

MEL-1B-R siRNA (h): sc-40114

BACKGROUND

The melatonin receptors, MEL-1A-R and MEL-1B-R, are members of the superfamily of guanine nucleotide-binding regulatory protein (G protein)-coupled receptors. The melatonin receptors are activated by the hormone melatonin (Mel), which is secreted by the pineal gland at night as part of the circadian clock. MEL-1A-R is thought to be involved in pacing the biological clock. Both MEL-1A-R and MEL-1B-R are implicated in controlling cellular growth in response to melatonin. MEL-1B-R is an integral membrane protein expressed in retina and, to a lesser extent, in brain and hippocampus. Functional studies of NIH/3T3 cells stably expressing the MEL-1B-R melatonin receptor indicate that it is coupled to inhibition of adenylyl cyclase.

REFERENCES

1. Reppert, S.M., et al. 1995. Molecular characterization of a second melatonin receptor expressed in human retina and brain: the Mel1b melatonin receptor. *Proc. Natl. Acad. Sci. USA* 92: 8734-8738.
2. Reppert, S.M., et al. 1996. Cloning of a melatonin-related receptor from human pituitary. *FEBS Lett.* 386: 219-224.
3. Brzezinski, A. 1997. Melatonin in humans. *N. Engl. J. Med.* 336: 186-195.
4. Niles, L.P., et al. 1999. Melatonin receptor mRNA expression in human granulosa cells. *Mol. Cell. Endocrinol.* 156: 107-110.
5. Ebisawa, T., et al. 2000. Genetic polymorphisms of human melatonin 1b receptor gene in circadian rhythm sleep disorders and controls. *Neurosci. Lett.* 280: 29-32.

CHROMOSOMAL LOCATION

Genetic locus: MTNR1B (human) mapping to 11q14.3.

PRODUCT

MEL-1B-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 MEL-1B-R shRNA Plasmid (h): sc-40114-SH and MEL-1B-R shRNA (h) Lentiviral Particles: sc-40114-V as alternate gene silencing products.

For independent verification of MEL-1B-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-40114A, sc-40114B and sc-40114C.

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

MEL-1B-R siRNA (h) is recommended for the inhibition of MEL-1B-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

MEL-1A/B-R (B-8): sc-398788 is recommended as a control antibody for monitoring of MEL-1B-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 MEL-1B-R gene expression knockdown using RT-PCR Primer: MEL-1B-R (h)-PR: sc-40114-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Fan, T., et al. 2018. Inhibiting MT2-TFE3-dependent autophagy enhances melatonin-induced apoptosis in tongue squamous cell carcinoma. *J. Pineal Res.* E-published.

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.