

POLR2E siRNA (m): sc-152372

BACKGROUND

RNA polymerase II (Pol II) is a multi-subunit complex responsible for catalyzing the transcription of DNA into RNA. POLR2E (polymerase (RNA) II (DNA directed) polypeptide E), also designated RPB5, XAP4, RPABC1, hRPB25 or hsRPB5, is a 210 amino acid nuclear protein belonging to the archaeal rpoH/ eukaryotic RPB5 RNA polymerase subunit family. POLR2E is a DNA-dependent RNA polymerase that catalyzes the transcription of DNA into RNA using the four ribonucleoside triphosphates as substrates. POLR2E is a component of the RNA polymerase I (Pol I), RNA polymerase II (Pol II) and RNA polymerase III (Pol III) complexes. The Pol complexes synthesize both mRNA and ribosomal RNA precursors, many functional non-coding RNAs and small RNAs including 5S rRNA and tRNAs. POLR2E interacts with TFIIF RAP 30 and is important for the association between Pol II and TFIIF.

REFERENCES

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2. Kershner, E., et al. 1998. Immunoaffinity purification and functional characterization of human transcription factor IIH and RNA polymerase II from clonal cell lines that conditionally express epitope-tagged subunits of the multiprotein complexes. *J. Biol. Chem.* 273: 34444-34453.
3. Wei, W., et al. 2001. Direct interaction between the subunit RAP30 of transcription factor IIF (TFIIF) and RNA polymerase subunit 5, which contributes to the association between TFIIF and RNA polymerase II. *J. Biol. Chem.* 276: 12266-12273.
4. Delgermaa, L., et al. 2004. Subcellular localization of RPB5-mediating protein and its putative functional partner. *Mol. Cell. Biol.* 24: 8556-8566.
5. Le, T.T., et al. 2005. Mutational analysis of human RNA polymerase II subunit 5 (RPB5): the residues critical for interactions with TFIIF subunit RAP30 and hepatitis B virus X protein. *J. Biochem.* 138: 215-224.
6. Panov, K.I., et al. 2006. RNA polymerase I-specific subunit CAST/hPAF49 has a role in the activation of transcription by upstream binding factor. *Mol. Cell. Biol.* 26: 5436-5448.

CHROMOSOMAL LOCATION

Genetic locus: Polr2e (mouse) mapping to 10 C1.

PRODUCT

POLR2E 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 POLR2E shRNA Plasmid (m): sc-152372-SH and POLR2E shRNA (m) Lentiviral Particles: sc-152372-V as alternate gene silencing products.

For independent verification of POLR2E (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-152372A and sc-152372B.

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

POLR2E siRNA (m) is recommended for the inhibition of POLR2E 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

POLR2E (B-5): sc-390979 is recommended as a control antibody for monitoring of POLR2E 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 POLR2E gene expression knockdown using RT-PCR Primer: POLR2E (m)-PR: sc-152372-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.

PROTOCOLS

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