SANTA CRUZ BIOTECHNOLOGY, INC.

NRAMP 2 siRNA (m): sc-40777

BACKGROUND

Natural resistance associated macrophage proteins (NRAMPs) belong to a super-family of highly conserved integral membrane proteins. NRAMP 1 is an intracellular macrophage protein located at the phagosomal membrane where it functions as a divalent cation transporter for Fe^{2+}, Zn^{2+} and Mn^{2+}. NRAMP 1 is a pH-dependent antiporter that transports metal ions either into or out of the phagosome against a proton gradient. The gene encoding human NRAMP 1 maps to chromosome 2q35. In humans, polymorphisms in the NRAMP 1 gene are linked to susceptibility to *M. tuberculosis* and leprosy. NRAMP 2 is another divalent cation transporter ubiquitously expressed as two splice variants, which are distinguished by the presence (isoform 1) or absence (isoform 2) of an iron response element. In the duodenum of the small intestine, dietary iron regulates NRAMP 2 expression at the brush border. The gene encoding human NRAMP 2 maps to chromosome 12q13. Mutations in the gene for NRAMP 2 in mice and rats result in severe anemia.

REFERENCES

1. Cellier, M., et al. 1994. Human natural resistance-associated macrophage protein: cDNA cloning, chromosomal mapping, genomic organization, and tissue-specific expression. J. Exp. Med. 180: 1741-1752.
2. Vidal, S., et al. 1995. Cloning and characterization of a second human NRAMP gene on chromosome 12q13. Mamm. Genome 6: 224-230.
3. Abel, L., et al. 1998. Susceptiblity to leprosy is linked to the human NRAMP1 gene. J. Infect. Dis. 177: 133-145.
4. Lee, P.L., et al. 1998. The human Nramp2 gene: characterization of the gene structure, alternative splicing, promoter region and polymorphisms. Blood Cells Mol. Dis. 24: 199-215.
5. Bellamy, R., et al. 1998. Variations in the NRAMP 1 gene and susceptibility to tuberculosis in West Africans. N. Engl. J. Med. 338: 640-644.
6. Su, M.A., et al. 1998. The G185R mutation disrupts function of the iron transporter NRAMP 2. Blood 92: 2157-2163.
7. Canonne-Hergaux, F., et al. 1999. Cellular and subcellular localization of the NRAMP 2 iron transporter in the intestinal brush border and regulation by dietary iron. Blood 93: 4406-4417.

CHROMOSOMAL LOCATION

Genetic locus: Slc11a2 (mouse) mapping to 15 F1.

PRODUCT

NRAMP 2 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 NRAMP 2 shRNA Plasmid (m): sc-40777-SH and NRAMP 2 shRNA (m) Lentiviral Particles: sc-40777-V as alternate gene silencing products.

For independent verification of NRAMP 2 (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-40777A, sc-40777B and sc-40777C.

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

NRAMP 2 siRNA (m) is recommended for the inhibition of NRAMP 2 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.

GENE EXPRESSION MONITORING

NRAMP 2 (G-5): sc-166884 is recommended as a control antibody for monitoring of NRAMP 2 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 NRAMP 2 gene expression knockdown using RT-PCR Primer: NRAMP 2 (m)-PR: sc-40777-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.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.