

HIP1 siRNA (h): sc-41982

BACKGROUND

Huntington disease is associated with the expansion of a polyglutamine tract, greater than 35 repeats, in the HD gene product huntingtin. HIP1 (huntingtin-interacting protein 1), a membrane-associated protein, binds specifically to the N-terminus of human huntingtin. HIP1 is ubiquitously expressed in different brain regions at low levels, and exhibits nearly identical subcellular fractionation as huntingtin. The huntingtin-HIP1 interaction is restricted to the brain and is inversely correlated to the polyglutamine length in the huntingtin, suggesting that loss of normal huntingtin-HIP1 interaction may compromise the membrane-cytoskeletal integrity in the brain. HIP1 contains an endocytic multidomain protein with a C-terminal Actin-binding domain, a central coiled-coil forming region and an N-terminal ENTH domain. HIP1 may be involved in vesicle trafficking; the structural integrity of HIP1 is crucial for maintenance of normal vesicle size *in vivo*. HIP12 is a non-proapoptotic member of the HIP gene family that is expressed in the brain and shares a similar subcellular distribution pattern with HIP1. However, HIP12 differs from HIP1 in its pattern of expression at both the mRNA and protein level. HIP12 does not directly interact with huntingtin but can interact with HIP1.

REFERENCES

1. Kalchman, M.A., et al. 1997. HIP1, a human homologue of *S. cerevisiae* Sla2p, interacts with membrane-associated huntingtin in the brain. *Nat. Genet.* 16: 44-53.
2. Wanker, E.E., et al. 1997. HIP1: a huntingtin interacting protein isolated by the yeast two-hybrid system. *Hum. Mol. Genet.* 6: 487-495.
3. Wedemeyer, N., et al. 1997. Localization of the human HIP1 gene close to the elastin (ELN) locus on 7q11.23. *Genomics* 46: 313-315.
4. Himmelbauer, H., et al. 1998. IRS-PCR-based genetic mapping of the huntingtin interacting protein gene (HIP1) on mouse chromosome 5. *Mamm. Genome* 9: 26-31.
5. Chopra, V.S., et al. 2000. HIP12 is a non-proapoptotic member of a gene family including HIP1, an interacting protein with huntingtin. *Mamm. Genome* 11: 1006-1015.
6. Waelter, S., et al. 2001. The huntingtin interacting protein HIP1 is a clathrin and α -adaptin-binding protein involved in receptor-mediated endocytosis. *Hum. Mol. Genet.* 10: 1807-1817.

CHROMOSOMAL LOCATION

Genetic locus: HIP1 (human) mapping to 7q11.23.

PRODUCT

HIP1 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 HIP1 shRNA Plasmid (h): sc-41982-SH and HIP1 shRNA (h) Lentiviral Particles: sc-41982-V as alternate gene silencing products.

For independent verification of HIP1 (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-41982A, sc-41982B and sc-41982C.

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

HIP1 siRNA (h) is recommended for the inhibition of HIP1 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

HIP1 (4B10): sc-47754 is recommended as a control antibody for monitoring of HIP1 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 MarkerTM 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 HIP1 gene expression knockdown using RT-PCR Primer: HIP1 (h)-PR: sc-41982-PR (20 μ l, 572 bp). 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.