# eIF2B $\delta$ siRNA (h): sc-35276



The Power to Question

## **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 eIF2B $\alpha$ , eIF2B $\beta$ , eIF2B $\beta$ , eIF2B $\beta$ , and eIF2B $\epsilon$ . 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 eIF2B $\epsilon$  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 eIF2B $\alpha$  subunit.

## **REFERENCES**

- 1. Henderson, R.A., et al. 1994. The  $\delta$ -subunit of murine guanine nucleotide exchange factor eIF-2B. Characterization of cDNAs predicts isoforms differing at the amino-terminal end. J. Biol. Chem. 269: 30517-30523.
- 2. Flowers, K.M., et al. 1995. Structure and sequence of the gene encoding the  $\alpha$ -subunit of rat translation initiation factor-2B. Biochim. Biophys. Acta 1264: 163-167.
- Price, N.T., et al. 1996. Cloning of cDNA for the γ-subunit of mammalian translation initiation factor 2B, the guanine nucleotide-exchange factor for eukaryotic initiation factor 2. Biochem. J. 318: 631-636.
- 4. Price, N.T., et al. 1996. eIF2B, the guanine nucleotide-exchange factor for eukaryotic initiation factor 2. Sequence conservation between the  $\alpha$ ,  $\beta$  and  $\delta$  subunits of eIF2B from mammals and yeast. Biochem. J. 318: 637-643.
- Asuru, A.I., et al. 1996. Cloning and characterization of cDNAs encoding the ε-subunit of eukaryotic initiation factor-2B from rabbit and human. Biochim. Biophys. Acta 1307: 309-317.
- 6. Webb, B.L. and Proud, C.G. 1997. Eukaryotic initiation factor 2B (eIF2B). Int. J. Biochem. Cell Biol. 29: 1127-1131.
- Fabian, J.R., et al. 1997. Subunit assembly and guanine nucleotide exchange activity of eukaryotic initiation factor-2B expressed in Sf9 cells. J. Biol. Chem. 272: 12359-12365.

## CHROMOSOMAL LOCATION

Genetic locus: EIF2B4 (human) mapping to 2p23.3.

## **PRODUCT**

elF2B $\delta$  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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see elF2B $\delta$  shRNA Plasmid (h): sc-35276-SH and elF2B $\delta$  shRNA (h) Lentiviral Particles: sc-35276-V as alternate gene silencing products.

For independent verification of elF2B $\delta$  (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-35276A, sc-35276B and sc-35276C.

## **RESEARCH USE**

For research use only, not for use in diagnostic procedures.

### 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  $\mu$ l of the RNAse-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNAse-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## **APPLICATIONS**

eIF2B $\delta$  siRNA (h) is recommended for the inhibition of eIF2B $\delta$  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**

eIF2B\(\delta\) (P-6): sc-9981 is recommended as a control antibody for monitoring of eIF2B\(\delta\) 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-lgG $\kappa$  BP-HRP: sc-516102 or m-lgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>TM</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-lgG $\kappa$  BP-FITC: sc-516140 or m-lgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

## **RT-PCR REAGENTS**

Semi-quantitative RT-PCR may be performed to monitor eIF2B $\delta$  gene expression knockdown using RT-PCR Primer: eIF2B $\delta$  (h)-PR: sc-35276-PR (20  $\mu$ I). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

#### **PROTOCOLS**

See our web site at www.scbt.com for detailed protocols and support products.

**Santa Cruz Biotechnology, Inc.** 1.800.457.3801 831.457.3801 **Europe** +00800 4573 8000 49 6221 4503 0 **www.scbt.com**