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# TRAF3 siRNA (h2): sc-44277

## BACKGROUND

Tumor necrosis factor (TNF)-activated cell signaling is mediated primarily through the TNF receptor 1 (TNF-R1) and, to a lesser extent, TNF-R2. Both TNF receptors are members of the expanding TNF receptor superfamily, which includes the FAS antigen and CD40. Potential insight into an understanding of TNF receptor-mediated signaling was provided by the identification of two related proteins, TRAF1 and TRAF2 (for TNF receptor-associated factors 1 and 2, respectively). Both function to form heterodimeric complexes and associate with the cytoplasmic domain of TNF-R2. A third member of this protein family, alternatively designated CD40 bp, CRAF1, LAP1 or TRAF3, has been identified and shown to associate with the cytoplasmic domain of CD40. The similarity between a specific region of TRAF3 with regions of TRAF1 and TRAF2 define a "TRAF-C" domain that is necessary and sufficient for CD40 binding and homodimerization.

## REFERENCES

1. Tartaglia, L.A., et al. 1992. Two TNF receptors. *Immunol. Today* 13: 151-153.
2. Smith, C.A., et al. 1994. The TNF receptor superfamily of cellular and viral proteins: activation, costimulation, and death. *Cell* 76: 959-962.
3. Rothe, M., et al. 1994. A novel family of putative signal transducers associated with the cytoplasmic domain of the 75 kDa tumor necrosis factor receptor. *Cell* 78: 681-692.
4. Hu, H.M., et al. 1994. A novel RING finger protein interacts with the cytoplasmic domain of CD40. *J. Biol. Chem.* 269: 30069-30072.
5. Cheng, G., et al. 1995. Involvement of CRAF1, a relative of TRAF, in CD40 signaling. *Science* 267: 1494-1498.
6. Mosiaios, G., et al. 1995. The Epstein-Barr virus transforming protein LMP1 engages signaling proteins for the tumor necrosis factor receptor family. *Cell* 80: 389-399.
7. Hsu, H., et al. 1995. The TNF receptor 1-associated protein TRADD signals cell death and NF $\kappa$ B activation. *Cell* 81: 495-504.

## CHROMOSOMAL LOCATION

Genetic locus: TRAF3 (human) mapping to 14q32.32.

## PRODUCT

TRAF3 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see TRAF3 shRNA Plasmid (h2): sc-44277-SH and TRAF3 shRNA (h2) Lentiviral Particles: sc-44277-V as alternate gene silencing products.

For independent verification of TRAF3 (h2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-44277A, sc-44277B and sc-44277C.

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) for detailed protocols and support products.

## 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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

TRAF3 siRNA (h2) is recommended for the inhibition of TRAF3 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  $\mu$ M in 66  $\mu$ 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

TRAF3 (G-6): sc-6933 is recommended as a control antibody for monitoring of TRAF3 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 $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor TRAF3 gene expression knockdown using RT-PCR Primer: TRAF3 (h2)-PR: sc-44277-PR (20  $\mu$ l, 449 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.