SANTA CRUZ BIOTECHNOLOGY, INC.

GPR14 siRNA (m): sc-37107

BACKGROUND

GPR14 (G protein-coupled receptor), also designated SENR (sensory epithelium neuropeptide-like receptor), was initially cloned as an "orphan" receptor, which is a receptor that binds an unidentified natural ligand. Further studies have shown that urotensin II (UII), a cyclic neuropeptide, binds to GPR14 with very high affinity. Subsequently, cells transfected with GPR14 experience an increase in calcium concentration upon binding of urotensin II. It is the calcium influx and localization of GPR14 in heart tissues that suggests GPR14 may play a role in the contraction of vascular smooth muscles in response to the specific binding of urotensin II. GPRI4 is also detected in pancreas and, to a lesser extent, in brain tissues.

REFERENCES

1. Coulouarn, Y., et al. 1998. Cloning of the cDNA encoding the urotensin II precursor in frog and human reveals intense expression of the urotensin II gene in motoneurons of the spinal cord. Proc. Natl. Acad. Sci. USA 95: 15803-15808.
2. Civelli, O. 1998. Functional genomics: the search for novel neurotransmitters and neuropeptides. FEBS Lett. 430: 55-58.
3. Nothacker, H.P., et al. 1999. Identification of the natural ligand of an orphan G protein-coupled receptor involved in the regulation of vasoconstriction. Cell Biol. 1: 383-385.
4. Ames, R.S., et al. 1999. Human urotensin-II is a potent vasoconstrictor and agonist for the orphan receptor GPR14. Nature 401: 282-286.
5. Mori, M., et al. 1999. Urotensin II is the endogenous ligand of a G protein-coupled orphan receptor, SENR (GPR14). Biochem. Biophys. Res. Commun. 265: 123-129.
6. Coulouarn, Y., et al. 1999. Cloning, sequence analysis and tissue distribution of the mouse and rat urotensin II precursors. FEBS Lett. 457: 28-32.

CHROMOSOMAL LOCATION

Genetic locus: Uts2r (mouse) mapping to 11 E2.

PRODUCT

GPR14 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see GPR14 shRNA Plasmid (m): sc-37107-SH and GPR14 shRNA (m) Lentiviral Particles: sc-37107-V as alternate gene silencing products.

For independent verification of GPR14 (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-37107A, sc-37107B and sc-37107C.

PROTOCOLS

See our web site at 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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

GPR14 siRNA (m) is recommended for the inhibition of GPR14 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.

GENE EXPRESSION MONITORING

GPR14 (D-4): sc-514460 is recommended as a control antibody for monitoring of GPR14 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 GPR14 gene expression knockdown using RT-PCR Primer: GPR14 (m)-PR: sc-37107-PR (20 µl). 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.