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SANTA CRUZ BIOTECHNOLOGY, INC.

NPAT siRNA (m): sc-44782

BACKGROUND

The nuclear protein ataxia telangiectasia locus (NPAT), an essential downstream component of the cyclin E/Cdk2 signaling pathway, acts as a critical regulator for S phase entry, histone gene expression and Cajal body maintenance in somatic cells. This protein was originally identified by its chromosomal location, 11q23, and its proximity to the ATM gene, which is responsible for the autosomal recessive disease ataxia telangiectasia (AT). The NPAT protein sequence is strongly conserved in eukaryotes and its expression is ubiquitous. The C-terminal half of the NPAT protein contains multiple elements required for induction of S phase, while the N-terminal half appears to be crucial for the activation of Histone H4 and H2B. NPAT contains several Cdk2 phosphorylation sites, but they do not appear to affect protein function.

REFERENCES

1. Imai, T., et al. 1996. Identification and characterization of a new gene physically linked to the ATM gene. Genome Res. 6: 439-447.
2. Ma, T., et al. 2000. Cell cycle-regulated phosphorylation of $p220^{NPAT}$ by cyclin E/Cdk2 in Cajal bodies promotes histone gene transcription. Genes Dev. 14: 2298-2313.
3. Sagara, M., et al. 2002. Characterization of functional regions for nuclear localization of NPAT. J. Biochem. 132: 875-879.
4. Gao, G., et al. 2003. NPAT expression is regulated by E2F and is essential for cell cycle progression. Mol. Cell. Biol. 23: 2821-2833.
5. Wei, Y., et al. 2003. The cyclin E/Cdk2 substrate and Cajal body component $p220^{NPAT}$ activates histone transcription through a novel LisH-like domain. Mol. Cell. Biol. 23: 3669-3680.
6. Wang, A., et al. 2004. Dynamic interaction of $p220^{NPAT}$ and CBP/p300 promotes S phase entry. Biochem. Biophys. Res. Commun. 325: 1509-1516.
7. Miele, A., et al. 2005. HiNF-P directly links the cyclin E/Cdk2/ $p220^{NPAT}$ pathway to Histone H4 gene regulation at the G_1/S phase cell cycle transition. Mol. Cell. Biol. 25: 6140-6153.

CHROMOSOMAL LOCATION

Genetic locus: Npat (mouse) mapping to 9 A5.3.

PRODUCT

NPAT 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 NPAT shRNA Plasmid (m): sc-44782-SH and NPAT shRNA (m) Lentiviral Particles: sc-44782-V as alternate gene silencing products.

For independent verification of NPAT (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-44782A, sc-44782B and sc-44782C.

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

NPAT siRNA (m) is recommended for the inhibition of NPAT 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 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor NPAT gene expression knockdown using RT-PCR Primer: NPAT (m)-PR: sc-44782-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.