

INSL4 siRNA (h): sc-60856

BACKGROUND

Insulin is a secreted peptide hormone that elicits metabolic effects such as increases in glucose uptake and glycogen synthesis leading to a decrease in blood glucose concentration. Insulin is first formed as a precursor molecule, proinsulin, which is later cleaved to proinsulin and finally to the mature Insulin hormone. Insulin-like peptides (INSL proteins), also designated Relaxin-like factors, are members of the Insulin family, which regulate cell growth, metabolism and tissue-specific functions. INSL1-7 are mostly secreted proteins that are expressed mainly in testis, placenta, uterus or prenatal tissues. INSL4 (insulin-like peptide 4) is a 139 amino acid secreted protein expressed in the placenta, uterus and in fetal perichondrium. It may play an important role in the regulation of bone formation and in trophoblast development.

REFERENCES

1. Chassin, D., et al. 1996. Cloning of a new member of the Insulin gene superfamily (INSL4) expressed in human placenta. *Genomics* 29: 465-470.
2. Bellet, D., et al. 1997. Identification of pro-EPIL and EPIL peptides translated from Insulin-like 4 (INSL4) mRNA in human placenta. *J. Clin. Endocrinol. Metab.* 82: 3169-3172.
3. Laurent, A., et al. 1999. Insulin-like 4 (INSL4) gene expression in human embryonic and trophoblastic tissues. *Mol. Reprod. Dev.* 51: 123-129.
4. Bièche, I., et al. 2003. Placenta-specific INSL4 expression is mediated by a human endogenous element. *Biol. Reprod.* 68: 1422-1429.
5. Faye, A., et al. 2005. Evaluation of the placental environment with a new *in vitro* model of histocultures of early and term placenta: determination of cytokine and chemokine expression profiles. *Placenta* 26: 262-267.
6. Hsu, S.Y., et al. 2005. Evolution of the signaling system in relaxin-family peptides. *Ann. N.Y. Acad. Sci.* 11041: 520-529.
7. Millar, L., et al. 2005. Early placental Insulin-like protein (INSL4 or EPIL) in placental and fetal membrane growth. *Biol. Reprod.* 73: 695-702.
8. Wilkinson, T.N., et al. 2005. Evolution of the Relaxin-like peptide family. *BMC. Evol. Biol.* 5: 14.

CHROMOSOMAL LOCATION

Genetic locus: INSL4 (human) mapping to 9p24.1.

PRODUCT

INSL4 siRNA (h) 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 INSL4 shRNA Plasmid (h): sc-60856-SH and INSL4 shRNA (h) Lentiviral Particles: sc-60856-V as alternate gene silencing products.

For independent verification of INSL4 (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-60856A, sc-60856B and sc-60856C.

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

INSL4 siRNA (h) is recommended for the inhibition of INSL4 expression in human 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

INSL4 (G-2): sc-373728 is recommended as a control antibody for monitoring of INSL4 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 INSL4 gene expression knockdown using RT-PCR Primer: INSL4 (h)-PR: sc-60856-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.