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Diagnostik & molekulare Diagnostik



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CaSR siRNA (m): sc-44374



The Power to Question

BACKGROUND

Extracellular calcium-sensing receptor (CaSR), also designated parathyroid cell calcium-sensing receptor, is an integral membrane protein that belongs to the G protein-coupled receptor 3 family. CaSR is involved in maintaining a stable calcium concentration by acting as an sensor of the extracellular calcium levels for the parathyroid and kidney. Its activity is mediated by a G protein which activates a phosphatidylinositol-calcium second messenger system. Defects that activate CaSR cause autosomal dominant hypocalcemia, whereas mutations that inactivate the protein cause familial hypocalciuric hypercalcemia. CaSR is expressed mainly in kidney, and is also expressed in intestine, placenta and brain.

REFERENCES

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- Freichel, M., et al. 1996. Expression of a calcium-sensing receptor in a human medullary thyroid carcinoma cell line and its contribution to calcitonin secretion. Endocrinology 137: 3842-3848.
- Bikle, D.D., et al. 1996. Changes in calcium responsiveness and handling during keratinocyte differentiation. Potential role of the calcium receptor. J. Clin. Invest. 97: 1085-1093.
- Stock, J.L., et al. 1999. Autosomal dominant hypoparathyroidism associated with short stature and premature osteoarthritis. J. Clin. Endocrinol. Metab. 84: 3036-3040.
- Nakayama, T., et al. 2001. A novel mutation in Ca²⁺-sensing receptor gene in familial hypocalciuric hypercalcemia. Endocrine 15: 277-82.
- 7. Uckun-Kitapci, A., et al. 2005. A novel mutation (E767K) in the second extracellular loop of the calcium sensing receptor in a family with autosomal dominant hypocalcemia. Am. J. Med. Genet. A 132A: 125-129.

CHROMOSOMAL LOCATION

Genetic locus: Casr (mouse) mapping to 16 B3.

PRODUCT

CaSR 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 CaSR shRNA Plasmid (m): sc-44374-SH and CaSR shRNA (m) Lentiviral Particles: sc-44374-V as alternate gene silencing products.

For independent verification of CaSR (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-44374A, sc-44374B and sc-44374C.

RESEARCH USE

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

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

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

CaSR (6D4): sc-47741 is recommended as a control antibody for monitoring of CaSR 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 CaSR gene expression knockdown using RT-PCR Primer: CaSR (m)-PR: sc-44374-PR (20 μ l, 500 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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

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