SANTA CRUZ BIOTECHNOLOGY, INC.

INSM1 siRNA (m): sc-72310

BACKGROUND

INSM1 (insulinoma-associated protein 1), also known as zinc finger protein IA-1, is a developmentally regulated zinc finger transcription factor. It localizes to the nucleus and is expressed in embryonic tissues undergoing neuroendocrine differentiation. INSM1 is not expressed in normal adult tissues but it can be found highly expressed in neuroendocrine tumors. INSM1 contains five Cys_2-His_2-type zinc finger DNA binding domains and a prohormone domain. INSM1 acts as a transcriptional repressor of the Neuro D promoter and recruits cyclin D1 as a co-repressor. It plays an important role in neuroendocrine development and is required for normal differentiation of pancreatic endocrine cells. Inhibition of INSM1 results in decreased formation of glucagon and Insulin positive cells. The gene encoding INSM1 is directly regulated by Neurogenin 3 which binds chromatin in the INSM1 promoter region and induces transcription.

REFERENCES

1. Li, Q., et al. 1997. Molecular characterization of the promoter region of a neuroendocrine tumor marker, IA-1. Biochem. Biophys. Res. Commun. 236: 776-781.
2. Breslin, M.B., et al. 2002. Neuroendocrine differentiation factor, IA-1, is a transcriptional repressor and contains a specific DNA-binding domain: identification of consensus IA-1 binding sequence. Nucleic Acids Res. 30: 1038-1045.
3. Xie, J., et al. 2002. The zinc-finger transcription factor INSM1 is expressed during embryo development and interacts with the Cbl-associated protein. Genomics 80: 54-61.
4. Breslin, M.B., et al. 2003. NeuroD1/E47 regulates the E-box element of a novel zinc finger transcription factor, IA-1, in developing nervous system. J. Biol. Chem. 278: 38991-38997.
5. Liu, W.D., et al. 2006. INSM1 functions as a transcriptional repressor of the neuroD/β2 gene through the recruitment of cyclin D1 and histone deacetylases. Biochem. J. 397: 169-177.
6. Mellitzer, G., et al. 2006. IA1 is NGN3-dependent and essential for differentiation of the endocrine pancreas. EMBO J. 25: 1344-1352.

CHROMOSOMAL LOCATION

Genetic locus: Insm1 (mouse) mapping to 2 G1.

PRODUCT

INSM1 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 INSM1 shRNA Plasmid (m): sc-72310-SH and INSM1 shRNA (m) Lentiviral Particles: sc-72310-V as alternate gene silencing products.

For independent verification of INSM1 (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-72310A, sc-72310B and sc-72310C.

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

INSM1 siRNA (m) is recommended for the inhibition of INSM1 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

INSM1 (A-8): sc-271408 is recommended as a control antibody for monitoring of INSM1 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 INSM1 gene expression knockdown using RT-PCR Primer: INSM1 (m)-PR: sc-72310-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.

PROTOCOLS

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