

NQO2 siRNA (h): sc-41575

BACKGROUND

NAD(P)H:quinone oxidoreductase 1 (NQO1) and NRH:quinone oxidoreductase (NQO2) are flavoproteins that catalyze the metabolic detoxification of quinones and their derivatives to hydroquinones. This detoxification process protects cells against quinone-induced oxidative stress, cytotoxicity and mutagenicity. NQO2 is a 231 amino acid protein and is 43 amino acids shorter than NQO1 at its C-terminus. NQO2 is an isozyme of NQO1 and transfers two electrons to a quinone, resulting in the formation of a hydroquinone product. The NQO2 gene is ubiquitously expressed and induced in response to TCDD. NQO2 has a higher level of expression in mouse liver and testis than NQO1, which is highly expressed in the heart. NQO2 has a different cofactor requirement than NQO1 and uses dihydronicotinamide riboside (NRH) rather than NAD(P)H as an electron donor. Unlike NQO1, NQO2 is resistant to typical inhibitors of NQO1 such as dicoumarol, Cibacron blue and phenindone, but is inhibited by quercetin and benzo(a)pyrene. NQO2 contains a specific metal binding site, which is absent in NQO1 and several *cis*-elements including SP1 binding sites, CCAAT box, XRE and ARE, which are located at the NQO2 gene promoter.

REFERENCES

1. Knox, R.J., et al. 2000. Bioactivation of 5-(aziridin-1-yl)-2,4-dinitrobenzamide (CB 1954) by human NAD(P)H quinone oxidoreductase 2: a novel co-substrate-mediated antitumor prodrug therapy. *Cancer Res.* 60: 4179-4186.
2. Jaiswal, A.K. 2000. Regulation of genes encoding NAD(P)H:quinone oxidoreductase. *Free Radic. Biol. Med.* 29: 254-262.
3. Chen, S., et al. 2000. Structure-function studies of DT-diaphorase (NQO1) and NRH:quinone oxidoreductase (NQO2). *Free Radic. Biol. Med.* 29: 276-284.
4. Long, D.J., 2nd. and Jaiswal, A.K. 2000. Mouse NRH:quinone oxidoreductase (NQO2): cloning of cDNA and gene- and tissue-specific expression. *Gene* 252: 107-117.
5. Long, D.J., 2nd. and Jaiswal, A.K. 2000. NRH:quinone oxidoreductase2 (NQO2). *Chem. Biol. Interact.* 129: 99-112.

CHROMOSOMAL LOCATION

Genetic locus: NQO2 (human) mapping to 6p25.2.

PRODUCT

NQO2 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see NQO2 shRNA Plasmid (h): sc-41575-SH and NQO2 shRNA (h) Lentiviral Particles: sc-41575-V as alternate gene silencing products.

For independent verification of NQO2 (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-41575A, sc-41575B and sc-41575C.

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 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

NQO2 siRNA (h) is recommended for the inhibition of NQO2 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

NQO2 (A-5): sc-271665 is recommended as a control antibody for monitoring of NQO2 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 λ BP-HRP: sc-516132 or m-IgG λ 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-IgG λ BP-FITC: sc-516185 or m-IgG λ 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 NQO2 gene expression knockdown using RT-PCR Primer: NQO2 (h)-PR: sc-41575-PR (20 μ l, 504 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.