TBK1 siRNA (h): sc-39058



The Power to Question

BACKGROUND

The transcription factor NF κ B is retained in the cytoplasm in an inactive form by the inhibitory protein I κ B. Activation of NF κ B requires that I κ B be phosphorylated on specific serine residues, which results in targeted degradation of I κ B. I κ B kinase α (IKK α), previously designated CHUK, interacts with I κ B- α and specifically phosphorylates I κ B- α on the sites that trigger its degradation, Serines 32 and 36. The functional IKK complex contains three subunits, IKK α , IKK β and IKK γ (also designated NEMO), and each appear to make essential contributions to I κ B phosphorylation. TANK binding kinase (TBK1), also designated T2K, is a novel IKK-related kinase that has been identified in murine and human tissues. TBK1 was shown to complex with TRAF2 and TANK in the NF κ B activation pathway. TBK1 shares homology with IKK α and IKK β in the amino-terminal half, which includes the kinase domain.

CHROMOSOMAL LOCATION

Genetic locus: TBK1 (human) mapping to 12q14.2.

PRODUCT

TBK1 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 μM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see TBK1 shRNA Plasmid (h): sc-39058-SH and TBK1 shRNA (h) Lentiviral Particles: sc-39058-V as alternate gene silencing products.

For independent verification of TBK1 (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-39058A, sc-39058B and sc-39058C.

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

TBK1 siRNA (h) is recommended for the inhibition of TBK1 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 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

TBK1 (A-6): sc-398366 is recommended as a control antibody for monitoring of TBK1 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-lgG κ BP-HRP: sc-516102 or m-lgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-lgG κ BP-FITC: sc-516140 or m-lgG 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 TBK1 gene expression knockdown using RT-PCR Primer: TBK1 (h)-PR: sc-39058-PR (20 μ l, 410 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

- Lee, S.H., et al. 2009. The major outer membrane protein of a periodontopathogen induces IFN-β and IFN-stimulated genes in monocytes via lipid raft and TANK-binding kinase 1/IFN regulatory factor-3. J. Immunol. 182: 5823-5835.
- 2. Fischer, H., et al. 2010. Pathogen specific, IRF3-dependent signaling and innate resistance to human kidney infection. PLoS Pathog. 6: e1001109.
- Moore, A.S. and Holzbaur, E.L. 2016. Dynamic recruitment and activation of ALS-associated TBK1 with its target optineurin are required for efficient mitophagy. Proc. Natl. Acad. Sci. USA 113: E3349-E3358.
- Davis, S.E., et al. 2019. Nucleosomal dsDNA stimulates APOL1 expression in human cultured podocytes by activating the cGAS/IFI16-STING signaling pathway. Sci. Rep. 9: 15485.
- Yang, H., et al. 2020. The GTPase KRAS suppresses the P53 tumor suppressor by activating the Nrf2-regulated antioxidant defense system in cancer cells. J. Biol. Chem. 295: 3055-3063.
- MIcochova, P., et al. 2020. TLR4-mediated pathway triggers interferonindependent G₀ arrest and antiviral SAMHD1 activity in macrophages. Cell Rep. 30: 3972-3980.e5.
- Sawaged, S., et al. 2022. TBK1 and GABARAP family members suppress coxsackievirus B infection by limiting viral production and promoting autophagic degradation of viral extracellular vesicles. PLoS Pathog. 18: e1010350.
- 8. Uchida, T., et al. 2023. Promotion of knee cartilage degradation by $I\kappa B$ kinase ϵ in the pathogenesis of osteoarthritis in human and murine models. Arthritis Rheumatol. 75: 937-949.

RESEARCH USE

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