SANTA CRUZ BIOTECHNOLOGY, INC.

GGCX siRNA (h): sc-75125

BACKGROUND

GGCX (γ-glutamyl carboxylase), also known as GC or VKCFD1 (vitamin K-dependent γ-carboxylase), is a 758 amino acid multi-pass membrane protein. Localized to the membrane of the endoplasmic reticulum, GGCX functions to mediate the vitamin K-dependent carboxylation of glutamate residues on target proteins, thereby producing calcium binding γ-carboxyglutamate (Gla) residues on these proteins and simultaneously converting vitamin K to vitamin K epoxide. GGCX exists as a monomer and, via its ability to modify glutamate residues, it accomplishes the post-translational changes that are necessary for the activity of all vitamin K-dependent proteins (such as blood coagulation and bone matrix proteins). Defects in the gene encoding GGCX are the cause of combined deficiency of vitamin K-dependent clotting factors 1 (VKCFD1) and PXE-like disorder with multiple coagulation factor deficiency, both of which are characterized by abnormal skin, blood or bone function.

REFERENCES

1. Brenner, B., et al. 1998. A missense mutation in γ-glutamyl carboxylase gene causes combined deficiency of all vitamin K-dependent blood coagulation factors. Blood 92: 4554-4559.
2. Lin, P.J., et al. 2004. Binding of the factor IX γ-carboxyglutamic acid domain to the vitamin K-dependent γ-glutamyl carboxylase active site induces an allosteric effect that may ensure processive carboxylation and regulate the release of carboxylated product. J. Biol. Chem. 279: 6560-6566.
3. Rishavy, M.A., et al. 2004. A new model for vitamin K-dependent carboxylation: the catalytic base that deprotonates vitamin K hydroquinone is not Cys but an activated amine. Proc. Natl. Acad. Sci. USA 101: 13732-13737.
4. Wadelius, M., et al. 2005. Common VKORC1 and GGCX polymorphisms associated with warfarin dose. Pharmacogenomics J. 5: 262-270.
5. Tie, J.K., et al. 2006. Identification of the N-linked glycosylation sites of vitamin K-dependent carboxylase and effect of glycosylation on carboxylase function. Biochemistry 45: 14755-14763.
6. Kimura, R., et al. 2006. Polymorphisms in vitamin K-dependent γ-carboxylation-related genes influence interindividual variability in plasma protein C and protein S activities in the general population. Int. J. Hematol. 84: 387-397.
7. Cha, P.C., et al. 2007. High-resolution SNP and haplotype maps of the human γ-glutamyl carboxylase gene (GGCX) and association study between polymorphisms in GGCX and the warfarin maintenance dose requirement of the Japanese population. J. Hum. Genet. 52: 856-864.
8. Sogabe, N., et al. 2007. Nutritional effects of γ-glutamyl carboxylase gene polymorphism on the correlation between the vitamin K status and γ-carboxylation of osteocalcin in young males. J. Nutr. Sci. Vitaminol. 53: 419-425.
9. Schmidt-Krey, I., et al. 2007. Two-dimensional crystallization of human vitamin K-dependent γ-glutamyl carboxylase. J. Struct. Biol. 157: 437-442.

PROTOCOLS

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

CHROMOSOMAL LOCATION

Genetic locus: GGCX (human) mapping to 2p11.2.

PRODUCT

GGCX 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 GGCX shRNA Plasmid (h): sc-75125-SH and GGCX shRNA (h) Lentiviral Particles: sc-75125-V as alternate gene silencing products.

For independent verification of GGCX (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-75125A, sc-75125B and sc-75125C.

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

GGCX siRNA (h) is recommended for the inhibition of GGCX 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor GGCX gene expression knockdown using RT-PCR Primer: GGCX (h)-PR: sc-75125-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.