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APLNR siRNA (h): sc-44732

BACKGROUND

The apelin receptor (APLNR) is a G protein-coupled integral membrane protein exhibiting a hypothalamic distribution in brain, glial cells, astrocytes and neuronal subpopulations. APLNR is bound by its cognate neuropeptide ligand, apelin, promoting receptor internalization to the nucleus and dose-dependent inhibition of forskolin-induced cAMP production. However, deletion studies of the apelin agonist have shown that internalization is not mandatory for decreasing vasopressin release, a hypotensive action of APLNR signaling. Further evidence for functional dissociation of APLNR stimulation and internalization was exhibited *in vitro* using mutational studies of a nuclear localization signal sequence. These findings may suggest the presence of multiple, functionally-differing conformational states for the receptor. Stress studies in rodents have shown APLNR is under negative regulation by glucocorticoids and may be involved in controlling hypothalamic function. APLNR also functions as an alternate coreceptor with CD4 for HIV-1 infection.

REFERENCES

1. De Mota, N., et al. 2000. Cloning, pharmacological characterization and brain distribution of the rat apelin receptor. *Neuroendocrinology* 72: 400-407.
2. Reaux, A., et al. 2001. Physiological role of a novel neuropeptide, apelin, and its receptor in the rat brain. *J. Neurochem.* 77: 1085-1096.
3. O'Carroll, A.M., et al. 2003. APJ receptor mRNA expression in the rat hypothalamic paraventricular nucleus: regulation by stress and glucocorticoids. *J. Neuroendocrinol.* 15: 1095-1101.
4. El Messari, S., et al. 2004. Functional dissociation of apelin receptor signaling and endocytosis: implications for the effects of apelin on arterial blood pressure. *J. Neurochem.* 90: 1290-1301.
5. Lee, D.K., et al. 2004. Agonist-independent nuclear localization of the apelin, Angiotensin AT1, and Bradykinin B2 receptors. *J. Biol. Chem.* 279: 7901-7908.
6. Klein, M.J., et al. 2005. Immunocytochemical localisation of the apelin receptor, APJ, to human cardiomyocytes, vascular smooth muscle and endothelial cells. *Regul. Pept.* 126: 233-240.

CHROMOSOMAL LOCATION

Genetic locus: APLNR (human) mapping to 11q12.1.

PRODUCT

APLNR 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 APLNR shRNA Plasmid (h): sc-44732-SH and APLNR shRNA (h) Lentiviral Particles: sc-44732-V as alternate gene silencing products.

For independent verification of APLNR (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-44732A, sc-44732B and sc-44732C.

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

APLNR siRNA (h) is recommended for the inhibition of APLNR 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

APLNR (3C3-7): sc-517300 is recommended as a control antibody for monitoring of APLNR 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 APLNR gene expression knockdown using RT-PCR Primer: APLNR (h)-PR: sc-44732-PR (20 μ l, 550 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.