



HIP-55 siRNA (h): sc-75255

BACKGROUND

Drebrins (developmentally regulated brain proteins) are cytoplasmic proteins that bind F-Actin in the brain and are involved in cell migration, extension of neuronal processes and plasticity of dendrites. HIP-55 (HPK1-interacting protein of 55 kDa), also known as ABP1, SH3P7 or DBNL (Drebrin-like), is a 430 amino acid cytoplasmic protein that belongs to the ABP1 family. HIP-55 binds to F-Actin but is not involved in Actin polymerization, capping or bundling. In addition to containing an ADF-H domain, HIP-55 also consists of a SH3 domain, which mediates interaction with Shank 2, Shank 3 and PRAM-1. HIP-55 acts as an Actin-binding adapter protein and as a common effector of antigen receptor-signaling pathways in leukocytes. As a key component of the immunological synapse, HIP-55 regulates T-cell activation by bridging TCRs and the Actin cytoskeleton to gene activation and endocytic processes. HIP-55 is degraded by caspases during apoptosis.

REFERENCES

1. Chen, Y.R., et al. 2001. Caspase-mediated cleavage of Actin-binding and SH3-domain-containing proteins Cortactin, HS1, and HIP-55 during apoptosis. *Biochem. Biophys. Res. Commun.* 288: 981-989.
2. Kessels, M.M., et al. 2001. Mammalian Abp1, a signal-responsive F-Actin-binding protein, links the Actin cytoskeleton to endocytosis via the GTPase dynamin. *J. Cell Biol.* 153: 351-366.
3. Mise-Omata, S., et al. 2003. Mammalian Actin binding protein 1 is essential for endocytosis but not lamellipodia formation: functional analysis by RNA interference. *Biochem. Biophys. Res. Commun.* 301: 704-710.
4. Han, J., et al. 2003. The SH3 domain-containing adaptor HIP-55 mediates c-Jun N-terminal kinase activation in T cell receptor signaling. *J. Biol. Chem.* 278: 52195-52202.
5. Le Bras, S., et al. 2004. Recruitment of the Actin-binding protein HIP-55 to the immunological synapse regulates T cell receptor signaling and endocytosis. *J. Biol. Chem.* 279: 15550-15560.

CHROMOSOMAL LOCATION

Genetic locus: DBNL (human) mapping to 7p13.

PRODUCT

HIP-55 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 HIP-55 shRNA Plasmid (h): sc-75255-SH and HIP-55 shRNA (h) Lentiviral Particles: sc-75255-V as alternate gene silencing products.

For independent verification of HIP-55 (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-75255A, sc-75255B and sc-75255C.

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

HIP-55 siRNA (h) is recommended for the inhibition of HIP-55 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

HIP-55 (E-11): sc-398498 is recommended as a control antibody for monitoring of HIP-55 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 HIP-55 gene expression knockdown using RT-PCR Primer: HIP-55 (h)-PR: sc-75255-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.