

CT-R siRNA (m): sc-39909

BACKGROUND

Calcitonin (CT) is a circulating peptide hormone that is secreted from the thyroid and specifically binds to surface calcitonin receptors (CT-R) to regulate calcium homeostasis. These receptors represent a distinct family of seven-transmembrane proteins, which include receptors for parathyroid hormone/parathyroid-related peptide, secretin and glucagon. CT-Rs induce intracellular signaling by coupling to multiple heterotrimeric G proteins, where they then activate several signal transduction pathways involving adenylyl cyclase, phospholipase C and map kinases. The gene encoding CT-R consists of numerous exons separated by larger introns, which are modified to produce multiple splice variants. These functionally unique isoforms display differential tissue distribution and preferentially associate with specific G proteins to recruit distinct signaling intermediates. In osteoclasts and embryonic kidney cells, CT binding to the CT-R stimulates the map kinases ERK 1/2 and PKC activity through the phosphorylation of the adaptor proteins Shc and HEF1, and this induction occurs independently from PKA and adenylyl cyclase mediated signaling.

REFERENCES

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2. Kuestner, R.E., et al. 1994. Cloning and characterization of an abundant subtype of the human calcitonin receptor. *Mol. Pharmacol.* 46: 246-255.
3. Yamin, M., et al. 1994. Cloning and characterization of a mouse brain calcitonin receptor complementary deoxyribonucleic acid and mapping of the calcitonin receptor gene. *Endocrinology* 135: 2635-2643.
4. Chen, Y., et al. 1998. The calcitonin receptor stimulates Shc tyrosine phosphorylation and ERK 1/2 activation. Involvement of G_i, protein kinase C, and calcium. *J. Biol. Chem.* 273: 19809-19816.
5. Shyu, J.F., et al. 1999. Protein kinase C antagonizes pertussis-toxin-sensitive coupling of the calcitonin receptor to adenylyl cyclase. *Eur. J. Biochem.* 262: 95-101.
6. Zhang, Z., et al. 1999. Cytoskeleton-dependent tyrosine phosphorylation of the p130^{Cas} family member HEF1 downstream of the G protein-coupled calcitonin receptor. Calcitonin induces the association of HEF1, paxillin, and focal adhesion kinase. *J. Biol. Chem.* 274: 25093-25098.
7. Nishikawa, T., et al. 1999. A novel calcitonin receptor gene in human osteoclasts from normal bone marrow. *FEBS Lett.* 458: 409-414.

CHROMOSOMAL LOCATION

Genetic locus: Calcr (mouse) mapping to 6 A1.

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.

PRODUCT

CT-R 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 CT-R shRNA Plasmid (m): sc-39909-SH and CT-R shRNA (m) Lentiviral Particles: sc-39909-V as alternate gene silencing products.

For independent verification of CT-R (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-39909A, sc-39909B and sc-39909C.

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

CT-R siRNA (m) is recommended for the inhibition of CT-R 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor CT-R gene expression knockdown using RT-PCR Primer: CT-R (m)-PR: sc-39909-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.