

Nur77 siRNA (h2): sc-156146

BACKGROUND

Nurr1 (Nur-related factor 1) and Nur77 (also designated NGFI-B) encode orphan nuclear receptors which may comprise an additional subfamily within the nuclear receptor superfamily. The rat and human homologs of mouse Nurr1 are designated RNR1 and NOT, respectively. Both Nurr1 and Nur77 are growth factor inducible immediate early response genes. Induction of both Nurr1 and Nur77 is seen after membrane depolarization while only Nur77 induction is seen with NGF stimulation. JunD acts as a mediator for Nur77. An increase in Nur77 expression is seen in activated T cells during G₀ to G₁ transition and throughout the G₁ phase. In addition to its function as an immediate early gene, Nur77 may play a role in TCR-mediated apoptosis. Cyclosporin A, a potent immunosuppressant, has been shown to inhibit the ability of Nur77 to bind DNA. A dominant negative form of Nur77 can protect T cell hybridomas from activation-induced apoptosis. However, the absolute requirement of Nur77 for TCR-mediated apoptosis is still under debate.

REFERENCES

1. Law, S.W., et al. 1992. Identification of a new brain-specific transcription factor, Nurr1. *Mol. Endocrinol.* 6: 2129-2135.
2. Mages, H.W., et al. 1994. NOT, a human immediate-early response gene closely related to the steroid/thyroid hormone receptor NAK1/TR3. *Mol. Endocrinol.* 8: 1583-1591.
3. Davis, I.J. and Lau, L.F. 1994. Endocrine and neuro-genic regulation of the orphan nuclear receptors Nur77 and Nurr1 in the adrenal glands. *Mol. Cell. Biol.* 14: 3469-3483.
4. Garcia, I., et al. 1994. Induction of NGFI-B gene expression during T cell activation. Role of protein phosphatases. *J. Immunol.* 153: 3417-3425.

CHROMOSOMAL LOCATION

Genetic locus: NR4A1 (human) mapping to 12q13.13.

PRODUCT

Nur77 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Nur77 shRNA Plasmid (h2): sc-156146-SH and Nur77 shRNA (h2) Lentiviral Particles: sc-156146-V as alternate gene silencing products.

For independent verification of Nur77 (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-156146A, sc-156146B and sc-156146C.

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

Nur77 siRNA (h2) is recommended for the inhibition of Nur77 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

Nur77 (C-5): sc-365113 is recommended as a control antibody for monitoring of Nur77 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 Nur77 gene expression knockdown using RT-PCR Primer: Nur77 (h2)-PR: sc-156146-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Malewicz, M., et al. 2011. Essential role for DNA-PK-mediated phosphorylation of NR4A nuclear orphan receptors in DNA double-strand break repair. *Genes Dev.* 25: 2031-2040.
2. Hinze, A.V., et al. 2013. P2X1 receptor-mediated inhibition of the proliferation of human coronary smooth muscle cells involving the transcription factor NR4A1. *Purinergic Signal.* 9: 677-686.

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.