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SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

mail@szabo-scandic.com

www.szabo-scandic.com

[linkedin.com/company/szaboscandic](https://www.linkedin.com/company/szaboscandic) 



robo3 siRNA (h): sc-44498

BACKGROUND

Secreted leucine-rich repeat-containing proteins 1-3 (Slit1-3) are secreted glycoproteins that influence axonal guidance and mediate normal neural progression by acting as high-affinity signaling ligands for the repulsive guidance receptors, robo 1 and robo 2 (also designated Roundabout 1 and 2). Within the developing CNS of different vertebrate systems, Slit proteins are expressed in equivalent regions, suggesting there is a conservation of function for vertebrate homologs. Robo3 plays a crucial role in controlling axon guidance at the midline of the CNS. Two human robo3 isoforms, robo3A and robo3B, which differ by the insertion of 26 amino acids at the N-terminus, appear to be evolutionary conserved. Robo3 guides commissural axons by preventing premature sensitivity to Slit proteins thus inhibiting Slit signaling through robo1. Together, the robo proteins prescribe developmental paths during neural development.

REFERENCES

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2. Woods, C.G., et al. 2004. Neuroscience. Crossing the midline. *Science* 304: 1455-1456.
3. Tayler, T.D., et al. 2004. Compartmentalization of visual centers in the *Drosophila* brain requires Slit and robo proteins. *Development* 131: 5935-5945.
4. Marillat, V., et al. 2004. The slit receptor RIG-1/robo3 controls midline crossing by hindbrain precerebellar neurons and axons. *Neuron* 43: 69-79.
5. Bosley, T.M., et al. 2005. Neurologic features of horizontal gaze palsy and progressive scoliosis with mutations in robo3. *Neurology* 64: 1196-1203.
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CHROMOSOMAL LOCATION

Genetic locus: ROBO3 (human) mapping to 11q24.2.

PRODUCT

robo3 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 robo3 shRNA Plasmid (h): sc-44498-SH and robo3 shRNA (h) Lentiviral Particles: sc-44498-V as alternate gene silencing products.

For independent verification of robo3 (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-44498A, sc-44498B and sc-44498C.

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

robo3 siRNA (h) is recommended for the inhibition of robo3 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor robo3 gene expression knockdown using RT-PCR Primer: robo3 (h)-PR: sc-44498-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.