *J. Immunother.* 23: 17-27.
4. Walsh, D.W., Wolfe, K.H. and Butler, G. 2002. Genomic differences between *Candida glabrata* and *Saccharomyces cerevisiae* around the MRPL28 and GCN3 loci. *Yeast* 19: 991-994.
5. Wong, S., Butler, G. and Wolfe, K.H. 2002. Gene order evolution and paleopolyploidy in hemiascomycete yeasts. *Proc. Natl. Acad. Sci. USA* 99: 9272-9277.

CHROMOSOMAL LOCATION

Genetic locus: Mrpl28 (mouse) mapping to 17 A3.3.

PRODUCT

MRP-L28 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 MRP-L28 shRNA Plasmid (m): sc-62644-SH and MRP-L28 shRNA (m) Lentiviral Particles: sc-62644-V as alternate gene silencing products.

For independent verification of MRP-L28 (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-62644A, sc-62644B and sc-62644C.

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

MRP-L28 siRNA (m) is recommended for the inhibition of MRP-L28 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

MRP-L28 (A-10): sc-390714 is recommended as a control antibody for monitoring of MRP-L28 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 MRP-L28 gene expression knockdown using RT-PCR Primer: MRP-L28 (m)-PR: sc-62644-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.