

PTH/PTHrP-R siRNA (h): sc-36327

BACKGROUND

Parathyroid hormone (PTH), which is also designated parathyrin, is an 84 amino acid single chain peptide that functions to regulate calcium metabolism by raising blood levels of calcium through various mechanisms. PTH stimulates bone formation to increase bone mass and strength in rats and humans. Within the PTH molecule, the essential activity is associated with the first 34 amino acids at the amino-terminus of the molecule. The gene encoding PTH maps to human chromosome 11p15.3-p15.1. Parathyroid hormone-related protein (PTH-rP) is an autocrine factor that is structurally related to PTH yet, unlike PTH, which is synthesized only by the parathyroid cells, PTH-rP is synthesized by several cell types. PTH-rP regulates endochondral bone development and epithelial-mesenchymal interactions during the formation of the mammary glands and teeth. Isolated from the culture medium of a human lung cancer cell line, PTH-rP produces PTH-like effects that are characterized as humoral hypercalcemia of malignancy. PTH and PTH-rP are both regulated by vitamin D and steroid hormones and preferentially bind to specific PTH/PTH-rP receptors, then activate adenylate cyclase or PLC β via PKC activation.

REFERENCES

1. Bruns, M.E., et al. 1995. Expression of parathyroid hormone-related peptide and its receptor messenger ribonucleic acid in human amnion and chorion-decidua: implications for secretion and function. *Am. J. Obstet. Gynecol.* 173: 739-746.
2. Iezzoni, J.C., et al. 1998. Coexpression of parathyroid hormone-related protein and its receptor in breast carcinoma: a potential autocrine effector system. *Mod. Pathol.* 11: 265-270.
3. Takasu, H., et al. 1999. Dual signaling and ligand selectivity of the human PTH/PTHrP receptor. *J. Bone Miner. Res.* 14: 11-20.
4. Huang, Z., et al. 1999. Role of signal transduction in internalization of the G protein-coupled receptor for parathyroid hormone (PTH) and PTH-related protein. *Endocrinology* 140: 1294-1300.
5. Mannstadt, M., et al. 1999. Receptors for PTH and PTHrP: their biological importance and functional properties. *Am. J. Physiol.* 277: 665-675.

CHROMOSOMAL LOCATION

Genetic locus: PTH1R (human) mapping to 3p21.31.

PRODUCT

PTH/PTHrP-R 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 PTH/PTHrP-R shRNA Plasmid (h): sc-36327-SH and PTH/PTHrP-R shRNA (h) Lentiviral Particles: sc-36327-V as alternate gene silencing products.

For independent verification of PTH/PTHrP-R (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-36327A, sc-36327B and sc-36327C.

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

PTH/PTHrP-R siRNA (h) is recommended for the inhibition of PTH/PTHrP-R 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.

GENE EXPRESSION MONITORING

PTH/PTHrP-R (3D1.1): sc-12722 is recommended as a control antibody for monitoring of PTH/PTHrP-R 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-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG 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 PTH/PTHrP-R gene expression knockdown using RT-PCR Primer: PTH/PTHrP-R (h)-PR: sc-36327-PR (20 μ l, 448 bp). 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.