RAMP3 siRNA (m): sc-40897



The Power to Question

BACKGROUND

Receptor activity-modifying proteins (RAMPs) are transmembrane accessory proteins that influence the pharmacological profiles of the calcitonin receptor-like receptors (CRLR). RAMPs associate with CRLR in the endoplasmic reticulum and facilitate the glycosylation and transport of CRLR to the cell surface, where the mature protein then operates as a receptor for two structurally related vasodilatory peptides, calcitonin-gene-related peptide (CGRP) or adrenomedullin (ADM). RAMP-1 associating with CRLR confers a CGRP receptor, while RAMP-2 and RAMP-3 preferentially induce a responsiveness to ADM. RAMP proteins, including RAMP-1, RAMP-2 and RAMP-3, are structurally similar as they are type I receptors, which have a single extracellular N-terminus and a cytoplasmic C-terminus, and they share approximately 55% sequence similarity. RAMP-1 expression is highest in the uterus, brain and gastrointestinal tract, whereas RAMP-2 and RAMP-3 are highest in lung, breast and fetal tissues.

REFERENCES

- McLatchie, L.M., et al. 1998. RAMPs regulate the transport and ligand specificity of the calcitonin-receptor-like receptor. Nature 393: 333-339.
- Sams, A., et al. 1998. Expression of calcitonin receptor-like receptor and receptor-activity-modifying proteins in human cranial arteries. Neurosci. Lett. 258: 41-44.
- Fraser, N.J., et al. 1999. The amino terminus of receptor activity modifying proteins is a critical determinant of glycosylation state and ligand binding of calcitonin receptor-like receptor. Mol. Pharmacol. 55: 1054-1059.
- 4. Foord, S.M., et al. 1999. RAMPs: accessory proteins for seven transmembrane domain receptors. Trends Pharmacol. Sci. 20: 184-187.
- Kamitani, S., et al. 1999. The RAMP2/CRLR complex is a functional adrenomedullin receptor in human endothelial and vascular smooth muscle cells. FEBS Lett. 448: 111-114.
- Drake, W.M., et al. 1999. Desensitization of CGRP and adrenomedullin receptors in SK-N-MC cells: implications for the RAMP hypothesis. Endocrinology 140: 533-537.

CHROMOSOMAL LOCATION

Genetic locus: Ramp3 (mouse) mapping to 11 A1.

PRODUCT

RAMP3 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 RAMP3 shRNA Plasmid (m): sc-40897-SH and RAMP3 shRNA (m) Lentiviral Particles: sc-40897-V as alternate gene silencing products.

For independent verification of RAMP3 (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-40897A, sc-40897B and sc-40897C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20 $^{\circ}$ C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20 $^{\circ}$ 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

RAMP3 siRNA (m) is recommended for the inhibition of RAMP3 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-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

RAMP3 (G-1): sc-365313 is recommended as a control antibody for monitoring of RAMP3 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-lgG κ BP-HRP: sc-516102 or m-lgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-lgG κ BP-FITC: sc-516140 or m-lgG 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 RAMP3 gene expression knockdown using RT-PCR Primer: RAMP3 (m)-PR: sc-40897-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.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

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