SANTA CRUZ BIOTECHNOLOGY, INC.

eIF2β siRNA (h): sc-35270

BACKGROUND

The initiation of protein synthesis in eukaryotic cells is regulated by interactions between protein initiation factors and RNA molecules. The eukaryotic initiation complex eIF2B exists as a five subunit complex composed of eIF2Bα, eIF2Bβ, eIF2Bγ, eIF2Bδ, and eIF2Bε. The eIF2B complex catalyzes the exchange of GDP for GTP on the eIF2 complex, following the interaction of eIF2/GTP with the 40S ribosomal subunit. Guanine nucleotide exchange factor (GEF) activity is exhibited by the eIF2Bε subunit alone, but is greater in the presence of all five eIF2B subunits. Phosphorylation of eIF2 inhibits GEF activity of eIF2B, an inhibition that requires the eIF2Bα subunit.

REFERENCES

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4. Pathak, V.K., Nielsen, P.J., Trachsel, H. and Hershey, J.W. 1988. Structure of the β subunit of translational initiation factor eIF2. Cell 54: 633-639.
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6. Gaspar, N.J., Kinzy, T.G., Scherer, B.J., Humbelin, M., Hershey, J.W. and Merrick, W.C. 1994. Translation initiation factor eIF2. Cloning and expression of the human cDNA encoding the γ-subunit. J. Biol. Chem. 269: 3415-3422.

CHROMOSOMAL LOCATION

Genetic locus: EIF2S2 (human) mapping to 20q11.22.

PRODUCT

eIF2β 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 eIF2β shRNA Plasmid (h): sc-35270-SH and eIF2β shRNA (h) Lentiviral Particles: sc-35270-V as alternate gene silencing products.

For independent verification of eIF2β (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-35270A, sc-35270B and sc-35270C.

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 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

eIF2β siRNA (h) is recommended for the inhibition of eIF2β 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

eIF2β (P-3): sc-9978 is recommended as a control antibody for monitoring of eIF2β 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 eIF2β gene expression knockdown using RT-PCR Primer: eIF2β (h)-PR: sc-35270-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.