SANTA CRUZ BIOTECHNOLOGY, INC.

GTBP siRNA (m): sc-35529

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. A member of the mismatch repair family, GTBP (also designated MSH6), is a 160 kDa MSH2-related protein that binds to DNA containing G/T mismatches. Findings suggest that the mismatch-binding factor in human cells is composed of a heterodimer of GTBP and MSH2.

REFERENCES

1. Peltomäki, P., et al. 1993. Genetic mapping of a locus predisposing to human colorectal cancer. Science 260: 810-812.
2. Palombo, F., et al. 1994. Mismatch repair and cancer. Nature 367: 417-418.
3. Bronner, C.E., et al. 1994. Mutation in the DNA mismatch repair gene homologue hMLH1 is associated with hereditary non-polyposis colon cancer. Nature 368: 258-261.
4. Papadopoulos, N., et al. 1994. Mutation of a MutL homolog in hereditary colon cancer. Science 263: 1625-1629.
5. Nicolaides, N.C., et al. 1994. Mutations of two PMS homologues in hereditary nonpolyposis colon cancer. Nature 371: 75-80.
6. Prolla, T.A., et al. 1994. MLH1, Pms1, and Msh2 interactions during the initiation of DNA mismatch repair in yeast. Science 265: 1091-1092.
7. Palombo, F., et al. 1995. GTBP, a 160 kDa protein essential for mismatch-binding activity in human cells. Science 268: 1912-1914.
8. Shiwaku, H.O., et al. 1997. Alternative splicing of GTBP in normal human tissues. DNA Res. 4: 359-362.
9. Ercoli, A., et al. 1999. hMSH2 and GTBP expression in advanced stage epithelial ovarian cancer. Br. J. Cancer 80: 1665-1671.

CHROMOSOMAL LOCATION

Genetic locus: Msh6 (mouse) mapping to 17 E4.

PRODUCT

GTBP 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see GTBP shRNA Plasmid (m): sc-35529-SH and GTBP shRNA (m) Lentiviral Particles: sc-35529-V as alternate gene silencing products.

For independent verification of GTBP (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-35529A, sc-35529B and sc-35529C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

GTBP siRNA (m) is recommended for the inhibition of GTBP 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 µM in 66 µl. 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

GTBP (E-8): sc-137015 is recommended as a control antibody for monitoring of GTBP 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 GTBP gene expression knockdown using RT-PCR Primer: GTBP (m)-PR: sc-35529-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.