

DR1 siRNA (m): sc-62239

BACKGROUND

DR1 (down-regulator of transcription 1), also known as NC2 β (negative cofactor 2 subunit β), is a TFIID (TATA box-binding protein)-associated protein. DR1 localizes to the nucleus and contains an N-terminal histone fold motif, a TFIID-binding domain and an alanine and glutamine rich region. Via its histone fold motif, DR1 forms a heterodimer with NC2 α (DRAP1) to comprise the conserved eukaryotic complex, NC2 (negative cofactor 2). The NC2 complex can both positively and negatively regulate transcription by RNA Pol II. More specifically, NC2 acts as a repressor of TATA-dependent transcription and acts as an activator for DPE-dependent transcription. NC2 represses RNA Pol II transcription by binding to TFIID and inhibiting association of the transcription factors TFIIA and TFIIB. NC2 activity is regulated by phosphorylation. Both subunits, NC2 α and DR1, are phosphorylated *in vivo*.

REFERENCES

1. Creton, S., et al. 2002. The NC2 α and β subunits play different roles *in vivo*. *Genes Dev.* 16: 3265-3276.
2. Kanbe, E., et al. 2003. DR1-like element in human topoisomerase II α gene involved in enhancement of etoposide-induced apoptosis by PPAR γ ligand. *Exp. Hematol.* 31: 300-308.
3. Kadonaga, J.T. 2003. The DPE, a core promoter element for transcription by RNA polymerase II. *Exp. Mol. Med.* 34: 259-264.
4. Klejman, M.P., et al. 2004. NC2 α interacts with BTA1 and stimulates its ATP-dependent association with TATA-binding protein. *Mol. Cell. Biol.* 24: 10072-10082.
5. Gilfillan, S., et al. 2005. Efficient binding of NC2.TATA-binding protein to DNA in the absence of TATA. *J. Biol. Chem.* 280: 6222-6230.
6. Klejman, M.P., et al. 2005. Mutational analysis of BTA1-TBP interaction: BTA1 can rescue DNA-binding defective TBP mutants. *Nucleic Acids Res.* 33: 5426-5436.

CHROMOSOMAL LOCATION

Genetic locus: Dr1 (mouse) mapping to 5 F.

PRODUCT

DR1 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see DR1 shRNA Plasmid (m): sc-62239-SH and DR1 shRNA (m) Lentiviral Particles: sc-62239-V as alternate gene silencing products.

For independent verification of DR1 (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-62239A, sc-62239B and sc-62239C.

PROTOCOLS

See our web site at 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 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

DR1 siRNA (m) is recommended for the inhibition of DR1 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 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

DR1 (D-1): sc-515083 is recommended as a control antibody for monitoring of DR1 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 DR1 gene expression knockdown using RT-PCR Primer: DR1 (m)-PR: sc-62239-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.