

ENSA siRNA (h): sc-78564

BACKGROUND

ATP-dependent potassium K_{ATP} channels regulate the polarity of the cell membrane, which affects cell metabolism and Insulin secretion. When ATP levels rise in response to an increased rate of glucose metabolism, the K_{ATP} channels close, which stimulates the cells to secrete Insulin. K_{ATP} channels are composed of two structurally unrelated subunits; a Kir6.0 subfamily component and a sulfonylurea receptor (SUR) component. ENSA (α -endosulfine), also known as ARPP-19e, is a 121 amino acid endogenous ligand for SUR. ENSA inhibits the binding of sulfonylurea to the SUR component of the K_{ATP} channel, thereby reducing channel activity and stimulating the secretion of Insulin. ENSA is localized to the cytoplasm and widely expressed in tissues, with high expression in brain and muscle and low expression in pancreas. ENSA is phosphorylated by PKA and exists as 2 isoforms, designated α and β , produced by alternative splicing.

REFERENCES

1. Heron, L., Virsolvy, A., Peyrollier, K., Gribble, F.M., Le Cam, A., Ashcroft, F.M. and Bataille, D. 1998. Human α -endosulfine, a possible regulator of sulfonylurea-sensitive K_{ATP} channel: molecular cloning, expression and biological properties. *Proc. Natl. Acad. Sci. USA* 95: 8387-8391.
2. Héron, L., Virsolvy, A., Apiou, F., Le Cam, A. and Bataille, D. 1999. Isolation, characterization, and chromosomal localization of the human ENSA gene that encodes α -endosulfine, a regulator of β -cell K_{ATP} channels. *Diabetes* 48: 1873-1876.
3. Bataille, D. 2000. Endosulfines: novel regulators of Insulin secretion. *Drug News Perspect.* 13: 453-462.
4. Kim, S.H. and Lubec, G. 2001. Decreased α -endosulfine, an endogenous regulator of ATP-sensitive potassium channels, in brains from adult Down syndrome patients. *J. Neural Transm. Suppl.* 61: 1-9.
5. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 603061. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>

CHROMOSOMAL LOCATION

Genetic locus: ENSA (human) mapping to 1q21.3.

PRODUCT

ENSA siRNA (h) is a target-specific 19-25 nt siRNA 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 ENSA shRNA Plasmid (h): sc-78564-SH and ENSA shRNA (h) Lentiviral Particles: sc-78564-V as alternate gene silencing products.

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.

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

ENSA siRNA (h) is recommended for the inhibition of ENSA 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

ENSA (L7Q): sc-81883 is recommended as a control antibody for monitoring of ENSA 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 ENSA gene expression knockdown using RT-PCR Primer: ENSA (h)-PR: sc-78564-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.