SANTA CRUZ BIOTECHNOLOGY, INC.

NF-YB siRNA (m): sc-29946

BACKGROUND

The CCAAT-binding factor NF-Y is a heteromeric transcription factor that specifically binds to CCAAT sequences in many eukaryotic genes. NF-Y is made up of three subunits, NF-YA, NF-YB and NF-YC. All three components are necessary for DNA binding. In each NF-Y subunit, the segment needed for formation of the NF-Y-DNA complex is conserved from yeast to human. These conserved segments are homologous to the histone-fold motif of eukaryotic histones. The DNA-binding domains of the NF-YB and NF-YC subunits have been suggested to interact through a protein-protein, histone-fold "handshake" motif in a manner analogous to the histone proteins H2B and H2A, respectively.

REFERENCES

1. Baxevanis, A.D., et al. 1995. A variety of DNA-binding and multimeric proteins contain the histone-fold motif. Nucleic Acids Res. 23: 2685-2691.
2. Sinha, S., et al. 1996. Three classes of mutations in the A subunit of the CCAAT-binding factor CBF delineate functional domains involved in the three-step assembly of the CBF-DNA complex. Mol. Cell. Biol. 16: 328-337.
3. Currie, R.A. 1997. Functional interaction between the DNA-binding subunit trimerization domain of NF-Y and high mobility group protein HMG-I(Y). J. Biol. Chem. 272: 30880-30888.
4. Maity, S.N., et al. 1998. Role of the CCAAT-binding protein CBF/NF-Y in transcription. Trends Biochem. Sci. 23: 174-178.
5. Liang, S.G., et al. 1998. Pathway of complex formation between DNA and three subunits of CBF/NF-Y. Photocross-linking analysis of DNA-protein interaction and characterization of equilibrium steps of subunit interaction and DNA binding. J. Biol. Chem. 273: 31590-31598.
6. Mantovani, R. 1998. A survey of 178 NF-Y-binding CCAAT boxes. Nucleic Acids Res. 26: 1135-1143.

CHROMOSOMAL LOCATION

Genetic locus: Nfyb (mouse) mapping to 10 C1.

PRODUCT

NF-YB 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 NF-YB shRNA Plasmid (m): sc-29946-SH and NF-YB shRNA (m) Lentiviral Particles: sc-29946-V as alternate gene silencing products.

For independent verification of NF-YB (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-29946A, sc-29946B and sc-29946C.

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 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

NF-YB siRNA (m) is recommended for the inhibition of NF-YB 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

NF-YB (G-2): sc-376546 is recommended as a control antibody for monitoring of NF-YB 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 NF-YB gene expression knockdown using RT-PCR Primer: NF-YB (m)-PR: sc-29946-PR (20 µl, 413 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.