

KV1.5 siRNA (h): sc-42716

BACKGROUND

Voltage-gated K⁺ channels in the plasma membrane control the repolarization and the frequency of action potentials in neurons, muscles and other excitable cells. The KV gene family encodes more than 30 genes that comprise the subunits of the K⁺ channels, and they vary in their gating and permeation properties, subcellular distribution and expression patterns. Functional KV channels assemble as tetramers consisting of pore-forming α subunits (KV α), which include the KV1, KV2, KV3 and KV4 proteins, and accessory or KV β subunits that modify the gating properties of the coexpressed KV α subunits. Differences exist in the patterns of trafficking, biosynthetic processing and surface expression of the major KV1 subunits (KV1.1, KV1.2, KV1.4, KV1.5 and KV1.6) expressed in rat and human brain, suggesting that the individual protein subunits are highly regulated to control for the assembly and formation of functional neuronal channels.

REFERENCES

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- Veh, R.W., et al. 1995. Immunohistochemical localization of five members of the KV1 channel subunits: contrasting subcellular locations and neuron-specific co-localizations in rat brain. *Eur. J. Neurosci.* 7: 2189-2205.
- Shi, G., et al. 1996. β subunits promote K⁺ channel surface expression through effects early in biosynthesis. *Neuron* 16: 843-852.
- Rhodes, K.J., et al. 1997. Association and colocalization of the KV β 1 and KV β 2 β subunits with KV1 α subunits in mammalian brain K⁺ channel complexes. *J. Neurosci.* 17: 8246-8258.
- Coleman, S.K., et al. 1999. Subunit composition of KV1 channels in human CNS. *J. Neurochem.* 73: 849-858.
- Manganas, L.N., et al. 2000. Subunit composition determines KV1 potassium channel surface expression. *J. Biol. Chem.* 275: 29685-29693.

CHROMOSOMAL LOCATION

Genetic locus: KCNA5 (human) mapping to 12p13.32.

PRODUCT

KV1.5 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 KV1.5 shRNA Plasmid (h): sc-42716-SH and KV1.5 shRNA (h) Lentiviral Particles: sc-42716-V as alternate gene silencing products.

For independent verification of KV1.5 (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-42716A, sc-42716B and sc-42716C.

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

KV1.5 siRNA (h) is recommended for the inhibition of KV1.5 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

KV1.5 (A-3): sc-377110 is recommended as a control antibody for monitoring of KV1.5 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 KV1.5 gene expression knockdown using RT-PCR Primer: KV1.5 (h)-PR: sc-42716-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.