

TREK-1 siRNA (r): sc-270403

BACKGROUND

TREK-1 (also designated TWIK-related K⁺ channel) and TREK-2 are members of the tandem-pore K⁺ channel family and belong to the class of mechanosensitive and fatty acid-stimulated K⁺ channels. TREK-1 has an outwardly rectifying current-voltage relationship, while TREK-2 shows inward rectification. Both TREK-1 and TREK-2 are activated by arachidonic acid and other naturally occurring unsaturated free fatty acids. These family members possess two pore-forming domains and four transmembrane segments. TREK-2 is a 538-amino acid protein and shares 65% amino acid sequence identity with TREK-1. TREK-1 is expressed in many different tissues, particularly lung and brain, while TREK-2 is expressed mainly in the cerebellum, spleen, and testis.

REFERENCES

1. Pongs, O. 1992. Molecular biology of voltage-dependent potassium channels. *Physiol. Rev.* 72: 569-588.
2. Jan, L.Y., et al. 1994. Potassium channels and their evolving gates. *Nature* 371: 119-122.
3. Wei, A., et al. 1996. Eight potassium channel families revealed by the *C. elegans* genome project. *Neuropharmacology* 35: 805-829.
4. Fink, M., et al. 1996. Cloning, functional expression and brain localization of a novel unconventional outward rectifier K⁺ channel. *EMBO J.* 15: 6854-6862.
5. Patel, A.J., et al. 1998. A mammalian two pore domain mechano-gated S-like K⁺ channel. *EMBO J.* 17: 4283-4290.
6. Maingret, F., et al. 1999. TRAAK is a mammalian neuronal mechano-gated K⁺ channel. *J. Biol. Chem.* 274: 1381-1387.
7. Bang, H., et al. 2000. TREK-2, a new member of the mechanosensitive tandem-pore K⁺ channel family. *J. Biol. Chem.* 275: 17412-17419.

CHROMOSOMAL LOCATION

Genetic locus: Kcnk2 (rat) mapping to 13q26.

PRODUCT

TREK-1 siRNA (r) 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 TREK-1 shRNA Plasmid (r): sc-270403-SH and TREK-1 shRNA (r) Lentiviral Particles: sc-270403-V as alternate gene silencing products.

For independent verification of TREK-1 (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-270403A, sc-270403B and sc-270403C.

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

TREK-1 siRNA (r) is recommended for the inhibition of TREK-1 expression in rat 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

TREK-1 (F-6): sc-398449 is recommended as a control antibody for monitoring of TREK-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 TREK-1 gene expression knockdown using RT-PCR Primer: TREK-1 (r)-PR: sc-270403-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.