



eIF4B siRNA (m): sc-77254

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. eIF4B (eukaryotic translation initiation factor 4B) is a 611 amino acid protein that contains one RNA recognition motif and belongs to the eIF4 protein family. Required for proper mRNA/ribosome binding, eIF4B associates with other eIF4 proteins, such as eIF4A, and promotes the ATP-dependent unwinding activity of select eukaryotic initiation factors. The gene encoding eIF4B maps to human chromosome 12, which encodes over 1,100 genes and comprises approximately 4.5% of the human genome.

REFERENCES

1. Howe, J.G. and Hershey, J.W. 1984. Translational initiation factor and ribosome association with the cytoskeletal framework fraction from HeLa cells. *Cell* 37: 85-93.
2. Milburn, S.C., et al. 1990. Cloning and expression of eukaryotic initiation factor 4B cDNA: sequence determination identifies a common RNA recognition motif. *EMBO J.* 9: 2783-2790.
3. Naranda, T., et al. 1994. Two structural domains of initiation factor eIF4B are involved in binding to RNA. *J. Biol. Chem.* 269: 14465-14472.
4. Methot, N., et al. 1994. The translation initiation factor eIF4B contains an RNA-binding region that is distinct and independent from its ribonucleoprotein consensus sequence. *Mol. Cell. Biol.* 14: 2307-2316.
5. Methot, N., et al. 1996. A region rich in aspartic acid, arginine, tyrosine, and glycine (DRYG) mediates eukaryotic initiation factor 4B (eIF4B) self-association and interaction with eIF3. *Mol. Cell. Biol.* 16: 5328-5334.
6. Online Mendelian Inheritance in Man, OMIM™. 1999. Johns Hopkins University, Baltimore, MD. MIM Number: 603928. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>

CHROMOSOMAL LOCATION

Genetic locus: Eif4b (mouse) mapping to 15 F3.

PRODUCT

eIF4B 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 eIF4B shRNA Plasmid (m): sc-77254-SH and eIF4B shRNA (m) Lentiviral Particles: sc-77254-V as alternate gene silencing products.

For independent verification of eIF4B (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-77254A, sc-77254B and sc-77254C.

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

eIF4B siRNA (m) is recommended for the inhibition of eIF4B 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

eIF4B (D-4): sc-376062 is recommended as a control antibody for monitoring of eIF4B 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 eIF4B gene expression knockdown using RT-PCR Primer: eIF4B (m)-PR: sc-77254-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.