Orexin R-1 siRNA (m): sc-36132



The Power to Question

BACKGROUND

The hypothalamus is essential for maintaining homeostasis by integrating the vertebrate endocrine and nervous systems, thereby controlling temperature, thrist and hunger. Orexin-A and Orexin-B (also designated hypocretins) are hypothalamic neuropeptides that are derived from a single precursor, preproorexin, by proteolytic processing. These peptides bind to and activate two closely related, G protein-coupled receptors, designated Orexin receptor-1 (Orexin R-1) and Orexin receptor-2 (Orexin R-2). Orexin-A protein and preproorexin mRNA are localized to neurons within the lateral section of the hypothalamus, designated the "feeding center". Prepro-orexin mRNA is up-regulated during fasting, suggesting that orexins may play a role in the central feedback mechanism that regulates feeding behavior. Orexin has been shown to increase the release of GABA and glutamate from axons, a response seen as a result of most synaptic activities in the hypothalamic region.

REFERENCES

- Sakurai, T., et al. 1998. Orexins and Orexin receptors: a family of hypothalamic neuropeptides and G protein-coupled receptors that regulate feeding behavior. Cell 92: 573-585.
- Lund, P.E., et al. 2000. The Orexin OX1 receptor activates a novel Ca²⁺ influx pathway necessary for coupling to phospholipase C. J. Biol. Chem. 275: 30806-30812.
- Hervieu, G.J., et al. 2001. Gene expression and protein distribution of the Orexin-1 receptor in the rat brain and spinal cord. Neuroscience 103: 777-797.
- Guan, J.L., et al. 2002. Ultrastructural localization of Orexin-1 receptor in pre and post-synaptic neurons in the rat arcuate nucleus. Neurosci. Lett. 329: 209-212.
- Hilairet, S., et al. 2003. Hypersensitization of the Orexin 1 receptor by the CB1 receptor: evidence for cross-talk blocked by the specific CB1 antagonist, SR141716. J. Biol. Chem 278: 23731-23737.
- Barreiro, M.L., et al. 2004. Orexin 1 receptor messenger ribonucleic acid expression and stimulation of testosterone secretion by Orexin-A in rat testis. Endocrinology 145: 2297-2306.
- 7. Lang, M., et al. 2004. Structure-activity studies of orexin a and Orexin-B at the human Orexin 1 and Orexin 2 receptors led to orexin 2 receptor selective and Orexin 1 receptor preferring ligands. J. Med. Chem. 47: 1153-1160.
- 8. Milasta, S., et al. 2005. The sustainability of interactions between the Orexin-1 receptor and β -arrestin-2 is defined by a single C-terminal cluster of hydroxy amino acids and modulates the kinetics of ERK MAP kinase regulation. Biochem. J. 387: 573-584.

CHROMOSOMAL LOCATION

Genetic locus: Hcrtr1 (mouse) mapping to 4 D2.2.

PROTOCOLS

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

PRODUCT

Orexin R-1 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 Orexin R-1 shRNA Plasmid (m): sc-36132-SH and Orexin R-1 shRNA (m) Lentiviral Particles: sc-36132-V as alternate gene silencing products.

For independent verification of Orexin R-1 (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-36132A, sc-36132B and sc-36132C.

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

Orexin R-1 siRNA (m) is recommended for the inhibition of Orexin R-1 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Orexin R-1 gene expression knockdown using RT-PCR Primer: Orexin R-1 (m)-PR: sc-36132-PR (20 μ l). 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.

Santa Cruz Biotechnology, Inc. 1.800.457.3801 831.457.3801 Fax 831.457.3801 Europe +00800 4573 8000 49 6221 4503 0 www.scbt.com