SANTA CRUZ BIOTECHNOLOGY, INC.

eIF3 p110 siRNA (m): sc-40546

BACKGROUND

Translation initiation in eukaryotes necessitates the assembly of an 80S ribosomal complex containing methionyl initiator tRNA (Met-tRNAiMet), which is base paired at the initiation codon (AUG, GUG) in eligible transcripts. Eukaryotic initiation factors (eIFs) are utilized in a sequence of reactions that leads to 80S ribosomal assembly and initiation of translation. Eukaryotic initiation factor-3 (eIF3) is the largest family of eIFs and consists of at least 10 unique subunits (p170, p116, p110, p66, p48, p47, p44, p40, p36, and p35) in mammals. eIF3 subunit-9 (eIF3-h, eIF3-p116, p116, eIF3-S9, PRT1) is a 873 amino acid component of the eIF3 multi-
subunit complex that is involved in ribosomal 48S complex formation. Association of the eIF3 complex with the 40S ribosomal subunit stabilizes eIF2-GTP-Met-tRNAiMet complex association and mRNA binding, and promotes dissociation of 80S ribosomes into 40S and 60S subunits.

REFERENCES

1. Asano, K., et al 1997. Conservation and diversity of eukaryotic translation initiation factor eIF3. J. Biol. Chem. 272: 1101-1109.
2. Chaudhuri, J., et al. 1999. Distinct functions of eukaryotic translation initiation factors eIF1A and eIF3 in the formation of the 40 S ribosomal preinitiation complex. J. Biol. Chem. 274: 17975-17980.
3. Online Mendelian Inheritance in Man, OMIM™. 1999. Johns Hopkins University, Baltimore, MD. MIM Number: 603916. World Wide Web URL: http://www.ncbi.nlm.nih. gov/omim/
4. Korneeva, N.L., et al. 2000. Mutually cooperative binding of eukaryotic translation initiation factor (eIF) 3 and eIF4A to human eIF4G-1. J. Biol. Chem. 275: 41369-41376.
5. Asano, K., et al. 2000. A multifactor complex of eukaryotic initiation factors, eIF1, eIF2, eIF3, eIF5, and initiator tRNA(Met) is an important translation initiation intermediate *in vivo*. Genes Dev. 14: 2534-2546.

CHROMOSOMAL LOCATION

Genetic locus: Eif3s8 (mouse) mapping to 7.

PRODUCT

eIF3 p110 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see eIF3 p110 shRNA Plasmid (m): sc-40546-SH and eIF3 p110 shRNA (m) Lentiviral Particles: sc-40546-V as alternate gene silencing products.

For independent verification of eIF3 p110 (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-40546A, sc-40546B and sc-40546C.

PROTOCOLS

See our web site at www.scbt.com or our catalog 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

eIF3 p110 siRNA (m) is recommended for the inhibition of eIF3 p100 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 µM in 60 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

eIF3 p110 (B-6): sc-74507 is recommended as a control antibody for monitoring of eIF3 p110 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor eIF3 p100 gene expression knockdown using RT-PCR Primer: eIF3 p110 (m)-PR: sc-40546-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.