

PCBD1 siRNA (m): sc-152050

BACKGROUND

PCBD1 (pterin-4 α -carbinolamine dehydratase), also known as PCD, PHS, DCOH (dimerization cofactor of hepatocyte nuclear factor 1 α) or PCBD, is a component of the phenylalanine hydroxylase (PAH) system and participates in tetrahydrobiopterin biosynthesis. More specifically, PCBD1 catalyzes the dehydration of pterin-4 α -carbinolamine (4-OH-BH4) to quinonoid dihydrobiopterin (q-BH2), an essential reaction for the regeneration of 6(R)-L-erythro-5,6,7,8-tetrahydrobiopterin (6(R)BH4). In addition, PCBD1 can homodimerize and, in this dimer, can function as a transcriptional activator cofactor for HNF-1 α . Mutations in the gene encoding PCBD1 lead to an accumulation of 4-OH-BH4 which subsequently produces 7-BH4 (a potent inhibitor of PAH), and may result in primapterinuria. Patients with primapterinuria, a mild form of phenylketonuria (PKU), exhibit both hyperphenylalaninemia (HPA) and excretion of 7-substituted pterins.

REFERENCES

- Thöny, B., et al. 1998. Hyperphenylalaninemia with high levels of 7-biopterin is associated with mutations in the PCBD gene encoding the bifunctional protein pterin-4 α -carbinolamine dehydratase and transcriptional coactivator (DCoH). *Am. J. Hum. Genet.* 62: 1302-1311.
- Online Mendelian Inheritance in Man, OMIM[™]. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 126090. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
- Schallreuter, K.U., et al. 2003. Molecular evidence that halo in Sutton's naevus is not vitiligo. *Arch. Dermatol. Res.* 295: 223-228.
- Schallreuter, K.U., et al. 2004. Activation/deactivation of acetylcholinesterase by H₂O₂: more evidence for oxidative stress in vitiligo. *Biochem. Biophys. Res. Commun.* 315: 502-508.
- Melo, A.M., et al. 2005. A nhaD Na⁺/H⁺ antiporter and a pcd homologues are among the Rhodothermus marinus complex I genes. *Biochim. Biophys. Acta* 1709: 95-103.
- Hasse, S., et al. 2005. *In vivo* and *in vitro* evidence for autocrine DCoH/HNF-1 α transcription of albumin in the human epidermis. *Exp. Dermatol.* 14: 182-187.

CHROMOSOMAL LOCATION

Genetic locus: Pcbd1 (mouse) mapping to 10 B4.

PRODUCT

PCBD1 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PCBD1 shRNA Plasmid (m): sc-152050-SH and PCBD1 shRNA (m) Lentiviral Particles: sc-152050-V as alternate gene silencing products.

For independent verification of PCBD1 (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-152050A, sc-152050B and sc-152050C.

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

PCBD1 siRNA (m) is recommended for the inhibition of PCBD1 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 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

PCBD1 (E-7): sc-518184 is recommended as a control antibody for monitoring of PCBD1 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 Marker[™] 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 PCBD1 gene expression knockdown using RT-PCR Primer: PCBD1 (m)-PR: sc-152050-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.