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GGT1/2 siRNA (h): sc-43802

BACKGROUND

GGT (γ -glutamyltranspeptidase) acts as a glutathionase and catalyzes the transfer of the glutamyl moiety of glutathione to a variety of amino acids and dipeptide acceptors. This enzyme is located on the outer surface of the cell membrane and is widely distributed in mammalian tissues involved in absorption and secretion. In humans, hepatic GGT activity is elevated in some liver diseases. GGT1 is released into the bloodstream after liver damage, and an elevated level of the enzyme may be a useful early sign of hepatocellular carcinoma. GGT5 converts leukotriene C4 to leukotriene D4; it does not, however, convert synthetic substrates that are commonly used to assay GGT. In human serum and in human tissues, there is a marked heterogeneity in GGT, but this heterogeneity can be attributed to different glycosylation of the same peptide rather than to the products of different genes.

REFERENCES

1. Bulle, F., Mattei, M.G., Siegrist, S., Pawlak, A., Passage, E., Chobert, M.N., Laperche, Y. and Guellaen, G. 1987. Assignment of the human γ -glutamyl transferase gene to the long arm of chromosome 22. *Hum. Genet.* 76: 283-286.
2. Heisterkamp, N., Rajpert-De Meyts, E., Uribe, L., Forman, H.J. and Groffen, J. 1991. Identification of a human γ -glutamyl cleaving enzyme related to, but distinct from, γ -glutamyltranspeptidase. *Proc. Natl. Acad. Sci. USA* 88: 6303-6307.
3. Visvikis, A., Thioudellet, C., Oster, T., Fournel-Gigleux, S., Wellman, M. and Siest, G. 1991. High-level expression of enzymatically active mature human γ -glutamyltransferase in transgenic V79 Chinese hamster cells. *Proc. Natl. Acad. Sci. USA* 88: 7361-7365.
4. LocusLink Report (LocusID: 137181). <http://www.ncbi.nlm.nih.gov/LocusLink>

CHROMOSOMAL LOCATION

Genetic locus: GGT2 (human) mapping to 22q11.21.

PRODUCT

GGT1/2 siRNA (h) is a pool of 2 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 GGT1/2 shRNA Plasmid (h): sc-43802-SH and GGT1/2 shRNA (h) Lentiviral Particles: sc-43802-V as alternate gene silencing products.

For independent verification of GGT1/2 (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-43802A and sc-43802B.

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

GGT1/2 siRNA (h) is recommended for the inhibition of GGT1/2 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

GGT1/2 (A-4): sc-393706 is recommended as a control antibody for monitoring of GGT1/2 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 GGT1/2 gene expression knockdown using RT-PCR Primer: GGT1/2 (h)-PR: sc-43802-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.