

XRCC3 siRNA (h): sc-37403

BACKGROUND

The x-ray repair cross-complementing (XRCC) proteins are responsible for efficiently repairing and maintaining genetic stability following DNA base damage. These genes share sequence similarity with the yeast DNA repair protein Rad51. XRCC1 is a protein that facilitates the DNA base excision repair pathway by interacting with DNA ligase III and DNA polymerase to repair DNA single-strand breaks. XRCC2 and XRCC3 are both involved in maintaining chromosome stability during cell division. XRCC2 is required for efficient repair of DNA double-strand breaks by homologous recombination between sister chromatids, and XRCC3 interacts directly with Rad51 to co-operate with Rad51 during recombinational repair. XRCC4 is an accessory factor of DNA ligase IV that preferentially binds DNA with nicks or broken ends. XRCC4 binds to DNA ligase IV and enhances its joining activity, and it is also involved in V(D)J recombination. Any defect in one of the known components of the DNA repair/V(D)J recombination machinery (Ku-70, Ku-80, DNA-PK α , XRCC4 and DNA ligase IV) leads to abortion of the V(D)J rearrangement process and early block in both T and B cell maturation.

REFERENCES

1. Nash, R.A., et al. 1997. XRCC1 protein interacts with one of two distinct forms of DNA ligase III. *Biochemistry* 36: 5207-5211.
2. Liu, N., et al. 1998. XRCC2 and XRCC3, new human Rad51-family members, promote chromosome stability and protect against DNA cross-links and other damages. *Mol. Cell* 1: 783-793.
3. Thacker, J. 1999. The role of homologous recombination processes in the repair of severe forms of DNA damage in mammalian cells. *Biochimie* 81: 77-85.
4. Johnson, R.D., et al. 1999. Mammalian XRCC2 promotes the repair of DNA double-strand breaks by homologous recombination. *Nature* 401: 397-399.
5. Pierce, A.J., et al. 1999. XRCC3 promotes homology-directed repair of DNA damage in mammalian cells. *Genes Dev.* 13: 2633-2638.
6. Modesti, M., et al. 1999. DNA binding of Xrcc4 protein is associated with V(D)J recombination but not with stimulation of DNA ligase IV activity. *EMBO J.* 18: 2008-2018.

CHROMOSOMAL LOCATION

Genetic locus: XRCC3 (human) mapping to 14q32.33.

PRODUCT

XRCC3 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 XRCC3 shRNA Plasmid (h): sc-37403-SH and XRCC3 shRNA (h) Lentiviral Particles: sc-37403-V as alternate gene silencing products.

For independent verification of XRCC3 (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-37403A, sc-37403B and sc-37403C.

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

XRCC3 siRNA (h) is recommended for the inhibition of XRCC3 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

XRCC3 (D-7): sc-271714 is recommended as a control antibody for monitoring of XRCC3 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 XRCC3 gene expression knockdown using RT-PCR Primer: XRCC3 (h)-PR: sc-37403-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.