

Orai2 siRNA (m): sc-76004

BACKGROUND

Orai2 (orai calcium release-activated calcium modulator 2), also known as CBCIP2 (CAP-binding protein complex-interacting protein 2) or TMEM142B (transmembrane protein 142B), is a 254 amino acid multi-pass membrane protein that belongs to the orai family of proteins. Localizing to the plasma membrane, Orai2 plays an important role in store-operated calcium (SOC) 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. CRAC channels are responsible for mediating calcium influx in T cells and play an important role in the immune response. Orai2 specifically increases the Ca^{2+} -selective current through coaction with the Ca^{2+} sensor Stim1.

REFERENCES

1. Mercer, J.C., et al. 2006. Large store-operated calcium selective currents due to coexpression of Orai1 or Orai2 with the intracellular calcium sensor, Stim1. *J. Biol. Chem.* 281: 24979-24990.
2. Feske, S., et al. 2006. A mutation in Orai1 causes immune deficiency by abrogating CRAC channel function. *Nature* 441: 179-185.
3. DeHaven, W.I., et al. 2007. Calcium inhibition and calcium potentiation of Orai1, Orai2 and Orai3 calcium release-activated calcium channels. *J. Biol. Chem.* 282: 17548-17556.
4. Online Mendelian Inheritance in Man, OMIM[™]. 2007. Johns Hopkins University, Baltimore, MD. MIM Number: 610929. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>

CHROMOSOMAL LOCATION

Genetic locus: Orai2 (mouse) mapping to 5 G2.

PRODUCT

Orai2 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 Orai2 shRNA Plasmid (m): sc-76004-SH and Orai2 shRNA (m) Lentiviral Particles: sc-76004-V as alternate gene silencing products.

For independent verification of Orai2 (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-76004A, sc-76004B and sc-76004C.

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

Orai2 siRNA (m) is recommended for the inhibition of Orai2 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

Orai2 (G-5): sc-376757 is recommended as a control antibody for monitoring of Orai2 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 Orai2 gene expression knockdown using RT-PCR Primer: Orai2 (m)-PR: sc-76004-PR (20 μl). Annealing temperature for the primers should be $55-60^{\circ}\text{C}$ and the extension temperature should be $68-72^{\circ}\text{C}$.

SELECT PRODUCT CITATIONS

1. Sundivakkam, P.C., et al. 2012. The Ca^{2+} sensor stromal interaction molecule 1 (STIM1) is necessary and sufficient for the store-operated Ca^{2+} entry function of transient receptor potential canonical (TRPC) 1 and 4 channels in endothelial cells. *Mol. Pharmacol.* 81: 510-526.
2. Goldberg, J., et al. 2020. Targeting of intracellular Ca^{2+} stores as a therapeutic strategy against age-related neurotoxicities. *NPJ Aging Mech. Dis.* 6: 10.

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.