

# Orexin R-1 siRNA (r): sc-61898

## BACKGROUND

The hypothalamus is essential for maintaining homeostasis by integrating the vertebrate endocrine and nervous systems, thereby controlling temperature, thirst and hunger. Orexin-A and Orexin-B (also designated hypocretins) are hypothalamic neuropeptides that are derived from a single precursor, prepro-orexin, 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 prepro-orexin mRNA are localized to neurons within the lateral section of the hypothalamus, designated the "feeding center". Prepro-orexin mRNA is upregulated 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

1. 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.
2. Lund, P.E., et al. 2000. The Orexin OX1 receptor activates a novel  $Ca^{2+}$  influx pathway necessary for coupling to phospholipase C. *J. Biol. Chem.* 275: 30806-30812.
3. 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.
4. 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.

## CHROMOSOMAL LOCATION

Genetic locus: Hcrtr1 (rat) mapping to 5q36.

## PRODUCT

Orexin R-1 siRNA (r) 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Orexin R-1 shRNA Plasmid (r): sc-61898-SH and Orexin R-1 shRNA (r) Lentiviral Particles: sc-61898-V as alternate gene silencing products.

For independent verification of Orexin R-1 (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-61898A, sc-61898B and sc-61898C.

## RESEARCH USE

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

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) for detailed protocols and support products.

## 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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

Orexin R-1 siRNA (r) is recommended for the inhibition of Orexin R-1 expression in rat 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  $\mu$ M in 66  $\mu$ 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.

## 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 (r)-PR: sc-61898-PR (20  $\mu$ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.