

# FEN-1 siRNA (m): sc-37796

## BACKGROUND

DNA replication, recombination and repair, all of which are necessary for genome stability, require the presence of exonucleases. In DNA replication, these enzymes are involved in the processing of Okazaki fragments, whereas in DNA repair, they function to excise damaged DNA fragments and correct recombinational mismatches. FEN-1 (for flap endonuclease) is an endonuclease that specifically cleaves the 5' flap structure of DNA in the process of DNA repair. FEN-1 is highly homologous to yeast RAD2. The C-terminal region of FEN-1 may bind to PCNA, thus allowing FEN-1 to function as an exonuclease in DNA replication.

## REFERENCES

- Goulian, M., et al. 1990. Discontinuous DNA synthesis by purified mammalian proteins. *J. Biol. Chem.* 265: 18461-18471.
- Waga, S., et al. 1994. Reconstitution of complete SV40 DNA replication with purified replication factors. *J. Biol. Chem.* 269: 10923-10934.
- Harrington, J.J., et al. 1994. Functional domains within FEN-1 and RAD2 define a family of structure-specific endonucleases: implications for nucleotide excision repair. *Genes Dev.* 8: 1344-1355.
- Johnson, R.E., et al. 1995. Requirement of the yeast RTH1 5' to 3' exonuclease for the stability of simple repetitive DNA. *Science* 269: 238-240.
- Sommers, C.H., et al. 1995. Conditional lethality of null mutations in RTH1 that encodes the yeast counterpart of a mammalian 5'- to 3'-exonuclease required for lagging strand DNA synthesis in reconstituted systems. *J. Biol. Chem.* 270: 4193-4196.
- Hiraoka, L.R., et al. 1995. Sequence of human FEN-1, a structure-specific endonuclease, and chromosomal localization of the gene (FEN1) in mouse and human. *Genomics* 25: 220-225.
- Hosfield, D.J., et al. 1998. Structure of the DNA repair and replication endonuclease and exonuclease FEN-1: coupling DNA and PCNA binding to FEN-1 activity. *Cell* 95: 135-146.

## CHROMOSOMAL LOCATION

Genetic locus: Fen1 (mouse) mapping to 19 A.

## PRODUCT

FEN-1 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see FEN-1 shRNA Plasmid (m): sc-37796-SH and FEN-1 shRNA (m) Lentiviral Particles: sc-37796-V as alternate gene silencing products.

For independent verification of FEN-1 (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-37796A, sc-37796B and sc-37796C.

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) or our catalog 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  $\mu$ 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

FEN-1 siRNA (m) is recommended for the inhibition of FEN-1 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  $\mu$ M in 60  $\mu$ 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

FEN-1 (B-4): sc-28355 is recommended as a control antibody for monitoring of FEN-1 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor FEN-1 gene expression knockdown using RT-PCR Primer: FEN-1 (m)-PR: sc-37796-PR (20  $\mu$ l, 530 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.