SANTA CRUZ BIOTECHNOLOGY, INC.

BARD1 siRNA (m): sc-37312

BACKGROUND

Mutations within the BRCA1 gene, localized to chromosome 17q, are believed to account for approximately 45% of families with increased incidence of both early-onset breast cancer and ovarian cancer. The BRCA1 gene is expressed in numerous tissues, including breast and ovary, and encodes a predicted protein of 1,863 amino acids. This protein contains a RING domain near the N-terminus and appears to encode a tumor suppressor. BARD1 (BRCA1-associated RING domain protein 1) and BAP1 (BRCA1-associated protein 1) have both been shown to bind to the N-terminus of BRCA1 and are potential mediators of tumor suppression. BARD1 contains an N-terminal RING domain and three tandem ankyrin repeats. The C-terminus of BARD1 contains a region with sequence homology to BRCA1, termed the BRCT domain. BAP1 is a ubiquitin hydrolase and has been shown to enhance BRCA1-mediated cell growth suppression.

REFERENCES

1. Hall, J.M., et al. 1990. Linkage of early-onset familial breast cancer to chromosome 17q21. Science 250: 1684-1689.
2. Narod, S.A., et al. 1991. Familial breast-ovarian cancer locus on chromosome 17q12-q23. Lancet 338: 82-83.
3. Novak, R. 1994. Breast cancer gene offers surprises. Science 265: 1796-1799.
4. Futreal, P.A., et al. 1994. BRCA1 mutations in primary breast and ovarian carcinomas. Science 266: 120-122.
5. Wu, L.C., et al. 1996. Identification of a RING protein that can interact *in vivo* with the BRCA1 gene product. Nat. Genet. 14: 430-440.
6. Jin, Y., et al. 1997. Cell cycle-dependent colocalization of BARD1 and BRCA1 proteins in discrete nuclear domains. Proc. Natl. Acad. Sci. USA 94: 12075-12080.
7. Thai, T.H., et al. 1998. Mutations in the BRCA1-associated RING domain (BARD1) gene in primary breast, ovarian and uterine cancers. Hum. Mol. Genet. 7: 195-202.
8. Jensen, D.E., et al. 1998. BAP1: a novel ubiquitin hydrolase which binds to the BRCA1 RING finger and enhances BRCA1-mediated cell growth suppression. Oncogene 16: 1097-1112.

CHROMOSOMAL LOCATION

Genetic locus: Bard1 (mouse) mapping to 1 C3.

PRODUCT

BARD1 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 BARD1 shRNA Plasmid (m): sc-37312-SH and BARD1 shRNA (m) Lentiviral Particles: sc-37312-V as alternate gene silencing products.

For independent verification of BARD1 (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-37312A, sc-37312B and sc-37312C.

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

BARD1 siRNA (m) is recommended for the inhibition of BARD1 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

BARD1 (E-11): sc-74559 is recommended as a control antibody for monitoring of BARD1 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 BARD1 gene expression knockdown using RT-PCR Primer: BARD1 (m)-PR: sc-37312-PR (20 µl). 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.