SANTA CRUZ BIOTECHNOLOGY, INC.

FN3K siRNA (h): sc-60647

BACKGROUND

Amines, including those present on proteins, spontaneously react with glucose to make fructosamines in a reaction termed glycation. Fructosamine 3-kinase (FN3K), a 309-amino acid enzyme initially identified in erythrocytes, catalyzes the ATP-dependent phosphorylation of the third carbon on both D- and L-fructosamines, leading to their destabilization and eventually, their removal from the protein. FN3K is a monomer that is ubiquitously expressed in mammalian tissue and phosphorylates both low molecular mass and protein-bound fructosamines which are formed as a result of glycation of glucose with primary amines. FN3K protects proteins from the harmful effects of nonenzymatic glycation, and may also be involved in peptide repair and cell metabolism. Abnormal expression of FN3K may lead to diabetic complications.

REFERENCES

1. Delplanque, J., et al. 2004. Tissue distribution and evolution of fructosamine 3-kinase and fructosamine 3-kinase-related protein. J. Biol. Chem. 279: 46606-46613.
2. Conner, J.R., et al. 2005. Some clues as to the regulation, expression, function, and distribution of fructosamine 3-kinase and fructosamine 3-kinase-related protein. Ann. N.Y. Acad. Sci. 1043: 824-836.
3. Fortpied, J., et al. 2005. Plant ribulosamine/erythrulosamine 3-kinase, a putative protein-repair enzyme. Biochem. J. 388: 795-802.
4. Szwergold, B.S., et al. 2005. Intrinsic toxicity of glucose, due to non-enzymatic glycation, is controlled *in vivo* by deglycation systems including: FN3K-mediated deglycation of fructosamines and transglycation of aldosamines. Med. Hypotheses 65: 337-348.
5. Szwergold, B.S., et al. 2005. Transglycation—a potential new mechanism for deglycation of Schiff's bases. Ann. N.Y. Acad. Sci. 1043: 845-864.
6. Delpierre, G., et al. 2006. Variability in E with polymorphisms in the FN3K gene and impacts on haemoglobin glycation at specific sites. Diabetes Metab. 32: 31-39.
7. Monnier, V.M. 2006. The fructosamine 3-kinase knockout mouse: a tool for testing the glycation hypothesis of intracellular protein damage in diabetes and aging. Biochem. J. 399: 11-13.

CHROMOSOMAL LOCATION

Genetic locus: FN3K (human) mapping to 17q25.3.

PRODUCT

FN3K 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see FN3K shRNA Plasmid (h): sc-60647-SH and FN3K shRNA (h) Lentiviral Particles: sc-60647-V as alternate gene silencing products.

For independent verification of FN3K (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-60647A, sc-60647B and sc-60647C.

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

FN3K siRNA (h) is recommended for the inhibition of FN3K 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 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

FN3K (E-9): sc-271503 is recommended as a control antibody for monitoring of FN3K 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 FN3K gene expression knockdown using RT-PCR Primer: FN3K (h)-PR: sc-60647-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.