

Pan3 siRNA (h): sc-76039

BACKGROUND

Pan3 (PAB-dependent poly(A)-specific ribonuclease subunit 3) is a 741 amino acid cytoplasmic protein belonging to the protein kinase superfamily. Containing a protein kinase domain, Pan3 is a component of the Pan nuclease complex and recruits polyadenylate-binding protein, which in turn stimulates Pan2 nuclease activity. It is suggested that Pan3 may have a functional role in cytoplasmic mRNA decay. Pan3 exists as three isoforms produced by alternative splicing and the gene encoding Pan3 is located on human chromosome 13. Chromosome 13 houses over 400 genes, such as BRCA2 and RB1, and comprises nearly 4% of the human genome. Trisomy 13, also known as Patau syndrome, is deadly and the few who survive past one year suffer from permanent neurologic defects, difficulty eating and vulnerability to serious respiratory infections.

REFERENCES

1. Brown, C.E., et al. 1996. Pan3 encodes a subunit of the Pab1p-dependent poly(A) nuclease in *Saccharomyces cerevisiae*. *Mol. Cell. Biol.* 16: 5744-5753.
2. Hammet, A., et al. 2002. Posttranscriptional regulation of the Rad5 DNA repair gene by the Dun1 kinase and the Pan2-Pan3 poly(A)-nuclease complex contributes to survival of replication blocks. *J. Biol. Chem.* 277: 22469-22474.
3. Uchida, N., et al. 2004. Identification of a human cytoplasmic poly(A) nuclease complex stimulated by poly(A)-binding protein. *J. Biol. Chem.* 279: 1383-1391.
4. Funakoshi, Y., et al. 2007. Mechanism of mRNA deadenylation: evidence for a molecular interplay between translation termination factor eRF3 and mRNA deadenylases. *Genes Dev.* 21: 3135-3148.
5. Siddiqui, N., et al. 2007. Poly(A) nuclease interacts with the C-terminal domain of polyadenylate-binding protein domain from poly(A)-binding protein. *J. Biol. Chem.* 282: 25067-25075.

CHROMOSOMAL LOCATION

Genetic locus: PAN3 (human) mapping to 13q12.2.

PRODUCT

Pan3 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Pan3 shRNA Plasmid (h): sc-76039-SH and Pan3 shRNA (h) Lentiviral Particles: sc-76039-V as alternate gene silencing products.

For independent verification of Pan3 (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-76039A, sc-76039B and sc-76039C.

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

Pan3 siRNA (h) is recommended for the inhibition of Pan3 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 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

Pan3 (A-9): sc-376434 is recommended as a control antibody for monitoring of Pan3 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 Pan3 gene expression knockdown using RT-PCR Primer: Pan3 (h)-PR: sc-76039-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.