SANTA CRUZ BIOTECHNOLOGY, INC.

Nix siRNA (m): sc-37454

BACKGROUND

The adenovirus E1B is a viral homolog of the Bcl-2 family of proteins that are involved in regulating cell death. A family of interacting proteins, which are designated Nip or Bnip and include Nip1, Nip2, Nip3 and Nix, associate with both the E1B protein and Bcl-2 proteins to mediate apoptotic signaling. Nip1 contains a hydrophobic transmembrane domain which enables its localization to the nuclear envelope, endoplasmic recticulum and mitochondria. Nip2 shares homology with the non-catalytic domain of Cdc42 GTPase-activating protein (Cdc42GAP). Through binding to Cdc42GAP, Nip2 enhances the GTPase activity of Cdc42GAP, facilitating the hydrolysis of GTP bound to Cdc42 and thereby mediating the signaling pathways involving receptor kinases, small GTPases and apoptotic proteins. Nix, which is also designated Nip3L or Bnip3L, is highly related to Nip3, and both proteins localize to the mitochondria where they associate with Bcl-2 proteins. Nip3 preferentially binds to Bcl-x_L and induces apoptosis by suppressing the anti-apoptosis activity of Bcl-x_L.

REFERENCES

1. Boyd, J.M., et al. 1994. Adenovirus E1B 19 kDa and Bcl-2 proteins interact with a common set of cellular proteins. Cell 79: 341-351.
2. Chiou, S.K., et al. 1994. Functional complementation of the Adenovirus E1B 19 kDa protein with Bcl-2 in the inhibition of apoptosis in infected cells. J. Virol. 68: 6553-6566.
3. Subramanian, T., et al. 1995. Functional substitution identifies a cell survival promoting domain common to Adenovirus E1B 19 kDa and Bcl-2 proteins. Oncogene 11: 2403-2409.
4. Chen, G., et al. 1997. The E1B 19 kDa/Bcl-2-binding protein Nip3 is a dimeric mitochondrial protein that activates apoptosis. J. Exp. Med. 186: 1975-1983.
5. Zhang, H., et al. 1999. Novel Bnip1 variants and their interaction with Bcl-2 family members. FEBS Lett. 448: 23-27.
6. Chen, G., et al. 1999. Nix and Nip3 form a subfamily of pro-apoptotic mitochondrial proteins. J. Biol. Chem. 274: 7-10.

CHROMOSOMAL LOCATION

Genetic locus: Bnip3l (mouse) mapping to 14 D1.

PRODUCT

Nix 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 Nix shRNA Plasmid (m): sc-37454-SH and Nix shRNA (m) Lentiviral Particles: sc-37454-V as alternate gene silencing products.

For independent verification of Nix (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-37454A, sc-37454B and sc-37454C.

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

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

Nix (H-8): sc-166332 is recommended as a control antibody for monitoring of Nix 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-IgGλ BP-HRP: sc-516132 or m-IgGλ BP-HRP (Cruz Marker): sc-516132-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-IgGλ BP-FITC: sc-516185 or m-IgGλ BP-PE: sc-516186 (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 Nix gene expression knockdown using RT-PCR Primer: Nix (m)-PR: sc-37454-PR (20 µl, 494 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Kim, Y.J., et al. 2020. BNIP3L/NIX-mediated mitophagy plays an important role in the process of age-related hearing loss. Neuroscience 455: 39-51.

RESEARCH USE

For research use only, not for use in diagnostic procedures.