SANTA CRUZ BIOTECHNOLOGY, INC.

eIF4H siRNA (h): sc-89585

BACKGROUND

The initiation of protein synthesis in eukaryotic cells is regulated by interactions between protein initiation factors and RNA molecules. These interactions are facilitated, in part, by the eukaryotic initiation factor 4 family (eIF4) of proteins that are involved in the early initiation of protein synthesis. eIF4H (eukaryotic translation initiation factor 4H), also known as WSCR1 or WBSCR1, is a 248 amino acid protein that localizes to the perinuclear region of the cytoplasm and is expressed as two isoforms, designated short and long. While the short isoform is expressed predominately in liver and kidney, both isoforms are present in lung, pancreas, testis and spleen, where they function to stimulate RNA helicase activity. Specifically, eIF4H enhances the activity of eIF4A in the translation initiation complex, thereby promoting protein synthesis. Defects in the gene encoding eIF4H are associated with Williams-Beuren syndrome (WBS), a rare developmental disorder characterized by cardiovascular and musculo-skeletal abnormalities.

REFERENCES

1. Osborne, L.R., et al. 1996. Identification of genes from a 500 kb region at 7q11.23 that is commonly deleted in Williams syndrome patients. Genomics 36: 328-336.
2. Richter-Cook, N.J., et al. 1998. Purification and characterization of a new eukaryotic protein translation factor. Eukaryotic initiation factor 4H. J. Biol. Chem. 273: 7579-7587.
3. Bjork, P., et al. 2003. The Chironomus tentans translation initiation factor eIF4H is present in the nucleus but does not bind to mRNA until the mRNA reaches the cytoplasmic perinuclear region. J. Cell Sci. 116: 4521-4532.
4. Doepker, R.C., et al. 2004. Herpes simplex virus virion host shutoff protein is stimulated by translation initiation factors eIF4B and eIF4H. J. Virol. 78: 4684-4699.
5. Korneeva, N.L., et al. 2005. Interaction between the NH_2-terminal domain of eIF4A and the central domain of eIF4G modulates RNA-stimulated ATPase activity. J. Biol. Chem. 280: 1872-1881.
6. Feng, P., et al. 2005. mRNA decay during herpes simplex virus (HSV) infections: protein-protein interactions involving the HSV virion host shutoff protein and translation factors eIF4H and eIF4A. J. Virol. 79: 9651-9664.

CHROMOSOMAL LOCATION

Genetic locus: EIF4H (human) mapping to 7q11.23.

PRODUCT

eIF4H 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 eIF4H shRNA Plasmid (h): sc-89585-SH and eIF4H shRNA (h) Lentiviral Particles: sc-89585-V as alternate gene silencing products.

For independent verification of eIF4H (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-89585A, sc-89585B and sc-89585C.

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

eIF4H siRNA (h) is recommended for the inhibition of eIF4H 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

eIF4H (C-6): sc-515265 is recommended as a control antibody for monitoring of eIF4H 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 eIF4H gene expression knockdown using RT-PCR Primer: eIF4H (h)-PR: sc-89585-PR (20 µl). 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.