

eIF2D siRNA (h): sc-88042

BACKGROUND

Ligatin, also known as HCA56 (hepatocellular carcinoma-associated antigen 56) or LGTN, is a 584 amino acid peripheral membrane protein belonging to the ligatin family. Expressed during embryonic development and in early differentiated states, ligatin is a trafficking receptor for phosphoglycoproteins. Ligatin localizes to phosphoglycoproteins within endosomes and at the cell periphery where it participates in specific metabolic processes as well as intercellular adhesion. Involved in RNA binding and translation initiation through its single PUA domain and SUI1 domain, ligatin is down-regulated with long-lasting effects by the activation of Ca^{2+} dependent N-methyl-D-aspartate (NMDA) subclass of excitatory amino acid (EAA) receptors. Ligatin is considered a marker protein for membrane-vesicle transport systems. Ligatin exists as two alternatively splice variants and is encoded by a gene located on human chromosome 1.

REFERENCES

1. Marchase, R.B., et al. 1982. Retinal ligatin recognizes glycoproteins bearing oligosaccharides terminating in phosphodiester-linked glucose. *Cell* 28: 813-820.
2. Gaston, S.M., et al. 1982. Brain ligatin: a membrane lectin that binds acetylcholinesterase. *J. Cell. Biochem.* 18: 447-459.
3. Marchase, R.B., et al. 1982. A possible role for ligatin and the phosphoglycoproteins it binds in calcium-dependent retinal cell adhesion. *J. Cell. Biochem.* 18: 461-468.
4. Jakoi, E.R., et al. 1987. Ligatin: a peripheral membrane protein with covalently bound palmitic acid. *J. Biol. Chem.* 262: 1300-1304.
5. Jakoi, E.R., et al. 1989. Molecular cloning of the cDNA for ligatin. *J. Cell Sci.* 93: 227-232.
6. Jakoi, E.R., et al. 1992. Excitatory amino acid receptor activation produces a selective and long-lasting modulation of gene expression in hippocampal neurons. *Brain Res.* 582: 282-290.
7. Perlin, J.B., et al. 1993. Kindling produces long-lasting and selective changes in gene expression of hippocampal neurons. *Proc. Natl. Acad. Sci. USA* 90: 1741-1745.

CHROMOSOMAL LOCATION

Genetic locus: EIF2D (human) mapping to 1q32.1.

PRODUCT

eIF2D 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 μM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see eIF2D shRNA Plasmid (h): sc-88042-SH and eIF2D shRNA (h) Lentiviral Particles: sc-88042-V as alternate gene silencing products.

For independent verification of eIF2D (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-88042A, sc-88042B and sc-88042C.

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 μl of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μl of RNase-free water makes a 10 μM solution in a 10 μM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

eIF2D siRNA (h) is recommended for the inhibition of eIF2D 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 μM in 66 μl . 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

eIF2D (2D10): sc-293396 is recommended as a control antibody for monitoring of eIF2D 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 eIF2D gene expression knockdown using RT-PCR Primer: eIF2D (h)-PR: sc-88042-PR (20 μl). Annealing temperature for the primers should be $55-60^{\circ}\text{C}$ and the extension temperature should be $68-72^{\circ}\text{C}$.

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.