SANTA CRUZ BIOTECHNOLOGY, INC.

BCKDHB siRNA (m): sc-141668

BACKGROUND

BCKDHB (branched chain keto acid dehydrogenase E1, β polypeptide), also known as 2-oxoisovalerate dehydrogenase subunit β mitochondrial or E1B, is a 392 amino acid mitochondrial matrix protein and component of branched-chain keto acid dehydrogenase, a multienzyme complex involved in the catabolism of branched-chain amino acids. Existing as a heterodimer, BCKDHB is encoded by a gene mapping to human chromosome 6q14.1, whose defects are the cause of an autosomal recessive disorder known as maple syrup urine disease type IB (MSUD1B). Characterized by urine with maple syrup odor, patients with maple syrup urine disease may suffer severe neurological damage, mental retardation and feeding problems.

REFERENCES

1. Chuang, J.L., et al. 1990. Molecular cloning of the mature E1b-β subunit of human branched-chain α-keto acid dehydrogenase complex. FEBS Lett. 262: 305-309.
2. Zneimer, S.M., et al. 1991. Regional assignment of two genes of the human branched-chain α-keto acid dehydrogenase complex: the E1 β gene (BCKDHB) to chromosome 6p21-22 and the E2 gene (DBT) to chromosome 1p31. Genomics 103: 740-747.
3. Patel, M.S., et al. 1995. Mammalian α-keto acid dehydrogenase complexes: gene regulation and genetic defects. FASEB J. 9: 1164-1172.
4. Chuang, J.L., et al. 1996. Maple syrup urine disease: the E1 β gene of human branched-chain α-ketoacid dehydrogenase complex has 11 rather than 10 exons, and the 3' UTR in one of the two E1 β mRNAs arises from intronic sequences. Am. J. Hum. Genet. 586: 1373-1377.
5. Chuang, D.T. 1998. Maple syrup urine disease: it has come a long way. J. Pediatr. 132: S17-S23.
6. Chuang, J.L., et al. 2004. Structural and biochemical basis for novel mutations in homozygous Israeli maple syrup urine disease patients: a proposed mechanism for the thiamin-responsive phenotype. J. Biol. Chem. 279: 17792-17800.

CHROMOSOMAL LOCATION

Genetic locus: Bckdhb (mouse) mapping to 9 E2.

PRODUCT

BCKDHB siRNA (m) is a target-specific 19-25 nt siRNA 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 BCKDHB shRNA Plasmid (m): sc-141668-SH and BCKDHB shRNA (m) Lentiviral Particles: sc-141668-V as alternate gene silencing products.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

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

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

BCKDHB (H-6): sc-374630 is recommended as a control antibody for monitoring of BCKDHB 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 BCKDHB gene expression knockdown using RT-PCR Primer: BCKDHB (m)-PR: sc-141668-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.