

MSH2 siRNA (h): sc-35969

BACKGROUND

The finding that mutations in DNA mismatch repair genes are associated with hereditary nonpolyposis colorectal cancer (HNPCC) has resulted in considerable interest in the understanding of the mechanism of DNA mismatch repair. Initially, inherited mutations in the MSH2 and MLH1 homologs of the bacterial DNA mismatch repair genes MutS and MutL were demonstrated at high frequency in HNPCC and were shown to be associated with microsatellite instability. The demonstration that 10 to 45% of pancreatic, gastric, breast, ovarian and small cell lung cancers also display microsatellite instability has been interpreted to suggest that DNA mismatch repair is not restricted to HNPCC tumors but is a common feature in tumor initiation or progression. Two additional homologs of the prokaryotic MutL gene, designated PMS1 and PMS2, have been identified and shown to be mutated in the germline of HNPCC patients.

REFERENCES

1. Peltomäki, P., et al. 1993. Genetic mapping of a locus predisposing to human colorectal cancer. *Science* 260: 810-812.
2. Ionov, Y., et al. 1993. Ubiquitous somatic mutations in simple repeated sequences reveal a new mechanism for colonic carcinogenesis. *Nature* 363: 558-561.
3. Papadopoulos, N., et al. 1994. Mutation of a MutL homolog in hereditary colon cancer. *Science* 263: 1625-1629.
4. Prolla, T.A., et al. 1994. MLH1, PMS1, and MSH2 interactions during the initiation of DNA mismatch repair in yeast. *Science* 265: 1091-1092.
5. Palombo, F., et al. 1994. Mismatch repair and cancer. *Nature* 367: 417-418.

CHROMOSOMAL LOCATION

Genetic locus: MSH2 (human) mapping to 2p21.

PRODUCT

MSH2 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see MSH2 shRNA Plasmid (h): sc-35969-SH and MSH2 shRNA (h) Lentiviral Particles: sc-35969-V as alternate gene silencing products.

For independent verification of MSH2 (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-35969A, sc-35969B and sc-35969C.

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

MSH2 siRNA (h) is recommended for the inhibition of MSH2 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 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

MSH2 (D-6): sc-376384 is recommended as a control antibody for monitoring of MSH2 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 MSH2 gene expression knockdown using RT-PCR Primer: MSH2 (h)-PR: sc-35969-PR (20 μ l, 491 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Kang, X., et al. 2009. Regulation of the hTERT promoter activity by MSH2, the hnRNPs K and D, and GRHL2 in human oral squamous cell carcinoma cells. *Oncogene* 28: 565-574.
2. Gaymes, T.J., et al. 2013. Microsatellite instability induced mutations in DNA repair genes CtIP and MRE11 confer hypersensitivity to poly (ADP-ribose) polymerase inhibitors in myeloid malignancies. *Haematologica* 98: 1397-1406.

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.