

DR3 siRNA (m): sc-35217

BACKGROUND

Tumor necrosis factor (TNF)-related cytokines are pleiotropic factors that initiate various cellular processes including cell death, proliferation and differentiation. Their effects are mediated by a family of receptors which includes TNF-R1, TNF-R2, NGFR (nerve growth factor receptor) and Fas. The members of this family are type I membrane receptors and are characterized by the presence of cysteine-rich repeats in their extracellular domains. Several of these receptors, including TNF-R1 and Fas, contain a region of intracellular homology, designated the death domain, known to signal apoptosis. A new death domain member of this family, DR3 (also designated Wsl-1, APO-3, TRAMP and LARD) has been shown to induce apoptosis and activation of NFκB. DR3 is most similar in sequence to TNF-R1, but is more restricted in tissue distribution. DR3 is highly expressed in thymocytes and lymphocytes.

REFERENCES

1. Tartaglia, L.A., et al. 1993. A novel domain within the 55 kDa TNF receptor signals cell death. *Cell* 74: 845-853.
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. Nagata, S. and Golstein, P. 1995. The Fas death factor. *Science* 267: 1449-1456.
4. Ware, C.F., et al. 1996. Apoptosis mediated by the TNF-related cytokine and receptor families. *J. Cell. Biochem.* 60: 47-55.
5. Kitson, J., et al. 1996. A death-domain-containing receptor that mediates apoptosis. *Nature* 384: 372-375.

CHROMOSOMAL LOCATION

Genetic locus: Tnfrsf25 (mouse) mapping to 4 E2.

PRODUCT

DR3 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 DR3 shRNA Plasmid (m): sc-35217-SH and DR3 shRNA (m) Lentiviral Particles: sc-35217-V as alternate gene silencing products.

For independent verification of DR3 (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-35217A, sc-35217B and sc-35217C.

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

DR3 siRNA (m) is recommended for the inhibition of DR3 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

DR3 (B-8): sc-374203 is recommended as a control antibody for monitoring of DR3 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 DR3 gene expression knockdown using RT-PCR Primer: DR3 (m)-PR: sc-35217-PR (20 μl, 512 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Zhang, K., et al. 2016. TNFSF15 suppresses VEGF production in endothelial cells by stimulating miR-29b expression via activation of JNK-GATA3 signals. *Oncotarget* 7: 69436-69449.

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.