

TPX2 siRNA (m): sc-37654

BACKGROUND

TPX2 (targeting protein for Xklp2) is a microtubule-associated protein involved in targeting the motor protein Xklp2 to microtubules. Ran-GTP activates TPX2 for the chromatin-induced microtubule assembly during M phase. Aurora-A kinase associates with TPX2 at the spindle apparatus and may regulate TPX2 via phosphorylation during the spindle assembly. TPX2 appears to play a structural role in spindle formation. TPX2 activates Eg2 in a microtubule-dependent manner by stimulating the phosphorylation and kinase activity of Eg2. TPX2 is inactivated by binding to importin α , a nuclear import factor. Finally, the suppression of TPX2 with RNA interference causes defects in microtubule organization during mitosis.

REFERENCES

1. Wittmann, T., et al. 1998. Localization of the kinesin-like protein Xklp2 to spindle poles requires a leucine zipper, a microtubule-associated protein, and dynein. *J. Cell Biol.* 143: 673-685.
2. Gruss, O.J., et al. 2001. Ran induces spindle assembly by reversing the inhibitory effect of importin α on TPX2 activity. *Cell* 104: 83-93.
3. Kufer, T.A., et al. 2002. Human TPX2 is required for targeting Aurora-A kinase to the spindle. *J. Cell Biol.* 158: 617-623.
4. Garrett, S., et al. 2002. hTPX2 is required for normal spindle morphology and centrosome integrity during vertebrate cell division. *Curr. Biol.* 12: 2055-2059.
5. Gruss, O.J., et al. 2002. Chromosome-induced microtubule assembly mediated by TPX2 is required for spindle formation in HeLa cells. *Nat. Cell Biol.* 4: 871-879.
6. Tsai, M.Y., et al. 2003. A Ran signalling pathway mediated by the mitotic kinase Aurora A in spindle assembly. *Nat. Cell Biol.* 5: 242-248.

CHROMOSOMAL LOCATION

Genetic locus: Tpx2 (mouse) mapping to 2 H1.

PRODUCT

TPX2 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see TPX2 shRNA Plasmid (m): sc-37654-SH and TPX2 shRNA (m) Lentiviral Particles: sc-37654-V as alternate gene silencing products.

For independent verification of TPX2 (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-37654A, sc-37654B and sc-37654C.

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.

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

TPX2 siRNA (m) is recommended for the inhibition of TPX2 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

TPX2 (B-5): sc-376812 is recommended as a control antibody for monitoring of TPX2 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 TPX2 gene expression knockdown using RT-PCR Primer: TPX2 (m)-PR: sc-37654-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.