



TANK siRNA (h): sc-36612

BACKGROUND

The tumor necrosis factor (TNF) receptor superfamily is composed of several type I integral membrane glycoproteins that exhibit homology in their cysteine-rich extracellular domains. Members of this family include TNF-R1 and -R11, FAS, OX40, CD27, CD30 and CD40. Ligands for these receptors can be small, secreted proteins such as TNF, or type II integral membrane proteins, such as the CD40 ligand, CD40L. While the signal transduction mechanism of the TNF receptor superfamily is poorly understood, stimulation of cells with either TNF or soluble CD40L has been shown to induce the nuclear translocation of NF κ B. Members of the TRAF family associate with activated TNF-R and CD40 and have been implicated in this process. The discovery of a novel protein, designated TANK, has shed light on the means by which TRAF activation of NF κ B occurs. TANK is not only capable of binding to all three TRAFs, but also of synergizing with TRAF2 to activate the NF κ B signaling cascade. TANK contains a regulatory carboxy terminal domain that maintains its inactivity in unstimulated cells. Upon its association with TRAF2, the inhibitory effect of this domain is overcome.

REFERENCES

1. Smith, C.A., et al. 1994. The TNF receptor superfamily of cellular and viral proteins: activation, costimulation and death. *Cell* 76: 959-962.
2. Cleveland, J.L., et al. 1995. Contenders in FasL/TNF death signaling. *Cell* 81: 479-482.
3. Rothe, M., et al. 1995. TRAF2-mediated activation of NF κ B by TNF receptor 2 and CD40. *Science* 269: 1424-1427.
4. Baker, S.J., et al. 1996. Transducers of life and death: TNF receptor superfamily and associated proteins. *Oncogene* 12: 1-9.
5. McLellan, A.D., et al. 1996. Human dendritic cells activate T lymphocytes via a CD40: CD40 ligand-dependent pathway. *Eur. J. Immunol.* 26: 1204-1210.
6. Snapper, C.M., et al. 1996. B cells from p50/NF κ B knockout mice have selective defects in proliferation, differentiation, germ-line CH transcription, and Ig class switching. *J. Immunol.* 156: 183-191.
7. Cheng, G., et al. 1996. TANK, a co-inducer with TRAF2 of TNF- and CD40L-mediated NF κ B activation. *Genes Dev.* 10: 963-973.

CHROMOSOMAL LOCATION

Genetic locus: TANK (human) mapping to 2q24.2.

PRODUCT

TANK 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see TANK shRNA Plasmid (h): sc-36612-SH and TANK shRNA (h) Lentiviral Particles: sc-36612-V as alternate gene silencing products.

For independent verification of TANK (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-36612A, sc-36612B and sc-36612C.

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

TANK siRNA (h) is recommended for the inhibition of TANK 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 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.

GENE EXPRESSION MONITORING

TANK (D-2): sc-166643 is recommended as a control antibody for monitoring of TANK 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-IgG κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG 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 TANK gene expression knockdown using RT-PCR Primer: TANK (h)-PR: sc-36612-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.