IκB-β siRNA (h): sc-29362



The Power to Question

BACKGROUND

On the basis of both functional and structural considerations, members of the lkB family of proteins can be divided into four groups. The first of these groups, lkB- α , includes the avian protein pp40 and the mammalian MAD-3, both of which inhibit binding of p50-p65 NFkB complex or Rel protein to their cognate binding sites but do not inhibit the binding of p50 homodimer to kB sites, suggesting that the lkB- α family binds to the p65 subunit of p50-p65 heterocomplex through ankyrin repeats. The second member of the lkB family is represented by a protein designated lkB- β . The third group of lkB proteins is represented by lkB- γ , which is identical in sequence with the C-terminal domain of the p110 precursor of NFkB p50 and is expressed predominantly in lymphoid cells. An additional lkB family member, lkB- ϵ , has several phosphorylated forms and is primarily found complexed with Rel A and/or c-Rel.

REFERENCES

- Ghosh, S., et al. 1990. Activation in vitro to NFκB by phosphorylation of its inhibitor IκB. Nature 344: 678-682.
- 2. Kerr, L.D., et al. 1991. The Rel-associated pp40 protein prevents DNA binding of Rel and NF κ B: relationship with I κ B- β and regulation by phosphorylation. Genes Dev. 5: 1464-1476.
- 3. Davis, N., et al. 1991. Rel-associated pp40: an inhibitor of the Rel family of transcription factors. Science 252: 1268-1271.

CHROMOSOMAL LOCATION

Genetic locus: NFKBIB (human) mapping to 19q13.2.

PRODUCT

IκB- β 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 IκB- β shRNA Plasmid (h): sc-29362-SH and IκB- β shRNA (h) Lentiviral Particles: sc-29362-V as alternate gene silencing products.

For independent verification of lkB- β (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-29362A, sc-29362B and sc-29362C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20 $^{\circ}$ C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20 $^{\circ}$ 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.

PROTOCOLS

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

APPLICATIONS

lkB- β siRNA (h) is recommended for the inhibition of lkB- β 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

 l_{κ} B-β (D-3): sc-74451 is recommended as a control antibody for monitoring of l_{κ} B-β 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 $I_{\kappa}B$ - β gene expression knockdown using RT-PCR Primer: $I_{\kappa}B$ - β (h)-PR: sc-29362-PR (20 μ I, 543 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.

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