SANTA CRUZ BIOTECHNOLOGY, INC.

NEIL1 siRNA (h): sc-61166

BACKGROUND

NEIL1, NEIL2 and NEIL3, also known as endonuclease VIII-like 1,2 and 3 or DNA-(apurinic or apyrimidinic site) lyase NEIL 1, 2 and 3, are nuclear proteins involved in the repair of DNA damaged by oxidation. The NEIL proteins belong to the FPG familiy of proteins. They act as DNA glycosylases that can recognize and remove damaged bases, leaving an abasic site. NEIL3, however, lacks the proline residue at the N-terminus which acts as the active site residue found in NEIL1 and NEIL2. NEIL1 is a ubiquitiously expressed protein that is up-regulated during S-phase. NEIL2 is expressed primarily in testis, heart, skeletal muscle, placenta, brain, kidney and liver while NEIL3 is detected primarily in thymus and testis.

REFERENCES

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2. Dou, H., Mitra, S. and Hazra, T.K. 2003. Repair of oxidized bases in DNA bubble structures by human DNA glycosylases NEIL1 and NEIL2. J. Biol. Chem. 278: 49679-49684.
3. Shinmura, K., Tao, H., Goto, M., Igarashi, H., Taniguchi, T., Maekawa, M., Takezaki, T. and Sugimura, H. 2004. Inactivating mutations of the human base excision repair gene NEIL1 in gastric cancer. Carcinogenesis 25: 2311-2317.
4. Das, A., Rajagopalan, L., Mathura, V.S., Rigby, S.J., Mitra, S. and Hazra, T.K. 2004. Identification of a zinc finger domain in the human NEIL2 (Nei-like-2) protein. J. Biol. Chem. 279: 47132-47138.
5. Mokkapati, S.K., Wiederhold, L., Hazra, T.K. and Mitra, S. 2004. Stimulation of DNA glycosylase activity of OGG1 by NEIL1: functional collaboration between two human DNA glycosylases. Biochemistry 43: 11596-11604.
6. Hailer, M.K., Slade, P.G., Martin, B.D., Rosenquist, T.A. and Sugden, K.D. 2005. Recognition of the oxidized lesions spiroiminodihydantoin and guanidinohydantoin in DNA by the mammalian base excision repair. DNA 4: 41-50.

CHROMOSOMAL LOCATION

Genetic locus: NEIL1 (human) mapping to 15q24.2.

PRODUCT

NEIL1 siRNA (h) 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 NEIL1 shRNA Plasmid (h): sc-61166-SH and NEIL1 shRNA (h) Lentiviral Particles: sc-61166-V as alternate gene silencing products.

For independent verification of NEIL1 (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-61166A, sc-61166B and sc-61166C.

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

NEIL1 siRNA (h) is recommended for the inhibition of NEIL1 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 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

NEIL1 (D-1): sc-271164 is recommended as a control antibody for monitoring of NEIL1 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 NEIL1 gene expression knockdown using RT-PCR Primer: NEIL1 (h)-PR: sc-61166-PR (20 µl, 600 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.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.