

TWIK-1 siRNA (h): sc-42349

BACKGROUND

K⁺ channels are divided into three subclasses, reflecting the number of transmembrane segments (TMS), which are designated 6TMS, 4TMS and 2TMS. Members of the 4TMS class contain two distinct pore regions, and include TASK, TREK, TRAAK and TWIK. TWIK-1 mRNA is expressed abundantly in brain and at lower levels in lung, kidney and skeletal muscle. TWIK-2 shares low sequence homology with other mammalian family group members, and only 34% homology with TWIK-1. Human TWIK-2 is expressed in pancreas, placenta and heart, while mouse TWIK-2 is expressed in liver. TWIK-2 is inhibited by intracellular, but not extracellular, acidification.

REFERENCES

1. Lesage, F., et al. 1996. TWIK-1, a ubiquitous human weakly inward rectifying K⁺ channel with a novel structure. *EMBO J.* 15: 1004-1011.
2. Fink, M., et al. 1996. Cloning, functional expression and brain localization of a novel unconventional outward rectifier K⁺ channel. *EMBO J.* 15: 6854-6862.
3. Duprat, F., et al. 1997. TASK, a human background K⁺ channel to sense external pH variations near physiological pH. *EMBO J.* 16: 5464-5471.
4. Lesage, F., et al. 1997. The structure, function and distribution of the mouse TWIK-1 K⁺ channel. *FEBS Lett.* 402: 28-32.
5. Maingret, F., et al. 1999. TRAAK is a mammalian neuronal mechano-gated K⁺ channel. *J. Biol. Chem.* 274: 1381-1387.
6. Pountney, D.J., et al. 1999. Identification and cloning of TWIK-originated similarity sequence (TOSS): a novel human 2-pore K⁺ channel principal subunit. *FEBS Lett.* 450: 191-196.

CHROMOSOMAL LOCATION

Genetic locus: KCNK1 (human) mapping to 1q42.2.

PRODUCT

TWIK-1 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see TWIK-1 shRNA Plasmid (h): sc-42349-SH and TWIK-1 shRNA (h) Lentiviral Particles: sc-42349-V as alternate gene silencing products.

For independent verification of TWIK-1 (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-42349A, sc-42349B and sc-42349C.

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

TWIK-1 siRNA (h) is recommended for the inhibition of TWIK-1 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 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

TWIK-1 (4D7): sc-517040 is recommended as a control antibody for monitoring of TWIK-1 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 TWIK-1 gene expression knockdown using RT-PCR Primer: TWIK-1 (h)-PR: sc-42349-PR (20 μ l, 454 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Chai, S., et al. 2017. Contribution of two-pore K⁺ channels to cardiac ventricular action potential revealed using human iPSC-derived cardiomyocytes. *Am. J. Physiol. Heart Circ. Physiol.* 312: H1144-H1153.

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.