SANTA CRUZ BIOTECHNOLOGY, INC.

TNF-R1 siRNA (m): sc-36688

BACKGROUND

Tumor necrosis factor (TNF) is a pleiotropic cytokine whose function is mediated through two distinct cell surface receptors. These receptors, designated TNF-R1 and TNF-R2, are expressed on most cell types. The majority of TNF functions are primarily mediated through TNF-R1, while signaling through TNF-R2 occurs less extensively and is confined to cells of the immune system. Both of these proteins belong to the growing TNF and nerve growth factor (NGF) receptor superfamily, which includes FAS, CD30, CD27 and CD40. The members of this superfamily are type I membrane proteins that share sequence homology confined to the extracellular region. TNF-R1 shares a motif termed the "death domain" with FAS and three structurally unrelated signaling proteins, TRADD, FADD and RIP. This death domain is required for transduction of the apoptotic signal.

CHROMOSOMAL LOCATION

Genetic locus: Tnfrsf1a (mouse) mapping to 6 F3.

PRODUCT

TNF-R1 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see TNF-R1 shRNA Plasmid (m): sc-36688-SH and TNF-R1 shRNA (m) Lentiviral Particles: sc-36688-V as alternate gene silencing products.

For independent verification of TNF-R1 (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-36688A, sc-36688B and sc-36688C.

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

TNF-R1 siRNA (m) is recommended for the inhibition of TNF-R1 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 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

TNF-R1 (H-5): sc-8436 is recommended as a control antibody for monitoring of TNF-R1 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 TNF-R1 gene expression knockdown using RT-PCR Primer: TNF-R1 (m)-PR: sc-36688-PR (20 µl, 437 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Song, L., et al. 2007. p85α acts as a novel signal transducer for mediation of cellular apoptotic response to UV radiation. Mol. Cell. Biol. 27: 2713-2731.
2. Nishiumi, S., et al. 2010. 2,3,7,8-tetrachlorodibenzo-p-dioxin impairs an Insulin signaling pathway through the induction of tumor necrosis factor-α in adipocytes. Toxicol. Sci. 115: 482-491.
3. Wang, L., et al. 2013. IFN-γ and TNF-α synergistically induce mesenchymal stem cell impairment and tumorigenesis via NFκB signaling. Stem Cells 31: 1383-1395.
4. Li, H., et al. 2015. Administration of progranulin (PGRN) triggers ER stress and impairs Insulin sensitivity via PERK-eIF2α-dependent manner. Cell Cycle 14: 1893-1907.
5. Zhou, B., et al. 2015. Progranulin induces adipose Insulin resistance and autophagic imbalance via TNF-R1 in mice. J. Mol. Endocrinol. 55: 231-243.
6. Liu, J., et al. 2015. PGRN induces impaired Insulin sensitivity and defective autophagy in hepatic Insulin resistance. Mol. Endocrinol. 29: 528-541.

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.