

GAP-43 siRNA (h): sc-35446

BACKGROUND

GAP-43 (growth associated protein 43, B-50, PP46, calmodulin-binding protein P-57, neuromodulin, neuron growth-associated protein 43, protein F1) is a crucial component for regenerative response in the nervous system. It is present at high levels in neuronal growth cones during development and axonal regeneration. GAP-43 is normally produced by neurons during developmental growth and axonal regeneration, but it is also expressed in specific regions of the normal adult nervous system. The neuron-specific Elav/Hu family member, HuD, interacts with and stabilizes GAP-43 mRNA in developing neurons and leads to increased levels of GAP-43 protein. Heterozygous GAP-43 knockout mice with GAP-43 levels reduced by one-half display significant memory impairments in cued conditioning or on tests of nociceptive or auditory perception.

REFERENCES

1. Deloulme, J.C., et al. 1990. Neuromodulin (GAP43): a neuronal protein kinase C substrate is also present in O-2A glial cell lineage. Characterization of neuromodulin in secondary cultures of oligodendrocytes and comparison with the neuronal antigen. *J. Cell Biol.* 111: 1559-1569.
2. Neve, R.L., et al. 1998. The neuronal growth-associated protein GAP-43 interacts with rabaptin-5 and participates in endocytosis. *J. Neurosci.* 18: 7757-7767.
3. Sretavan, D.W., et al. 1998. Randomized retinal ganglion cell axon routing at the optic chiasm of GAP-43 deficient mice: association with midline recrossing and lack of normal ipsilateral axon turning. *J. Neurosci.* 18: 10502-10513.
4. Arni, S., et al. 1998. Association of GAP-43 with detergent-resisting membranes requires two palmitoylated cysteine residues. *J. Biol. Chem.* 273: 28478-28485.
5. Dent, E.W., et al. 1998. Distribution of phosphorylated GAP-43 (neuromodulin) in growth cones directly reflects growth cone behavior. *J. Neurobiol.* 35: 287-299.
6. Velasco, A., et al. 2003. Role of oleic acid as a neurotrophic factor is supported *in vivo* by the expression of GAP-43 subsequent to the activation of SREBP-1 and the up-regulation of stearoyl-CoA desaturase during postnatal development of the brain. *Brain Res.* 977: 103-111.

CHROMOSOMAL LOCATION

Genetic locus: GAP43 (human) mapping to 3q13.31.

PRODUCT

GAP-43 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 GAP-43 shRNA Plasmid (h): sc-35446-SH and GAP-43 shRNA (h) Lentiviral Particles: sc-35446-V as alternate gene silencing products.

For independent verification of GAP-43 (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-35446A, sc-35446B and sc-35446C.

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

GAP-43 siRNA (h) is recommended for the inhibition of GAP-43 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

GAP-43 (B-5): sc-17790 is recommended as a control antibody for monitoring of GAP-43 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 GAP-43 gene expression knockdown using RT-PCR Primer: GAP-43 (h)-PR: sc-35446-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.

PROTOCOLS

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