

# FANCM siRNA (h): sc-77313

## BACKGROUND

Fanconi anemia (FA) is an autosomal recessive disorder characterized by bone marrow failure, birth defects and chromosomal instability. At the cellular level, FA is characterized by spontaneous chromosomal breakage and a unique hypersensitivity to DNA cross-linking agents. The thirteen FA proteins that have been characterized are important for regulating chromosomal stability and genome surveillance. Eight of these proteins, namely FANCA, FANCB, FANCC, FANCE, FANCF, FANCG, FANCL and FANCM, comprise the FA core complex, which catalyzes a key reaction in DNA repair: the monoubiquitination of FANCD2. FANCM (Fanconi anemia, complementation group M) is a member of the DEAD-box helicase family of proteins and contains a DEAH helicase domain and a nuclease domain. Localizing to chromatin fractions, FANCM is phosphorylated in a cell cycle-dependent manner and is believed to function as an anchor, recruiting the FA core complex to chromatin. Mutations in the gene encoding FANCM can lead to Fanconi anemia.

## REFERENCES

1. de Winter, J.P., et al. 2000. The Fanconi anemia protein FANCF forms a nuclear complex with FANCA, FANCC and FANCG. *Hum. Mol. Genet.* 9: 2665-2674.
2. Yagasaki, H., et al. 2001. A cytoplasmic serine protein kinase binds and may regulate the Fanconi anemia protein FANCA. *Blood* 98: 3650-3657.
3. Wilson, J.B., et al. 2001. The Chinese hamster FANCG/XRCC9 mutant NM3 fails to express the monoubiquitinated form of the FANCD2 protein, is hyper-sensitive to a range of DNA damaging agents and exhibits a normal level of spontaneous sister chromatid exchange. *Carcinogenesis* 22: 1939-1946.
4. Siddique, M.A., et al. 2001. Function of the Fanconi anemia pathway in Fanconi anemia complementation group F and D1 cells. *Exp. Hematol.* 29: 1448-1455.
5. Otsuki, T., et al. 2002. Phosphorylation of Fanconi anemia protein, FANCA, is regulated by Akt kinase. *Biochem. Biophys. Res. Commun.* 291: 628-634.
6. Taniguchi, T. and Dandrea, A.D. 2002. Molecular pathogenesis of fanconi anemia. *Int. J. Hematol.* 75: 123-128.

## CHROMOSOMAL LOCATION

Genetic locus: FANCM (human) mapping to 14q21.2.

## PRODUCT

FANCM 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see FANCM shRNA Plasmid (h): sc-77313-SH and FANCM shRNA (h) Lentiviral Particles: sc-77313-V as alternate gene silencing products.

For independent verification of FANCM (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-77313A, sc-77313B and sc-77313C.

## 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

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

FANCM (M40-P2C6): sc-101389 is recommended as a control antibody for monitoring of FANCM 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 FANCM gene expression knockdown using RT-PCR Primer: FANCM (h)-PR: sc-77313-PR (20  $\mu$ l, 417 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](http://www.scbt.com) for detailed protocols and support products.