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SANTA CRUZ BIOTECHNOLOGY, INC.

NIK siRNA (h2): sc-44314

BACKGROUND

The NFκB transcription factor can be activated by several cytokines including TNF and IL-1. The TNF receptor activates NFκB through the TRAF2 adapter protein, whereas the IL-1 receptor activates NFκB in a pathway involving TRAF6. Both TRAF2 and TRAF6 have been shown to interact with a Serine/Threonine kinase designated NFκB inducing kinase (NIK), which appears to participate in the NFκB signaling cascades triggered by both TNF and IL-1. NIK associates with, and is a costimulator for IκB kinase a (IKKα). IKKα, in turn, phosphorylates IκB, resulting in IκB degradation and NFκB activation. NIK has sequence similarity to several kinases that participate in MAP kinase cascades. NIK appears to be uninvolved in the TRAF2-mediated activation of JNK by TNF.

REFERENCES

1. Rothe, M., et al. 1995. TRAF2-mediated activation of NFκB by TNF receptor 2 and CD40. Science 269: 1424-1427.
2. Hsu, H., et al. 1996. TRADD-TRAF2 and TRADD-FADD interactions define two distinct TNF receptor 1 signal transduction pathways. Cell 84: 299-308.
3. Cao, Z., et al. 1996. TRAF6 is a signal transducer for interleukin-1. Nature 383: 443-446.
4. Malinin, N.L., et al. 1997. MAP3K-related kinase involved in NFκB induction by TNF, CD95 and IL-1. Nature 385: 540-544.
5. Song, H.Y., et al. 1997. Tumor necrosis factor (TNF)-mediated kinase cascades: bifurcation of nuclear factor-κB and c-Jun N-terminal kinase (JNK/SAPK) pathways at TNF receptor-associated factor 2. Proc. Nat. Acad. Sci. USA 94: 9792-9796..

CHROMOSOMAL LOCATION

Genetic locus: MAP3K14 (human) mapping to 17q21.31.

PRODUCT

NIK siRNA (h2) 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 NIK shRNA Plasmid (h2): sc-44314-SH and NIK shRNA (h2) Lentiviral Particles: sc-44314-V as alternate gene silencing products.

For independent verification of NIK (h2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3 nmol of lyophilized siRNA. These include: sc-44314A, sc-44314B and sc-44314C.

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

NIK siRNA (h2) is recommended for the inhibition of NIK 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

NIK (A-12): sc-8417 is recommended as a control antibody for monitoring of NIK 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor NIK gene expression knockdown using RT-PCR Primer: NIK (h2)-PR: sc-44314-PR (20 µl, 572 bp). 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 or our catalog for detailed protocols and support products.