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GASP-2 siRNA (m): sc-145338

BACKGROUND

G protein-coupled receptors play a role in many different stimulus-response pathways. G protein-coupled receptors mediate extracellular signals into intracellular signals (G protein activation). They respond to a wide variety of signaling molecules, including hormones, neurotransmitters and other proteins and peptides. GASP-2 (G protein-coupled receptor associated sorting protein 2), also known as GPRASP2, is an 838 amino acid protein that regulates a number of G protein-coupled receptors, such as CT-R (calcitonin receptor) and mAChR M1 (muscarinic acetylcholine receptor M1), through interactions with their cytoplasmic tails. Expressed primarily in brain, GASP-2 is a member of the GPRASP family and forms a complex with Huntingtin, with which it is thought to influence receptor trafficking.

REFERENCES

1. Lee, D.K., et al. 2002. Novel G protein-coupled receptor genes expressed in the brain: continued discovery of important therapeutic targets. *Expert Opin. Ther. Targets* 6: 185-202
2. Simonin, F., et al. 2004. Identification of a novel family of G protein-coupled receptor associated sorting proteins. *J. Neurochem.* 89: 766-775.
3. Horn, S.C., et al. 2006. Huntingtin interacts with the receptor sorting family protein GASP2. *J. Neural. Transm.* 113: 1081-1090.
4. Rozenfeld, R. and Devi, L.A. 2010. Exploring a role for heteromerization in GPCR signalling specificity. *Biochem. J.* 433: 11-18.
5. Costanzi, S. 2010. Modeling G protein-coupled receptors: a concrete possibility. *Chim. Oggi* 28: 26-31.
6. Borroto-Escuela, D.O., et al. 2011. Muscarinic receptor family interacting proteins: role in receptor function. *J. Neurosci. Methods* 195: 161-169.

CHROMOSOMAL LOCATION

Genetic locus: Gprasp2 (mouse) mapping to X F1.

PRODUCT

GASP-2 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 GASP-2 shRNA Plasmid (m): sc-145338-SH and GASP-2 shRNA (m) Lentiviral Particles: sc-145338-V as alternate gene silencing products.

For independent verification of GASP-2 (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-145338A, sc-145338B and sc-145338C.

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.

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

GASP-2 siRNA (m) is recommended for the inhibition of GASP-2 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 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

GASP-2 (F-6): sc-390664 is recommended as a control antibody for monitoring of GASP-2 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 GASP-2 gene expression knockdown using RT-PCR Primer: GASP-2 (m)-PR: sc-145338-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.