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HERG siRNA (m): sc-42498

BACKGROUND

Human ether-a-go-go related gene (HERG) encodes the pore-forming α subunit of the delayed rectifier potassium channel IKr. The HERG subunit contains six transmembrane α -helices with a reentrant "pore-loop" between the fifth and the sixth transmembrane helices. The two N-terminal splice variants of HERG include the full-length isoform 1 α and the shorter isoform 1 β . Isoform 1 β lacks the PAS motif and deactivates at a faster rate than isoform 1 α . Residues within the C-terminal play a role in channel expression and channel gating, including voltage-dependent activation. HERG is expressed in the heart and is more abundantly expressed in the ventricles than in the atria. Mutations in the gene encoding HERG increase beat-to-beat variability and early after depolarization. These fluctuations facilitate the genesis and propagation of premature heartbeats associated with inheritable long QT syndrome.

REFERENCES

- Heginbotham, L., et al. 1994. Mutations in the K⁺ channel signature sequence. *Biophys. J.* 66: 1061-1067.
- Curran, M.E., et al. 1995. A molecular basis for cardiac arrhythmia: HERG mutations cause long QT syndrome. *Cell* 80: 795-803.
- Sanguinetti, M.C., et al. 1995. A mechanistic link between an inherited and an acquired cardiac arrhythmia: HERG encodes the IKr potassium channel. *Cell* 81: 299-307.
- Lees-Miller, J.P., et al. 1997. Electrophysiological characterization of an alternatively processed Erg K⁺ channel in mouse and human hearts. *Circ. Res.* 81: 719-726.

CHROMOSOMAL LOCATION

Genetic locus: Kcnh2 (mouse) mapping to 5 A3.

PRODUCT

HERG 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see HERG shRNA Plasmid (m): sc-42498-SH and HERG shRNA (m) Lentiviral Particles: sc-42498-V as alternate gene silencing products.

For independent verification of HERG (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-42498A, sc-42498B and sc-42498C.

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

HERG siRNA (m) is recommended for the inhibition of HERG 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 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

HERG (F-12): sc-377388 is recommended as a control antibody for monitoring of HERG 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 HERG gene expression knockdown using RT-PCR Primer: HERG (m)-PR: sc-42498-PR (20 μ l, 522 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

- Jehle, J., et al. 2012. Regulation of apoptosis in HL-1 cardiomyocytes by phosphorylation of the receptor tyrosine kinase EphA2 and protection by lithocholic acid. *Br. J. Pharmacol.* 167: 1563-1572.

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.