

GRIP1 siRNA (h): sc-42160

BACKGROUND

Glutamate receptors mediate most excitatory neurotransmission in the brain and play an important role in neural plasticity, neural development and neurodegeneration. The glutamate receptor interacting proteins, GRIP1 and GRIP2, are members of the PDZ domain-containing protein family, and they specifically bind to the carboxy-terminus of AMPA receptor subunits, GluR-2 and GluR-3. GRIP1 and GRIP2 are involved in the targeting of GluR-2 and GluR-3 to the synapse. GRIP1 and GRIP2 are widely expressed in brain, with the highest levels in the cerebral cortex, hippocampus and olfactory bulb. They are both enriched in synaptic plasma and postsynaptic density fractions. GRIP1 is expressed in early development before the expression of AMPA receptors, specifically postnatal days 8-10, while GRIP2 expression parallels that of AMPA receptors during later developmental stages. GRIP1 and GRIP2 may mediate the endocytotic rate of GluR-2 and GluR-3 in response to the phosphorylation of the receptors on Ser 880 by PKC, which is implicated in the induction of cerebellar long-term depression (LTD).

REFERENCES

1. Choi, D.W., et al. 1990. The role of glutamate neurotoxicity in hypoxic-ischemic neuronal death. *Annu. Rev. Neurosci.* 13: 171-182.
2. Nakanishi, S. 1992. Molecular diversity of glutamate receptors and implications for brain function. *Science* 258: 597-603.
3. Wyszynski, M., et al. 1999. Association of AMPA receptors with a subset of glutamate receptor-interacting protein *in vivo*. *J. Neurosci.* 19: 6528-6537.
4. Dong, H., et al. 1999. Characterization of the glutamate receptor-interacting proteins GRIP1 and GRIP2. *J. Neurosci.* 19: 6930-6941.
5. Osten, P., et al. 2000. Mutagenesis reveals a role for ABP/GRIP binding to GluR2 in synaptic surface accumulation of the AMPA receptor. *Neuron* 27: 313-325.
6. Xia, J., et al. 2000. Cerebellar long-term depression requires PKC-regulated interactions between GluR2/3 and PDZ domain-containing proteins. *Neuron* 28: 499-510.
7. Matsuda, S., et al. 2000. Disruption of AMPA receptor GluR2 clusters following long-term depression induction in cerebellar Purkinje neurons. *EMBO J.* 19: 2765-2774.

CHROMOSOMAL LOCATION

Genetic locus: GRIP1 (human) mapping to 12q14.3.

PRODUCT

GRIP1 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 GRIP1 shRNA Plasmid (h): sc-42160-SH and GRIP1 shRNA (h) Lentiviral Particles: sc-42160-V as alternate gene silencing products.

For independent verification of GRIP1 (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-42160A, sc-42160B and sc-42160C.

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

GRIP1 siRNA (h) is recommended for the inhibition of GRIP1 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

GRIP1 (H-4): sc-365937 is recommended as a control antibody for monitoring of GRIP1 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 GRIP1 gene expression knockdown using RT-PCR Primer: GRIP1 (h)-PR: sc-42160-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.