

FANCD2 siRNA (m): sc-35357

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. At least eight complementation groups (A-G) have been identified and six FA genes (for subtypes A, C, D2, E, F and G) have been cloned. The FA proteins lack sequence homologies or motifs that could point to a molecular function. Phosphorylation of FANCF (Fanconi anemia complementation group) proteins are thought to be important for the function of the FA pathway. Several FA proteins, including FANCA, FANCC, FANCF and FANCG, interact in a nuclear complex, and this complex is required for the activation (monoubiquitination) of the downstream FANCD2 protein. When monoubiquitinated, the FANCD2 protein co-localizes with the breast cancer susceptibility protein BRCA1 in DNA damage induced foci. In male meiosis, FANCD2 also co-localizes with BRCA1 at synaptonemal complexes. The human FANCD2 gene maps to chromosome 3p25.3, contains 44 exons and encodes a 1,451 amino acid nuclear protein that exists as 2 protein isoforms.

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 hypersensitive 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. Grompe, M. and D'Andrea, A. 2001. Fanconi anemia and DNA repair. *Hum. Mol. Genet.* 10: 2253-2259.

CHROMOSOMAL LOCATION

Genetic locus: *Fancd2* (mouse) mapping to 6 E3.

PRODUCT

FANCD2 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 FANCD2 shRNA Plasmid (m): sc-35357-SH and FANCD2 shRNA (m) Lentiviral Particles: sc-35357-V as alternate gene silencing products.

For independent verification of FANCD2 (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-35357A, sc-35357B and sc-35357C.

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

FANCD2 shRNA Plasmid (m) is recommended for the inhibition of FANCD2 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor FANCD2 gene expression knockdown using RT-PCR Primer: FANCD2 (m)-PR: sc-35357-PR (20 μ l, 489 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 for detailed protocols and support products.