

eIF4AIII siRNA (m): sc-77395

BACKGROUND

The eukaryotic translation factor 4A (eIF4A) is a member of DEA(D/H)-box RNA helicase family that couples ATP hydrolysis to RNA binding and duplex separation. eIF4A participates in the initiation of translation by unwinding secondary structure in the 5'-untranslated region of mRNAs and facilitating scanning by the 40 S ribosomal subunit for the initiation codon. eIF4AIII is a component of the exon junction complex (EJC) that assembles near exon-exon junctions of mRNAs as a result of splicing. eIF4AIII, but not eIF4AI or eIF4AII, preferentially associates with spliced mRNA. eIF4AIII is found in the nucleus whereas eIF4AI and eIF4AII are found in the cytoplasm.

REFERENCES

1. Lomakin, I.B., et al. 2000. Physical association of eukaryotic initiation factor 4G (eIF4G) with eIF4A strongly enhances binding of eIF4G to the internal ribosomal entry site of encephalomyocarditis virus and is required for internal initiation of translation. *Mol. Cell. Biol.* 20: 6019-6029.
2. Dominguez, D., et al. 2001. Structural and functional similarities between the central eukaryotic initiation factor (eIF)4A-binding domain of mammalian eIF4G and the eIF4A-binding domain of yeast eIF4G. *Biochem. J.* 355: 223-230.
3. Rogers, G.W., Jr., et al. 2001. Further characterization of the helicase activity of eIF4A. Substrate specificity. *J. Biol. Chem.* 276: 12598-12608.
4. Li, W., et al. 2001. Eukaryotic initiation factors 4A (eIF4A) and 4G (eIF4G) mutually interact in a 1:1 ratio *in vivo*. *J. Biol. Chem.* 276: 29111-29115.
5. Rogers, G.W., Jr., et al. 2001. Modulation of the helicase activity of eIF4A by eIF4B, eIF4H, and eIF4F. *J. Biol. Chem.* 276: 30914-30922.
6. Montero-Lomeli, M., et al. 2002. The initiation factor eIF4A is involved in the response to lithium stress in *Saccharomyces cerevisiae*. *J. Biol. Chem.* 277: 21542-21548.
7. Goke, A., et al. 2002. DUG is a novel homologue of translation initiation factor 4G that binds eIF4A. *Biochem. Biophys. Res. Commun.* 297: 78-82.

CHROMOSOMAL LOCATION

Genetic locus: Eif4a3 (mouse) mapping to 11 E2.

PRODUCT

eIF4AIII siRNA (m) is a pool of 2 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 eIF4AIII shRNA Plasmid (m): sc-77395-SH and eIF4AIII shRNA (m) Lentiviral Particles: sc-77395-V as alternate gene silencing products.

For independent verification of eIF4AIII (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-77395A and sc-77395B.

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

eIF4AIII siRNA (m) is recommended for the inhibition of eIF4AIII 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

eIF4AIII (B-2): sc-365549 is recommended as a control antibody for monitoring of eIF4AIII 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 eIF4AIII gene expression knockdown using RT-PCR Primer: eIF4AIII (m)-PR: sc-77395-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.