SANTA CRUZ BIOTECHNOLOGY, INC.

ALKBH4 siRNA (m): sc-141021

BACKGROUND

ALKBH4 (ALKB, alkylation repair homolog 4), also known as ABH4, is a 302 amino acid protein belonging to the ALKB family. ALKBH4 is one of many homo-logs of the *Escherichia coli* protein ALKB. ALKB functions to protect DNA and RNA against damage from environmental methylating compounds by directly reversing 1-methyladenine (1-meA) and 3-methylcytosine (3-meC) cytotoxic alkylation lesions in DNA and RNA. The enzyme acts by oxidative demethylation, utilizing ferrous iron and α-ketoglutarate as cofactors, 2-oxoglutarate as a co-substrate and molecular oxygen as the oxidizing agent. ALKBH4 exists as two alternatively spliced isoforms and is encoded by a gene located on human chromosome 7q22.1, which houses over 1,000 genes and comprises nearly 5% of the human genome.

REFERENCES

1. Kim, M.Y., et al. 2007. ALKB influences the chloroacetaldehyde-induced mutation spectra and toxicity in the pSP189 supF shuttle vector. Chem. Res. Toxicol. 20: 1075-1083.
2. Tsujikawa, K., et al. 2007. Expression and sub-cellular localization of human ABH family molecules. J. Cell. Mol. Med. 11: 1105-1116.
3. Bleijlevens, B., et al. 2007. Replacement of non-heme Fe(II) with Cu(II) in the α-ketoglutarate dependent DNA repair enzyme ALKB: spectroscopic characterization of the active site. J. Inorg. Biochem. 101: 1043-1048.
4. Roy, T.W. and Bhagwat, A.S. 2007. Kinetic studies of *Escherichia coli* ALKB using a new fluorescence-based assay for DNA demethylation. Nucleic Acids Res. 35: e147.
5. Ringvoll, J., et al. 2008. ALKB homologue 2-mediated repair of ethenoadenine lesions in mammalian DNA. Cancer Res. 68: 4142-4149.
6. Fix, D., et al. 2008. Transcription increases methylmethane sulfonate-induced mutations in ALKB strains of *Escherichia coli*. DNA Repair 7: 1289-1297.
7. Yang, C.G., et al. 2008. Crystal structures of DNA/RNA repair enzymes ALKB and ABH2 bound to dsDNA. Nature 452: 961-965.

CHROMOSOMAL LOCATION

Genetic locus: Alkbh4 (mouse) mapping to 5 G2.

PRODUCT

ALKBH4 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 ALKBH4 shRNA Plasmid (m): sc-141021-SH and ALKBH4 shRNA (m) Lentiviral Particles: sc-141021-V as alternate gene silencing products.

For independent verification of ALKBH4 (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-141021A, sc-141021B and sc-141021C.

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

ALKBH4 siRNA (m) is recommended for the inhibition of ALKBH4 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor ALKBH4 gene expression knockdown using RT-PCR Primer: ALKBH4 (m)-PR: sc-141021-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.