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UMPS siRNA (m): sc-154917

BACKGROUND

Uridine 5'-monophosphate synthase (UMPS) catalyzes the last two steps of the pyrimidine biosynthetic pathway. Unlike prokaryotes, UMPS in eukaryotes combines the orotate phosphoribosyltransferase and the orotidine-5'-monophosphate (OMP) decarboxylase activities into a single protein. The union of these two enzymes is thought to stabilize the catalytic centers due to the low molar concentration of the protein in mammalian cells. Loss of either enzymatic activity results in hereditary orotic aciduria, a rare autosomal recessive disorder characterized by retarded growth, anemia, and excessive urinary excretion of orotic acid. Two isoforms of UMPS exist as a result of alternative splicing events.

REFERENCES

1. Yablonski, M.J., et al. 1996. Intrinsic activity and stability of bifunctional human UMP synthase and its two separate catalytic domains, orotate phosphoribosyltransferase and orotidine-5'-phosphate decarboxylase. *J. Biol. Chem.* 271: 10704-10708.
2. Suchi, M., et al. 1997. Molecular cloning of the human UMP synthase gene and characterization of point mutations in two hereditary orotic aciduria families. *Am. J. Hum. Genet.* 60: 525-539.
3. Reisner, M., et al. 2004. The cyanobacterial toxin cylindrospermopsin inhibits pyrimidine nucleotide synthesis and alters cholesterol distribution in mice. *Toxicol. Sci.* 82: 620-627.
4. Wittmann, J.G. and Rudolph, M.G. 2007. Pseudo-merohedral twinning in monoclinic crystals of human orotidine-5'-monophosphate decarboxylase. *Acta Crystallogr. D Biol. Crystallogr.* 63: 744-749.
5. Brosnan, M.E. and Brosnan, J.T. 2007. Orotic acid excretion and arginine metabolism. *J. Nutr.* 137: 1656S-1661S.
6. Wittmann, J.G., et al. 2008. Structures of the human orotidine-5'-monophosphate decarboxylase support a covalent mechanism and provide a framework for drug design. *Structure* 16: 82-92.

CHROMOSOMAL LOCATION

Genetic locus: *Umps* (mouse) mapping to 16 B3.

PRODUCT

UMPS 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 UMPS shRNA Plasmid (m): sc-154917-SH and UMPS shRNA (m) Lentiviral Particles: sc-154917-V as alternate gene silencing products.

For independent verification of UMPS (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-154917A, sc-154917B and sc-154917C.

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

UMPS siRNA (m) is recommended for the inhibition of UMPS 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

UMPS (A-9): sc-398086 is recommended as a control antibody for monitoring of UMPS 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 UMPS gene expression knockdown using RT-PCR Primer: UMPS (m)-PR: sc-154917-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.