

# APNG siRNA (h): sc-37390

## BACKGROUND

Maintenance of DNA sequences is necessary for vertebrates and other life. DNA is under constant stress by a plethora of DNA-damaging agents present in both the environment and within cells. The potentially deleterious effects of DNA lesions in cells are elegantly resolved by sophisticated DNA repair systems, including base excision repair (BER), nucleotide excision repair (NER) and DNA repair methyltransferase (MTase). Methylated bases, such as 3-methyladenine (3MeA) and 7-methylguanine (7MeG) can be formed by agents in the environment and by endogenous cellular processes. Consequently, in the absence of exposure to environmental agents, DNA methylation damage can be incurred on the genomic DNA of normal mammalian cells. DNA N-glycosylases are base excision-repair proteins that locate and cleave damaged bases from DNA as the first step in restoring the sequence. 3MeA DNA glycosylases initiate base excision repair by removing 3MeA. These glycosylases also remove a broad spectrum of spontaneous and environmentally induced base lesions. The human N-methylpurine-DNA glycosylase gene maps to chromosome 16p13.3 and encodes a 298 amino acid protein, known as APNG.

## REFERENCES

1. O'Connor, T.R. 1993. Purification and characterization of human 3-methyladenine-DNA glycosylase. *Nucleic Acids Res.* 21: 5561-5569.
2. Friedberg, E.C., et al. 1995. *DNA Repair and Mutagenesis*. Washington, DC: ASM Press.
3. Allan, J.M., et al. 1998. Mammalian 3-methyladenine DNA glycosylase protects against the toxicity and clastogenicity of certain chemotherapeutic DNA cross-linking agents. *Cancer Res.* 58: 3965-3973.
4. Smith, S.A., et al. 2000. *In vivo* repair of methylation damage in Aag 3-methyladenine DNA glycosylase null mouse cells. *Nucleic Acids Res.* 28: 3294-3300.
5. Online Mendelian Inheritance in Man, OMIM<sup>™</sup>. 2000. Johns Hopkins University, Baltimore, MD. MIM Number: 156565. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
6. LocusLink Report (LocusID: 2243). <http://www.ncbi.nlm.nih.gov/LocusLink/>

## CHROMOSOMAL LOCATION

Genetic locus: MPG (human) mapping to 16p13.3.

## PRODUCT

APNG siRNA (h) is a pool of 2 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see APNG shRNA Plasmid (h): sc-37390-SH and APNG shRNA (h) Lentiviral Particles: sc-37390-V as alternate gene silencing products.

For independent verification of APNG (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-37390A and sc-37390B.

## 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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

APNG siRNA (h) is recommended for the inhibition of APNG 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  $\mu$ M in 66  $\mu$ 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

APNG (D-2): sc-390684 is recommended as a control antibody for monitoring of APNG 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 $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor APNG gene expression knockdown using RT-PCR Primer: APNG (h)-PR: sc-37390-PR (20  $\mu$ l). 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](http://www.scbt.com) for detailed protocols and support products.