SANTA CRUZ BIOTECHNOLOGY, INC.

DinB siRNA (h): sc-60537

BACKGROUND

Problems in DNA replication may lead to breaks in the replication fork, and recombinational reactions occur to restore the integrity of the fork via strand-invasion of the broken chromosome with its homologous strand. If this happens within repeated DNA sequences, genetic rearrangements may be produced. The bacterial UmuC/DinB family consists of bypass polymerases that are responsible for translesion DNA synthesis. DinB, also referred to as DNA polyermase IV or DNA polymerase κ, is an SOS-inducible, error-prone DNA polymerase that plays a role in DNA damage-induced mutagenesis by preferentially making frameshift mutations. DinB is uniquely and highly expressed in the adrenal cortex and testis, as well as in a variety of other tissues. p53 regulates DinB and exposure to various DNA-damaging agents causes an upregulation of DinB.

REFERENCES

1. Silvian, L.F., et al. 2001. Crystal structure of a DinB family error-prone DNA polymerase from *Sulfolobus solfataricus*. Nat. Struct. Biol. 8: 984-989.
2. Zhou, B.L., et al. 2001. Crystal structure of a DinB lesion bypass DNA polymerase catalytic fragment reveals a classic polymerase catalytic domain. Mol. Cell 8: 427-437.
3. McKenzie, G.J., et al. 2003. The DinB operon and spontaneous mutation in *Escherichia coli*. J. Bacteriol. 185: 3972-3977.
4. Velasco-Miguel, S., et al. 2003. Constitutive and regulated expression of the mouse DinB (pol-κ) gene encoding DNA polymerase κ. DNA Repair 2: 91-106.
5. Maisnier-Patin, S., et al. 2005. Genomic buffering mitigates the effects of deleterious mutations in bacteria. Nat. Genet. 37: 1376-1379.
6. Perez-Capilla, T., et al. 2005. SOS-independent induction of DinB transcription by β-lactam-mediated inhibition of cell wall synthesis in *Escherichia coli*. J. Bacteriol. 187: 1515-1518.
7. Beuning, P.J., et al. 2006. Two processivity clamp interactions differentially alter the dual activities of UmuC. Mol. Microbiol. 59: 460-474.
8. Hersh, M.N., et al. 2006. Single-strand-specific exonucleases prevent frameshift mutagenesis by suppressing SOS induction and the action of DinB/DNA polymerase IV in growing cells. J. Bacteriol. 188: 2336-2342.

CHROMOSOMAL LOCATION

Genetic locus: POLK (human) mapping to 5q13.3.

PRODUCT

DinB 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see DinB shRNA Plasmid (h): sc-60537-SH and DinB shRNA (h) Lentiviral Particles: sc-60537-V as alternate gene silencing products.

For independent verification of DinB (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-60537A, sc-60537B and sc-60537C.

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

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

DinB (A-9): sc-166667 is recommended as a control antibody for monitoring of DinB 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 DinB gene expression knockdown using RT-PCR Primer: DinB (h)-PR: sc-60537-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.