

IDE siRNA (m): sc-146140

BACKGROUND

Insulin degrading enzyme (IDE), initiates the cleavage of Insulin, resulting in Insulin response and resistance. However, IDE also degrades a variety of bioactive peptides, including amyloid- β peptides, implicating IDE in certain age-related changes seen in Alzheimer's disease. Studies show that when the expression of the IDE gene (chromosome 10q23.3) is altered, changes occur not only in glucose homeostasis, but also in the levels of brain A β 40 and A β 42 peptides. An IDE inhibitor, bacitracin, inhibits degradation of both Insulin and amylin, indicating that both are degraded through a common proteolytic pathway. Variations in the rate of proteolysis suggest that the function of IDE exhibits conformational dependence, which may lead to possible therapeutic interventions for diabetes, AD, and other diseases associated with IDE substrate proteolysis.

REFERENCES

1. Seta, K.A., et al. 1997. Overexpression of Insulin degrading enzyme: cellular localization and effects on Insulin signalling. *Biochem. Biophys. Res. Commun.* 231: 167-171.
2. Ling, Y., et al. 2003. Amyloid precursor protein (APP) and the biology of proteolytic processing: relevance to Alzheimer's disease. *Int. J. Biochem. Cell Biol.* 35: 1505-1535.
3. Miller, B.C., et al. 2003. Amyloid- β peptide levels in brain are inversely correlated with insulin activity levels *in vivo*. *Proc. Natl. Acad. Sci. USA* 100: 6221-6226.
4. Bennett, R.G., et al. 2003. An Insulin-degrading enzyme inhibitor decreases amylin degradation, increases amylin-induced cytotoxicity, and increases amyloid formation in Insulinoma cell cultures. *Diabetes* 52: 2315-2320.
5. Song, E.S., et al. 2003. Substrate activation of insulin degrading enzyme (Insulysin): a potential target for drug development. *J. Biol. Chem.* 278: 49789-49794.

CHROMOSOMAL LOCATION

Genetic locus: Ide (mouse) mapping to 19 C2.

PRODUCT

IDE 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 IDE shRNA Plasmid (m): sc-146140-SH and IDE shRNA (m) Lentiviral Particles: sc-146140-V as alternate gene silencing products.

For independent verification of IDE (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-146140A, sc-146140B and sc-146140C.

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

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

IDE (F-9): sc-393887 is recommended as a control antibody for monitoring of IDE 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 IDE gene expression knockdown using RT-PCR Primer: IDE (m)-PR: sc-146140-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Yun, Y.J., et al. 2022. Ginsenoside F1 protects the brain against Amyloid β -induced toxicity by regulating IDE and NEP. *Life* 12: 58.

RESEARCH USE

For research use only, not for use in diagnostic procedures.