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Homer-3 siRNA (m): sc-42193

BACKGROUND

Homer family proteins, also designated Vesl (for VASP/Ena-related gene upregulated during seizure) and LTP, are immediate early gene products that bind to group 1 metabotropic glutamate receptors (mGluRs), proteins involved in triggering intracellular calcium release. Unlike Homer-1a, the prototype member of the Homer family, other Homer family members (Homer-1b and -1c, Homer-2a, -2b and -2c and Homer-3) are constitutively expressed and contain a coiled-coil (CC) domain that mediates self-multimerization. Homer-1a is enriched at excitatory synapses, does not multimerize and appears to block the association of mGluRs to CC-Homer proteins. Homer proteins have also been shown to link mGluRs with the inositol triphosphate receptors (IP3R).

REFERENCES

1. Brakeman, P.R., et al. 1997. Homer: a protein that selectively binds metabotropic glutamate receptors. *Nature* 386: 284-288.
2. Kato, A., et al. 1997. Vesl, a gene encoding VASP/Ena family related protein, is upregulated during seizure, long-term potentiation and synaptogenesis. *FEBS Lett.* 412: 183-189.
3. Kato, A., et al. 1998. Novel members of the Vesl/Homer family of PDZ proteins that bind metabotropic glutamate receptors. *J. Biol. Chem.* 273: 23969-23975.
4. Xiao, B., et al. 1998. Homer regulates the association of group 1 metabotropic glutamate receptors with multivalent complexes of Homer-related, synaptic proteins. *Neuron* 21: 707-716.
5. Tu, J.C., et al. 1998. Homer binds a novel proline-rich motif and links group 1 metabotropic glutamate receptors with IP3 receptors. *Neuron* 21: 717-726.
6. Soloviev, M.M., et al. 2000. Molecular characterisation of two structurally distinct groups of human Homers, generated by extensive alternative splicing. *J. Mol. Biol.* 295: 1185-1200.
7. Soloviev, M.M., et al. 2000. Mouse brain and muscle tissues constitutively express high levels of Homer proteins. *Eur. J. Biochem.* 267: 634-639.
8. Ishiguro, K., et al. 2004. Homer-3 regulates activation of serum response element in T cells via its EVH1 domain. *Blood* 103: 2248-2256.

CHROMOSOMAL LOCATION

Genetic locus: Homer3 (mouse) mapping to 8 B3.3.

PRODUCT

Homer-3 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 Homer-3 shRNA Plasmid (m): sc-42193-SH and Homer-3 shRNA (m) Lentiviral Particles: sc-42193-V as alternate gene silencing products.

For independent verification of Homer-3 (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-42193A, sc-42193B and sc-42193C.

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

Homer-3 siRNA (m) is recommended for the inhibition of Homer-3 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

Homer-3 (E-2): sc-376154 is recommended as a control antibody for monitoring of Homer-3 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 Homer-3 gene expression knockdown using RT-PCR Primer: Homer-3 (m)-PR: sc-42193-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.