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SANTA CRUZ BIOTECHNOLOGY, INC.

TGase6 siRNA (m): sc-154239

BACKGROUND

Terminally differentiating mammalian epidermal cells acquire an insoluble, 10 to 20 nm thick protein deposit on the intracellular surface of the plasma membrane, known as the cross-linked cell envelope (CE). The CE is a component of the epidermis that is generated through the formation of disulfide bonds and γ-glutamyl-lysine isodipeptide bonds, which are formed by the action of transglutaminases (TGases). TGases are Ca^{2+}-dependent enzymes, which catalyze the formation of isopeptide bonds by transferring an amine to a glutaminyl residue, thereby cross-linking glutamine residues and lysine residues in substrate proteins. TGases influence numerous biological processes, including blood coagulation, epidermal differentiation, seminal fluid coagulation, fertilization, cell differentiation and apoptosis. TGase6 (transglutaminase 6), also known as TGM6, TGY or TGM3L, is a 706 amino acid protein that catalyzes the cross-linking of proteins and the conjugation of proteins to polyamines. As a result of alternative splicing, two TGase6 isoforms exist.

REFERENCES

1. Yamanishi, K., et al. 1991. Molecular cloning of human epidermal transglutaminase cDNA from keratinocytes in culture. Biochem. Biophys. Res. Commun. 175: 906-913.
2. Gentile, V., et al. 1991. Isolation and characterization of cDNA clones to mouse macrophage and human endothelial cell tissue transglutaminases. J. Biol. Chem. 266: 478-483.
3. Mariniello, L., et al. 1993. Human-immunodeficiency-virus transmembrane glycoprotein gp41 is an amino acceptor and donor substrate for transglutaminase *in vitro*. Eur. J. Biochem. 215: 99-104.
4. Ikura, K., et al. 1995. Site-directed mutation in conserved anionic regions of guinea pig liver transglutaminase. Arch. Biochem. Biophys. 318: 307-313.
5. Ueki, S., et al. 1996. Dual functions of transglutaminase in novel cell adhesion. J. Cell Sci. 109: 2727-2735.
6. Grenard, P., et al. 2001. Evolution of transglutaminase genes: identification of a transglutaminase gene cluster on human chromosome 15q15. Structure of the gene encoding transglutaminase X and a novel gene family member, transglutaminase Z. J. Biol. Chem. 276: 33066-33078.

CHROMOSOMAL LOCATION

Genetic locus: Tgm6 (mouse) mapping to 2 F1.

PRODUCT

TGase6 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see TGase6 shRNA Plasmid (m): sc-154239-SH and TGase6 shRNA (m) Lentiviral Particles: sc-154239-V as alternate gene silencing products.

For independent verification of TGase6 (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-154239A, sc-154239B and sc-154239C.

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

TGase6 siRNA (m) is recommended for the inhibition of TGase6 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 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

TGase6 (E-1): sc-398160 is recommended as a control antibody for monitoring of TGase6 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 TGase6 gene expression knockdown using RT-PCR Primer: TGase6 (m)-PR: sc-154239-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.