SANTA CRUZ BIOTECHNOLOGY, INC.

Pellino 3 siRNA (m): sc-44941

BACKGROUND

Mammalian Pellino proteins 1-3 (Pellino *Drosophila* homolog 1-3) are scaffolding components within Toll-like receptor (TLR) and interleukin-1 (IL-1) receptor signaling cascades. Pellino 1 and 3 interact with complexes that also contain IL-1R-associated kinase 4 (IRAK-4) and tumor necrosis factor receptor-associated factor-6 (TRAF6). Pellino 1, 2 and 3 interact with TAK1 (TGFβ activated kinase 1). Pellino 2 can initiate mitogen activated protein kinase pathways leading to activation of AP-1 and Elk-1. Pellino 3 promotes translocation of MAPK-activated protein kinase 2 from the nucleus to the cytoplasm and activates transcription factor CREB in a p38 MAPK-dependent manner. Pellino 3 physically interacts with NFκB-inducing kinase (NIK) in an IL-1-dependent manner and leads to activation of c-Jun, Elk-1 and c-Jun N-terminal kinase.

REFERENCES

1. Rich, T., et al. 2000. Pellino-related sequences from *Caenorhabditis elegans* and *Homo sapiens*. Immunogenetics 52: 145-149.
2. Resch, K., et al. 2001. Assignment of homologous genes, Peli1/PELI1 and Peli2/PELI2, for the Pelle adaptor protein Pellino to mouse chromosomes 11 and 14 and human chromosomes 2p13.3 and 14q21, respectively, by physical and radiation hybrid mapping. Cytogenet. Cell Genet. 92: 172-174.
3. Yu, K.Y., et al. 2002. Cutting edge: mouse Pellino 2 modulates IL-1 and lipopolysaccharide signaling. J. Immunol. 169: 4075-4078.
4. Jiang, Z., et al. 2003. Pellino 1 is required for interleukin-1 (IL-1)-mediated signaling through its interaction with the IL-1 receptor-associated kinase 4 (IRAK-4)-IRAK-tumor necrosis factor receptor-associated factor-6 (TRAF6) complex. J. Biol. Chem. 278: 10952-10956.
5. Jensen, L.E., et al. 2003. Pellino 2 activates the mitogen activated protein kinase pathway. FEBS Lett. 545: 199-202.
6. Jensen, L.E., et al. 2003. Pellino 3, a novel member of the Pellino protein family, promotes activation of c-Jun and Elk-1 and may act as a scaffolding protein. J. Immunol. 171: 1500-1506.
7. Butler, M.P., et al. 2005. Pellino 3 is a novel upstream regulator of p38 MAPK and activates CREB in a p38-dependent manner. J. Biol. Chem. 280: 27759-27768.

CHROMOSOMAL LOCATION

Genetic locus: Peli3 (mouse) mapping to 19 A.

PRODUCT

Pellino 3 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 Pellino 3 shRNA Plasmid (m): sc-44941-SH and Pellino 3 shRNA (m) Lentiviral Particles: sc-44941-V as alternate gene silencing products.

For independent verification of Pellino 3 (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-44941A, sc-44941B and sc-44941C.

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

Pellino 3 siRNA (m) is recommended for the inhibition of Pellino 3 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.

GENE EXPRESSION MONITORING

Pellino 3 (B-3): sc-376466 is recommended as a control antibody for monitoring of Pellino 3 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 Pellino 3 gene expression knockdown using RT-PCR Primer: Pellino 3 (m)-PR: sc-44941-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.

PROTOCOLS

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