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IMPDH siRNA (h): sc-45679

BACKGROUND

A member of the GMPR family, inosine-5'-monophosphate dehydrogenase 1 (IMPDH1) functions in the regulation of cell growth by catalyzing the rate-limiting step in the *de novo* synthesis of guanine nucleotides. IMPDH1 is a ubiquitously expressed homotetramer that plays an important role in cyclic nucleoside metabolism within photoreceptors. Expression of IMPDH1 is the main type found in normal leukocytes, while IMPDH2 predominates in tumors. Mutations in IMPDH1 are associated with the autosomal dominant retinitis pigmentosa type 10 (RP10), as well as the development of malignant tumors. Analysis of mutant IMPDH1 suggests that protein misfolding and aggregation leads to the severe phenotype rather than reduced IMPDH1 activity. Therefore, IMPDH1 may be a potential therapeutic target based upon a strategy combining simultaneous suppression of IMPDH1 transcripts with supplementation of GTP within retinal tissues.

REFERENCES

- Gorskii, B.V., et al. 1977. Effect of immune lymphocytes on the postvaccinal cytoserological reaction in foot-and-mouth disease. *Veterinariia* 5: 43-44.
- Bowne, S.J., et al. 2002. Mutations in the inosine monophosphate dehydrogenase 1 gene (IMPDH1) cause the RP10 form of autosomal dominant retinitis pigmentosa. *Hum. Mol. Genet.* 11: 559-568.
- Pankiewicz, K.W., et al. 2004. Cofactor mimics as selective inhibitors of NAD-dependent inosine monophosphate dehydrogenase (IMPDH)-the major therapeutic target. *Curr. Med. Chem.* 11: 887-900.
- Wright, D.G., et al. 2004. Effects of the IMP-dehydrogenase inhibitor, Tiazofurin, in Bcr-Abl positive acute myelogenous leukemia. Part II. *in vitro* studies. *Leuk. Res.* 28: 1137-1143.
- Aherne, A. et al. 2004. On the molecular pathology of neurodegeneration in IMPDH1-based retinitis pigmentosa. *Hum. Mol. Genet.* 13: 641-650.
- SWISS-PROT/TrEMBL (P20839). World Wide Web URL: <http://www.expasy.ch/sprot/sprot-top.html>

CHROMOSOMAL LOCATION

Genetic locus: IMPDH1 (human) mapping to 7q32.1.

PRODUCT

IMPDH 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 IMPDH shRNA Plasmid (h): sc-45679-SH and IMPDH shRNA (h) Lentiviral Particles: sc-45679-V as alternate gene silencing products.

For independent verification of IMPDH (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-45679A, sc-45679B and sc-45679C.

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 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

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

IMPDH (F-6): sc-166551 is recommended as a control antibody for monitoring of IMPDH 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 IMPDH gene expression knockdown using RT-PCR Primer: IMPDH (h)-PR: sc-45679-PR (20 μ l, 540 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.