

IKAP siRNA (m): sc-40693

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 the 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. IKK α appears to be critical for NF κ B activation in response to proinflammatory cytokines. Phosphorylation of the I κ B by IKK α is stimulated by the NF κ B inducing kinase (NIK), which itself is a central regulator for NF κ B activation in response to TNF and IL-1. The functional IKK complex contains three subunits, IKK α , IKK β and IKK γ (also designated NEMO) and each appears to make essential contributions to I κ B phosphorylation. IKAP (IKK-complex-associated protein) is a protein that acts as a scaffold, interacting with NIK, IKK α and IKK β and assembling them into an active kinase complex.

REFERENCES

1. Verma, I.M., et al. 1995. Rel/NF κ B/I κ B family: intimate tales of association and dissociation. *Genes Dev.* 9: 2723-2735.
2. Thanos, D., et al. 1995. NF κ B: a lesson in family values. *Cell* 80: 529-532.
3. Connelly, M.A., et al. 1995. CHUK, a new member of the helix-loop-helix and leucine zipper families of interacting proteins, contains a Serine/Threonine kinase catalytic domain. *Cell. Mol. Biol. Res.* 41: 537-549.
4. Malinin, N.L., et al. 1997. MAP3K-related kinase involved in NF κ B induction by TNF, CD95 and IL-1. *Nature* 385: 540-544.
5. DiDonato, J.A., et al. 1997. A cytokine-responsive I κ B kinase that activates the transcription factor NF κ B. *Nature* 388: 548-554.
6. Regnier, C.H., et al. 1997. Identification and characterization of an I κ B kinase. *Cell* 90: 373-383.
7. Scheidereit, C. 1998. Signal transduction. Docking I κ B kinases. *Nature* 395: 225-226.
8. Cohen, L., et al. 1998. IKAP is a scaffold protein of the I κ B kinase complex. *Nature* 395: 292-296.

CHROMOSOMAL LOCATION

Genetic locus: Ikbkap (mouse) mapping to 4 B3.

PRODUCT

IKAP 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see IKAP shRNA Plasmid (m): sc-40693-SH and IKAP shRNA (m) Lentiviral Particles: sc-40693-V as alternate gene silencing products.

For independent verification of IKAP (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-40693A, sc-40693B and sc-40693C.

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

IKAP siRNA (m) is recommended for the inhibition of IKAP 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 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor IKAP gene expression knockdown using RT-PCR Primer: IKAP (m)-PR: sc-40693-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.

PROTOCOLS

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