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CDKAL1 siRNA (m): sc-142228

BACKGROUND

Cell cycle progression is controlled, in part, by a family of cyclin dependent kinases (Cdks) that work to phosphorylate key substrates involved in each phase of cell cycle progression. Cdks are the catalytic subunits of serine/threonine protein kinases, a large family of proteins that act as regulators of the eukaryotic cell cycle. CDKAL1 (Cdk5 regulatory subunit associated protein 1-like 1) is a 579 amino acid single-pass membrane protein that contains one TRAM domain and is similar to Cdk5 regulatory subunit associated proteins (CDK5RAPs). Expressed in pancreas, brain and skeletal muscle, CDKAL1 uses iron as a cofactor and is involved in glucose-stimulated Insulin secretion. Defects in the gene encoding CDKAL1 impair Insulin secretion and are associated with the development of type 2 diabetes. Multiple isoforms of CDKAL1 exist due to alternative splicing events.

REFERENCES

1. Pascoe, L., et al. 2007. Common variants of the novel type 2 diabetes genes CDKAL1 and HHEX/IDE are associated with decreased pancreatic β -cell function. *Diabetes* 56: 3101-3104.
2. Steinthorsdottir, V., et al. 2007. A variant in CDKAL1 influences Insulin response and risk of type 2 diabetes. *Nat. Genet.* 39: 770-775.
3. Online Mendelian Inheritance in Man, OMIM[™]. 2007. Johns Hopkins University, Baltimore, MD. MIM Number: 611259. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
4. van Hoek, M., et al. 2008. Predicting type 2 diabetes based on polymorphisms from genome-wide association studies: a population-based study. *Diabetes* 57: 3122-3128.
5. Kirchhoff, K., et al. 2008. Polymorphisms in the TCF7L2, CDKAL1 and SLC30A8 genes are associated with impaired proinsulin conversion. *Diabetologia* 51: 597-601.
6. Groenewoud, M.J., et al. 2008. Variants of CDKAL1 and IGF2BP2 affect first-phase Insulin secretion during hyperglycaemic clamps. *Diabetologia* 51: 1659-1663.

CHROMOSOMAL LOCATION

Genetic locus: Cdkal1 (mouse) mapping to 13 A3.1.

PRODUCT

CDKAL1 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 CDKAL1 shRNA Plasmid (m): sc-142228-SH and CDKAL1 shRNA (m) Lentiviral Particles: sc-142228-V as alternate gene silencing products.

For independent verification of CDKAL1 (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-142228A, sc-142228B and sc-142228C.

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 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

CDKAL1 siRNA (m) is recommended for the inhibition of CDKAL1 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

CDKAL1 (E-9): sc-393447 is recommended as a control antibody for monitoring of CDKAL1 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 λ BP-HRP: sc-516132 or m-IgG λ BP-HRP (Cruz Marker): sc-516132-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 λ BP-FITC: sc-516185 or m-IgG λ BP-PE: sc-516186 (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 CDKAL1 gene expression knockdown using RT-PCR Primer: CDKAL1 (m)-PR: sc-142228-PR (20 μ l, 553 bp). 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.