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PLEKHM3 siRNA (m): sc-152322

BACKGROUND

PLEKHM3 (pleckstrin homology domain containing, family M, member 3), also known as DAPR or PLEKHM1L (pleckstrin homology domain containing, family M, member 1-like), is a 761 amino acid phosphoprotein that contains two pleckstrin homology (PH) domains and one phorbol-ester/DAG-type zinc finger. Conserved in chimpanzee, canine, bovine, mouse, rat, chicken and zebrafish, PLEKHM3 exists as three alternatively spliced isoforms that participate in metal ion binding. The gene that encodes PLEKHM3 maps to human chromosome 2q33.3. As the second largest human chromosome, chromosome 2 makes up approximately 8% of the human genome and contains 237 million bases encoding over 1,400 genes. Chromosome 2 contains a probable vestigial second centromere, as well as vestigial telomeres, which gives credence to the hypothesis that human chromosome 2 formed as a result of an ancient fusion of two ancestral chromosomes, which are still present in modern day apes.

REFERENCES

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2. Avarello, R., et al. 1992. Evidence for an ancestral alphoid domain on the long arm of human chromosome 2. *Hum. Genet.* 89: 247-249.
3. Hillier, L.W., et al. 2005. Generation and annotation of the DNA sequences of human chromosomes 2 and 4. *Nature* 434: 724-731.
4. Akiyama, M., et al. 2007. Compound heterozygous ABCA12 mutations including a novel nonsense mutation underlie harlequin ichthyosis. *Dermatology* 215: 155-159.
5. Marshall, J.D., et al. 2007. Spectrum of ALMS1 variants and evaluation of genotype-phenotype correlations in Alström syndrome. *Hum. Mutat.* 28: 1114-1123.
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CHROMOSOMAL LOCATION

Genetic locus: *Plekhh3* (mouse) mapping to 1 C2.

PRODUCT

PLEKHM3 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 PLEKHM3 shRNA Plasmid (m): sc-152322-SH and PLEKHM3 shRNA (m) Lentiviral Particles: sc-152322-V as alternate gene silencing products.

For independent verification of PLEKHM3 (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-152322A, sc-152322B and sc-152322C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

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

PLEKHM3 siRNA (m) is recommended for the inhibition of PLEKHM3 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

PLEKHM3 (A-2): sc-393066 is recommended as a control antibody for monitoring of PLEKHM3 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 PLEKHM3 gene expression knockdown using RT-PCR Primer: PLEKHM3 (m)-PR: sc-152322-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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