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DPH2 siRNA (m): sc-143158

BACKGROUND

DPH2 (diphthamide biosynthesis protein 2), also known as DPH2L2, is a 489 amino acid protein that shows strong expression in skeletal muscle; moderate expression in heart, small intestine, liver, pancreas, testis and colon; and lesser expression in brain, placenta, kidney, spleen, thymus, prostate, ovary and lymphocytes. DPH2 interacts with DPH1 and, functioning together as a dimer or multimer, DPH1 and DPH2 may participate in diphthamide biosynthesis. Diphthamide is a posttranslationally modified histidine residue which occurs in EF-2 (elongation factor 2) and targets diphtheria toxin ADP-ribosylation. The loss of DPH2 in *Saccharomyces cerevisiae* is believed to suppress zymocinicity. Two transcript variants encoding different isoforms have been found for this gene.

REFERENCES

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2. Phillips, N.J., Zeigler, M.R. and Deaven, L.L. 1996. A cDNA from the ovarian cancer critical region of deletion on chromosome 17p13.3. *Cancer Lett.* 102: 85-90.
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4. Fichtner, L., Jablonowski, D., Schierhorn, A., Kitamoto, H.K., Stark, M.J. and Schaffrath, R. 2003. Elongator's toxin-target (TOT) function is nuclear localization sequence dependent and suppressed by post-translational modification. *Mol. Microbiol.* 49: 1297-1307.
5. Liu, S., Milne, G.T., Kuremsky, J.G., Fink, G.R. and Leppla, S.H. 2004. Identification of the proteins required for biosynthesis of diphthamide, the target of bacterial ADP-ribosylating toxins on translation elongation factor 2. *Mol. Cell. Biol.* 24: 9487-9497.
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CHROMOSOMAL LOCATION

Genetic locus: Dph2 (mouse) mapping to 4 D2.1.

PRODUCT

DPH2 siRNA (m) is a target-specific 19-25 nt siRNA 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 DPH2 shRNA Plasmid (m): sc-143158-SH and DPH2 shRNA (m) Lentiviral Particles: sc-143158-V as alternate gene silencing products.

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

DPH2 siRNA (m) is recommended for the inhibition of DPH2 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

DPH2 (6E7): sc-101200 is recommended as a control antibody for monitoring of DPH2 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 DPH2 gene expression knockdown using RT-PCR Primer: DPH2 (m)-PR: sc-143158-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.