

## Pin1 siRNA (m): sc-36231

### BACKGROUND

NIMA was originally shown in *Aspergillus nidulans* to be necessary for entry into mitosis. NIMA-related mammalian proteins have since been identified as Nek1, Nek2 and Nek3. High expression of Nek1 is seen in male and female germ cell lines of mouse. Nek2 is the closest known mammalian relative to NIMA. Like NIMA, Nek2 expression peaks at the G<sub>2</sub> to M phase transition. Pin1 was originally identified as a NIMA-interacting protein. Pin1 is a peptidyl-prolyl *cis/trans* isomerase (PPlase), which specifically binds to phosphoserine-proline or phosphothreonine-proline bonds in mitotic phosphoproteins. While previously identified PPlases have been shown to be involved in protein folding, assembly and transport, Pin1 is the first PPlase to be identified as a required protein for cell viability.

### REFERENCES

- Osmani, S.A., et al. 1988. Mitotic induction and maintenance by overexpression of a G<sub>2</sub>-specific gene that encodes a potential protein kinase. *Cell* 53: 237-244.
- Letwin, K., et al. 1992. A mammalian dual specificity protein kinase, Nek1, is related to the NIMA cell cycle regulator and highly expressed in meiotic germ cells. *EMBO J.* 11: 3521-3531.
- Schultz, S.J., et al. 1994. Cell cycle-dependent expression of Nek2, a novel human protein kinase related to the NIMA mitotic regulator of *Aspergillus nidulans*. *Cell Growth Differ.* 5: 625-635.
- Lu, K.P., et al. 1996. A human peptidyl-prolyl isomerase essential for regulation of mitosis. *Nature* 380: 544-547.
- Yaffe, M.B., et al. 1997. Sequence-specific and phosphorylation-dependent proline isomerization: a potential mitotic regulatory mechanism. *Science* 278: 1957-1960.
- Ranganathan, R., et al. 1997. Structural and functional analysis of the mitotic rotamase Pin1 suggests substrate recognition is phosphorylation dependent. *Cell* 89: 875-886.
- Rhee, K., et al. 1997. The NIMA-related kinase 2, Nek2, is expressed in specific stages of the meiotic cell cycle and associates with meiotic chromosomes. *Development* 124: 2167-2177.

### CHROMOSOMAL LOCATION

Genetic locus: Pin1 (mouse) mapping to 9 A3.

### PRODUCT

Pin1 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Pin1 shRNA Plasmid (m): sc-36231-SH and Pin1 shRNA (m) Lentiviral Particles: sc-36231-V as alternate gene silencing products.

For independent verification of Pin1 (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-36231A, sc-36231B and sc-36231C.

### 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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

### APPLICATIONS

Pin1 siRNA (m) is recommended for the inhibition of Pin1 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  $\mu$ M in 66  $\mu$ 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

Pin1 (G-8): sc-46660 is recommended as a control antibody for monitoring of Pin1 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 $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

### RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Pin1 gene expression knockdown using RT-PCR Primer: Pin1 (m)-PR: sc-36231-PR (20  $\mu$ l, 583 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

### SELECT PRODUCT CITATIONS

- Baik, S.H., et al. 2015. Pin1 promotes neuronal death in stroke by stabilizing Notch intracellular domain. *Ann. Neurol.* 77: 504-516.

### RESEARCH USE

For research use only, not for use in diagnostic procedures.