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CaSR shRNA (h) Lentiviral Particles: sc-44373-V

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 a 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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- 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.
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CHROMOSOMAL LOCATION

Genetic locus: CASR (human) mapping to 3q21.1.

PRODUCT

CaSR shRNA (h) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 µl frozen stock containing 1.0 x 10⁶ infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see CaSR siRNA (h): sc-44373 and CaSR shRNA Plasmid (h): sc-44373-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

APPLICATIONS

CaSR shRNA (h) Lentiviral Particles is recommended for the inhibition of CaSR expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 µl frozen viral stock containing 1.0 x 10⁶ infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

CaSR (N-14): sc-32180 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 (secondary) reagents are recommended: 1) Western Blotting: use donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor CaSR gene expression knockdown using RT-PCR Primer: CaSR (h)-PR: sc-44373-PR (20 µl, 443 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

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