

BRIP1 siRNA (m): sc-61836

BACKGROUND

Genes that contribute to tumorigenesis can be broadly classified as either gatekeepers or caretakers. Genes in the gatekeeper class directly regulate cell division or cell death, and their alteration results in the uncontrolled cellular proliferation that characterizes tumor cells. Genes in the caretaker class are involved in DNA metabolic processes and are responsible for maintaining the overall stability of the genome. An unusual mutator phenotype in *Caenorhabditis elegans*, characterized by deletions that start around the 3' end of polyguanine tracts and terminate at variable positions 5' from such tracts, results from disruption of a gene that encodes BRIP1 (also designated BACH1 or BRCA1-associated carboxy-terminal helicase-1). BRCA1 interacts *in vivo* with BRIP1, a member of the DEAH helicase family. BRIP1 contains the seven helicase-specific motifs that are conserved among members of the DEAH family, and the helicase domain includes a nuclear localization signal. BRIP1 is ubiquitously expressed with highest levels in testis, an expression pattern similar to that of BRCA1. BRIP1 binds directly to the BRCT repeats of BRCA1 and the BRIP1-BRCA1 complex formation contributes to a key BRCA1 activity. BRIP1 is required to resolve the secondary structures of guanine-rich DNA that occasionally form during lagging-strand DNA synthesis. Phosphorylated BRIP1/BACH1 binds directly to the BRCT domain of BRCA1. This interaction is dependent on the phosphorylation of BRIP1/BACH1 at Ser 990, and is required for DNA damage-induced checkpoint control during the G₂ to M phase transition of the cell cycle.

REFERENCES

1. Cantor, S.B., et al. 2001. BACH1, a novel helicase-like protein, interacts directly with BRCA1 and contributes to its DNA repair function. *Cell* 105: 149-160.
2. Liu, Y., et al. 2002. Distinct functions of BRCA1 and BRCA2 in double-strand break repair. *Breast Cancer Res.* 4: 9-13.
3. Yu, X., et al. 2003. The BRCT domain is a phosphoprotein binding domain. *Science* 302: 639-642.
4. Rodriguez, M., et al. 2003. Phosphopeptide binding specificities of BRCA1 COOH-terminal (BRCT) domains. *J. Biol. Chem.* 278: 52914-52918.

CHROMOSOMAL LOCATION

Genetic locus: Brip1 (mouse) mapping to 11 C.

PRODUCT

BRIP1 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 BRIP1 shRNA Plasmid (m): sc-61836-SH and BRIP1 shRNA (m) Lentiviral Particles: sc-61836-V as alternate gene silencing products.

For independent verification of BRIP1 (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-61836A, sc-61836B and sc-61836C.

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

BRIP1 siRNA (m) is recommended for the inhibition of BRIP1 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.

GENE EXPRESSION MONITORING

BRIP1 (E-11): sc-365708 is recommended as a control antibody for monitoring of BRIP1 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 MarkerTM 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 BRIP1 gene expression knockdown using RT-PCR Primer: BRIP1 (m)-PR: sc-61836-PR (20 μ l). 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.