

FAT4 siRNA (h): sc-88877

BACKGROUND

The cadherins represent a family of Ca^{2+} -dependent adhesion molecules that function to mediate cell to cell binding that is critical for the maintenance of structure and morphogenesis. Cadherins contain a large extracellular domain at the N-terminus, which is characterized by a series of five homologous repeats, the most distal of which is thought to be responsible for binding specificity. The relatively short C-terminal intracellular domain interacts with a variety of cytoplasmic proteins, including β -catenin, to regulate cadherin function. The cadherin superfamily includes cadherins, protocadherins, desmogleins and desmocollins. FAT4 (FAT tumor suppressor homolog 4), also known as FAT-J, CDHF14 or CDHR11, is a 4,981 amino acid single-pass type I membrane protein that belongs to the protocadherin subfamily of cadherins and localizes to the primary cilia of kidney. Widely expressed, FAT4 contains 34 cadherin domains, 6 EGF-like domains and 2 laminin G-like domains. FAT4 may function in the regulation of planar cell polarity.

REFERENCES

1. Sano, K., et al. 1993. Protocadherins: a large family of cadherin-related molecules in central nervous system. *EMBO J.* 12: 2249-2256.
2. Wu, Q. and Maniatis, T. 1999. A striking organization of a large family of human neural cadherin-like cell adhesion genes. *Cell* 97: 779-790.
3. Suzuki, S.T. 2000. Recent progress in protocadherin research. *Exp. Cell Res.* 261: 13-18.
4. Hill, E., et al. 2001. Cadherin superfamily proteins in *Caenorhabditis elegans* and *Drosophila melanogaster*. *J. Mol. Biol.* 305: 1011-1024.

CHROMOSOMAL LOCATION

Genetic locus: FAT4 (human) mapping to 4q28.1.

PRODUCT

FAT4 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 FAT4 shRNA Plasmid (h): sc-88877-SH and FAT4 shRNA (h) Lentiviral Particles: sc-88877-V as alternate gene silencing products.

For independent verification of FAT4 (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-88877A, sc-88877B and sc-88877C.

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

FAT4 siRNA (h) is recommended for the inhibition of FAT4 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.

GENE EXPRESSION MONITORING

FAT4 (1654CT645.1.70): sc-517330 is recommended as a control antibody for monitoring of FAT4 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 MarkerTM 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 FAT4 gene expression knockdown using RT-PCR Primer: FAT4 (h)-PR: sc-88877-PR (20 μl). Annealing temperature for the primers should be $55-60^{\circ}\text{C}$ and the extension temperature should be $68-72^{\circ}\text{C}$.

SELECT PRODUCT CITATIONS

1. Malgundkar, S.H., et al. 2020. FAT4 silencing promotes epithelial-to-mesenchymal transition and invasion via regulation of YAP and β -catenin activity in ovarian cancer. *BMC Cancer* 20: 374.

RESEARCH USE

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

PROTOCOLS

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