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OGG1 siRNA (m): sc-44850

BACKGROUND

8-oxoguanine (8-oxoG), an oxidized form of guanine, is produced by reactive oxygen species in both DNA and nucleotide pools during normal aging. Accumulation of 8-oxoG increases the occurrence of A:T to C:G or G:C to T:A transversion mutations, because 8-oxoG forms a stable basepair with adenine as well as with cytosine. OGG1 (for 8-oxoG DNA glycosylase), also designated MMH, is a DNA repair enzyme that corrects these mutations. Inactivation of the OGG1 gene leads to a mutator phenotype, characterized by the increase in G:C to T:A transversions. The OGG1 gene encodes 8 isoforms (OGG1A-C, OGG2A-E) which result from alternative splicing of a single messenger RNA. The OGG1A splice variant is the most prevalent form and localizes to the nucleus, whereas the OGG2A splice variant is targeted to the mitochondria.

REFERENCES

- Shibutani, S., et al. 1991. Insertion of specific bases during DNA synthesis past the oxidation-damaged base 8-oxodG. *Nature* 349: 431-434.
- Cheng, K.C., et al. 1992. 8-hydroxyguanine, an abundant form of oxidative DNA damage, causes GT and AC substitutions. *J. Biol. Chem.* 267: 166-172.
- Ames, B.N., et al. 1993. Oxidants, antioxidants, and the degenerative diseases of aging. *Proc. Natl. Acad. Sci. USA* 90: 7915-7922.
- Hayakawa, M., et al. 1993. Age-associated damage in mitochondrial DNA in human hearts. *Mol. Cell. Biochem.* 119: 95-103.
- Nishioka, K., et al. 1999. Expression and differential intracellular localization of two major forms of human 8-oxoguanine DNA glycosylase encoded by alternatively spliced OGG1 mRNAs. *Mol. Biol. Cell* 10: 1637-1652.

CHROMOSOMAL LOCATION

Genetic locus: *Ogg1* (mouse) mapping to 6 E3.

PRODUCT

OGG1 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see OGG1 shRNA Plasmid (m): sc-44850-SH and OGG1 shRNA (m) Lentiviral Particles: sc-44850-V as alternate gene silencing products.

For independent verification of OGG1 (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-44850A, sc-44850B and sc-44850C.

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

OGG1 siRNA (m) is recommended for the inhibition of OGG1 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 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

OGG1/2 (G-5): sc-376935 is recommended as a control antibody for monitoring of OGG1 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 κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG κ 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 OGG1 gene expression knockdown using RT-PCR Primer: OGG1 (m)-PR: sc-44850-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

- Li, G., et al. 2012. 8-oxoguanine-DNA glycosylase 1 deficiency modifies allergic airway inflammation by regulating STAT6 and IL-4 in cells and in mice. *Free Radic. Biol. Med.* 52: 392-401.

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.