

Neurogenin 3 siRNA (m): sc-42080

BACKGROUND

The neurogenin family of proteins belongs to the basic helix-loop-helix (bHLH) superfamily and consists of Neurogenin 1, Neurogenin 2 and Neurogenin 3 (also designated *ngn3*). bHLH members are transcriptional regulators that determine cell fate. Neurogenin 3 is expressed in discrete regions of developing neurons and in the embryonic pancreatic islets of Langerhans. HNF-6 (hepatocyte nuclear factor 6) acts as a positive regulator of Neurogenin 3 by binding to and stimulating the neurogenin gene promoter. Neurogenin 3 is involved in the initial differentiation of the four islets cell types, while a network of transcription factors, including Hlx9, Isl1, Neuro D, Nkx-2.2, Nkx-6.4, Pax-4, Pax-6, PDX-1 and Mash1, are required for final differentiation. Neurogenin 3 acts upstream of Neuro D in a bHLH cascade. Neurogenin 3 activates the expression of Neuro D at the onset of islet cell differentiation through box sequences E1 and E3 in the Neuro D promoter.

REFERENCES

- Huang, H.P., et al. 2000. Regulation of the pancreatic islet-specific gene BETA2 (*neuroD*) by Neurogenin 3. *Mol. Cell. Biol.* 20: 3292-3307.
- Jacquemin, P., et al. 2000. Transcription factor hepatocyte nuclear factor 6 regulates pancreatic endocrine cell differentiation and controls expression of the proendocrine gene *ngn3*. *Mol. Cell. Biol.* 20: 4445-4454.
- Gradwohl, G., et al. 2000. Neurogenin 3 is required for the development of the four endocrine cell lineages of the pancreas. *Proc. Natl. Acad. Sci. USA* 97: 1607-1611.
- Schwitzgebel, V.M., et al. 2000. Expression of Neurogenin 3 reveals an islet cell precursor population in the pancreas. *Development* 127: 3533-3542.
- Jensen, J., et al. 2000. Independent development of pancreatic α - and β -cells from Neurogenin 3-expressing precursors: a role for the notch pathway in repression of premature differentiation. *Diabetes* 49: 163-176.

CHROMOSOMAL LOCATION

Genetic locus: *Neurog3* (mouse) mapping to 10 B4.

PRODUCT

Neurogenin 3 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 Neurogenin 3 shRNA Plasmid (m): sc-42080-SH and Neurogenin 3 shRNA (m) Lentiviral Particles: sc-42080-V as alternate gene silencing products.

For independent verification of Neurogenin 3 (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-42080A, sc-42080B and sc-42080C.

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 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

Neurogenin 3 siRNA (m) is recommended for the inhibition of Neurogenin 3 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

Neurogenin 3 (C-7): sc-374442 is recommended as a control antibody for monitoring of Neurogenin 3 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 Neurogenin 3 gene expression knockdown using RT-PCR Primer: Neurogenin 3 (m)-PR: sc-42080-PR (20 μ l, 548 bp). 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.