

TRPM6 siRNA (h): sc-76754

BACKGROUND

Transient receptor potential ion channels (TRPC) are a superfamily of six transmembrane segment-spanning gated cation channels. TRP subtypes mediate store-operated Ca^{2+} entry, a process involving Ca^{2+} influx and replenishment of Ca^{2+} stores formerly emptied through the action of inositol 1,4,5-trisphosphate production and other Ca^{2+} mobilizing agents. TRP ion channels influence calcium-depletion-induced calcium influx processes in response to chemo-, mechano- and osmo-regulatory events. TRPM6 (transient receptor potential cation channel, subfamily M, member 6), also known as HSH, HMGX, HOMG, CHAK2 or HOMG1, is a 2,022 amino acid multi-pass membrane protein that is highly expressed in kidney and colon. An essential ion channel and a serine/threonine-protein kinase, TRPM6 is crucial for magnesium homeostasis and has an important role in epithelial magnesium transport and in the active magnesium absorption in the gut and kidney.

REFERENCES

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- Rondón, L.J., et al. 2008. Dietary inulin in mice stimulates Mg^{2+} absorption and modulates TRPM6 and TRPM7 expression in large intestine and kidney. *Magnes. Res.* 21: 224-231.
- Song, Y., et al. 2009. Common genetic variants of the ion channel transient receptor potential membrane melastatin 6 and 7 (TRPM6 and TRPM7), magnesium intake, and risk of type 2 diabetes in women. *BMC Med. Genet.* 10: 4.
- Esteban-Oliva, D., et al. 2009. Long-term follow-up of a patient with primary hypomagnesaemia and secondary hypocalcaemia due to a novel TRPM6 mutation. *Eur. J. Pediatr.* 168: 439-442.
- Cao, G., et al. 2009. Regulation of the epithelial Mg^{2+} channel TRPM6 by estrogen and the associated repressor protein of estrogen receptor activity (REA). *J. Biol. Chem.* 284: 14788-14795.

CHROMOSOMAL LOCATION

Genetic locus: TRPM6 (human) mapping to 9q21.13.

PRODUCT

TRPM6 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 μM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see TRPM6 shRNA Plasmid (h): sc-76754-SH and TRPM6 shRNA (h) Lentiviral Particles: sc-76754-V as alternate gene silencing products.

For independent verification of TRPM6 (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-76754A, sc-76754B and sc-76754C.

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

TRPM6 siRNA (h) is recommended for the inhibition of TRPM6 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 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

TRPM6 (D-6): sc-365536 is recommended as a control antibody for monitoring of TRPM6 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor TRPM6 gene expression knockdown using RT-PCR Primer: TRPM6 (h)-PR: sc-76754-PR (20 μl). Annealing temperature for the primers should be $55-60^{\circ}\text{C}$ and the extension temperature should be $68-72^{\circ}\text{C}$.

RESEARCH USE

For research use only, not for use in diagnostic procedures.