SANTA CRUZ BIOTECHNOLOGY, INC.

FA2H siRNA (h): sc-93418

BACKGROUND

Sphingolipids are a class of lipids derived from sphinosine, an aliphatic amino alcohol, and play an important role in signaling and cell recognition. Ceramide is the fundamental structural unit that is common to all sphingolipids and it consists of a fatty acid chain attached to sphinosine. FA2H (fatty acid 2-hydroxylase) is a 372 amino acid multi-pass membrane protein that catalyzes the hydroxylation of ceramides at the 2-position of the N-acyl chain. Sphingolipids containing a 2-hydroxy fatty acid are common in the nervous system and epidermal tissue. Localized to the endoplasmic reticulum and microsomes, FA2H is expressed in epidermal keratinocytes, colon and brain. Defects in the gene encoding FA2H are the cause of leukodystrophy dysmyelinating with spastic paraparesis with or without dystonia.

REFERENCES

1. Loftus, B.J., et al. 1999. Genome duplications and other features in 12 Mb of DNA sequence from human chromosome 16p and 16q. Genomics 60: 295-308.
2. Uchida, Y., et al. 2007. Fatty acid 2-hydroxylase, encoded by FA2H, accounts for differentiation-associated increase in 2-OH ceramides during keratinocyte differentiation. J. Biol. Chem. 282: 13211-13219.
3. Edvardson, S., et al. 2008. Mutations in the fatty acid 2-hydroxylase gene are associated with leukodystrophy with spastic paraparesis and dystonia. Am. J. Hum. Genet. 83: 643-648.
4. Alderson, N.L., et al. 2009. Fatty acid 2-hydroxylase regulates cAMP-induced cell cycle exit in D6P2T schwannoma cells. J. Lipid Res. 50: 1203-1208.
5. Hama, H. 2010. Fatty acid 2-hydroxylation in mammalian sphingolipid biology. Biochim. Biophys. Acta 1801: 405-414.
6. Dick, K.J., et al. 2010. Mutation of FA2H underlies a complicated form of hereditary spastic paraplegia (SPG35). Hum. Mutat. 31: E1251-E1260.
7. Online Mendelian Inheritance in Man, OMIM™. 2010. Johns Hopkins University, Baltimore, MD. MIM Number: 611026. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/

CHROMOSOMAL LOCATION

Genetic locus: FA2H (human) mapping to 16q23.1.

PRODUCT

FA2H 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see FA2H shRNA Plasmid (h): sc-93418-SH and FA2H shRNA (h) Lentiviral Particles: sc-93418-V as alternate gene silencing products.

For independent verification of FA2H (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-93418A, sc-93418B and sc-93418C.

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 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

FA2H siRNA (h) is recommended for the inhibition of FA2H 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 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor FA2H gene expression knockdown using RT-PCR Primer: FA2H (h)-PR: sc-93418-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Li, H., et al. 2020. MicroRNA-1297 downregulation inhibits breast cancer cell epithelial-mesenchymal transition and proliferation in a FA2H-dependent manner. Oncol. Lett. 20: 277.
2. Hirao-Suzuki, M., et al. 2020. Fatty acid 2-hydroxylase (FA2H) as a stimulatory molecule responsible for breast cancer cell migration. Biochem. Biophys. Res. Commun. 531: 215-222.
3. Hong, B., et al. 2020. miR-300/FA2H affects gastric cancer cell proliferation and apoptosis. Open Med. 15: 882-889.
4. Sakai, G., et al. 2022. Perfluorooctanoic acid (PFOA) as a stimulator of estrogen receptor-negative breast cancer MDA-MB-231 cell aggressiveness: Evidence for involvement of fatty acid 2-hydroxylase (FA2H) in the stimulated cell migration. J. Toxicol. Sci. 47: 159-168.

PROTOCOLS

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