SANTA CRUZ BIOTECHNOLOGY, INC.

KIR3.1 siRNA (m): sc-42617

BACKGROUND

G protein-coupled inwardly rectifying potassium channels (KIR3.1 through KIR3.4) are coupled to numerous neurotransmitter receptors in the brain and are abundantly expressed in the olfactory bulb, hippocampus, neocortex, dentate gyrus, cerebellar cortex and thalamus regions of the brain. Also known as GIRK, KIR3 potassium channels localize to the soma and dendrites as well as axons of neurons. Liberated $G_{\beta\gamma}$ subunits from G protein heterotrimers bind to and regulate KIR3 channel activity. $G_{\beta 3}$- and $G_{\beta 4}$-containing $G_{\beta\gamma}$ dimers bind directly to cytoplasmic domains of KIR3 proteins and increase the K^+ current while $G_{\beta 5}$-containing $G_{\beta\gamma}$ dimers inhibit KIR3 K^+ current. KIR3 activity is also inhibited by tyrosine phosphorylation. Brain-derived neurotrophic factor activates receptor tyrosine kinase B, which then phosphorylates KIR3 tyrosine residues, effectively inactivating the KIR3 channels.

REFERENCES

1. Braun, A.P., et al, 1992. Activation of α 1-adrenoceptors modulates the inwardly rectifying potassium currents of mammalian atrial myocytes. Pflugers Arch. 421: 431-439.
2. Ponce, A., et al. 1996. G protein-gated inward rectifier K^+ channel proteins (GIRK1) are present in the soma and dendrites as well as in nerve terminals of specific neurons in the brain. J. Neurosci. 16: 1990-2001.
3. Farkas, R.H., et al. 1997. Neurotensin and dopamine D2 activation oppositely regulate the same K^+ conductance in rat midbrain dopaminergic neurons. Neurosci. Lett. 231: 21-24.
4. Lei, Q., et al. 2000. Activation and inhibition of G protein-coupled inwardly rectifying potassium (KIR3) channels by G protein by subunits. Proc. Natl. Acad. Sci. USA 97: 9771-9776.
5. Rogalski, S.L., et al. 2000. TrkB activation by brain-derived neurotrophic factor inhibits the G protein-gated inward rectifier KIR3 by tyrosine phosphorylation of the channel. J. Biol. Chem. 275: 25082-25088.

CHROMOSOMAL LOCATION

Genetic locus: Kcnj3 (mouse) mapping to 2 C1.1.

PRODUCT

KIR3.1 siRNA (m) 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 KIR3.1 shRNA Plasmid (m): sc-42617-SH and KIR3.1 shRNA (m) Lentiviral Particles: sc-42617-V as alternate gene silencing products.

For independent verification of KIR3.1 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-42617A, sc-42617B and sc-42617C.

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 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

KIR3.1 siRNA (m) is recommended for the inhibition of KIR3.1 expression in mouse 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

KIR3.1 (A-4): sc-365457 is recommended as a control antibody for monitoring of KIR3.1 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 KIR3.1 gene expression knockdown using RT-PCR Primer: KIR3.1 (m)-PR: sc-42617-PR (20 µl). 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.