SANTA CRUZ BIOTECHNOLOGY, INC.

NIP45 siRNA (m): sc-40773

BACKGROUND

The NFAT (nuclear factor of activated T cells) family of transcription factors regulates cytokine expression in T cells through *cis*-acting elements located in the promoters of cytokine genes. It is characteristic of members of the NFAT family to translocate to the nucleus, where they initiate transcription of cytokine genes subsequent to calcineurin activation. It is apparent that transcription factors such as NFAT and c-Maf require additional factors in order to mediate transcriptional activation of cytokine genes. NIP45 (for NFAT interacting protein) was identified as a protein that binds to the Rel homology domain (RHD) of NFAT c2. NIP45 and NFAT, in synergy with c-Maf, have been shown to transactivate the interleukin-4 promoter, resulting in gene transcription.

REFERENCES

1. Ho, S., et al. 1994. Cloning and characterization of NFATc and NFATp: the cytoplasmic components of NFAT. Adv. Exp. Med. Biol. 365: 167-173.
2. Ho, S.N., et al. 1995. NFATc3, a lymphoid-specific NFATc family member that is calcium-regulated and exhibits distinct DNA binding specificity. J. Biol. Chem. 270: 19898-19907.
3. Rao, A. 1995. NFATp, a cyclosporin-sensitive transcription factor implicated in cytokine gene induction. J. Leukoc. Biol. 57: 536-642.
4. Hoey, T., et al. 1995. Isolation of two new members of the NFAT gene family and functional characterization of the NFAT proteins. Immunity 2: 461-472.
5. Masuda, E.S., et al. 1995. NFATx, a novel member of the nuclear factor of activated T cells family that is expressed predominantly in the thymus. Mol. Cell. Biol. 15: 2697-2706.
6. Hodge, M.R., et al. 1996. NFAT-driven interleukin-4 transcription potentiated by NIP45. Science 274: 1903-1905.

CHROMOSOMAL LOCATION

Genetic locus: Nfatc2ip (mouse) mapping to 7 F3.

PRODUCT

NIP45 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 NIP45 shRNA Plasmid (m): sc-40773-SH and NIP45 shRNA (m) Lentiviral Particles: sc-40773-V as alternate gene silencing products.

For independent verification of NIP45 (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-40773A, sc-40773B and sc-40773C.

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.

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

NIP45 siRNA (m) is recommended for the inhibition of NIP45 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

NFATc2IP (B-1): sc-377461 is recommended as a control antibody for monitoring of NIP45 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 NIP45 gene expression knockdown using RT-PCR Primer: NIP45 (m)-PR: sc-40773-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.