

# TGase5 siRNA (h): sc-90196

## 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 formation of disulfide bonds and  $\gamma$ -glutamyl-lysine isodipeptide bonds, which are formed by the action of transglutaminases (TGases). TGases are intercellular localized,  $\text{Ca}^{2+}$ -dependent enzymes, which catalyze the formation of isopeptide bonds by transferring an amine to a glutamyl 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. TGase5 (transglutaminase 5), also known as TGM5 or TGX, is a 720 amino acid cytoplasmic protein that uses calcium to catalyze the cross-linking of proteins and plays an important role in the formation of the cornified cell envelope of keratinocytes. Defects in the gene encoding TGase5 are associated with peeling skin syndrome acral type (APSS), an autosomal recessive disease characterized by the continuous shedding of the outer layers of the epidermis.

## REFERENCES

1. Aeschlimann, D., et al. 1998. Isolation of a cDNA encoding a novel member of the transglutaminase gene family from human keratinocytes. Detection and identification of transglutaminase gene products based on reverse transcription-polymerase chain reaction with degenerate primers. *J. Biol. Chem.* 273: 3452-3460.
2. 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.
3. Candi, E., et al. 2002. Expression of transglutaminase 5 in normal and pathologic human epidermis. *J. Invest. Dermatol.* 119: 670-677.
4. Online Mendelian Inheritance in Man, OMIM<sup>™</sup>. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 603805. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
5. Cadot, B., et al. 2004. Overexpressed transglutaminase 5 triggers cell death. *Amino Acids* 26: 405-408.
6. Rufini, A., et al. 2004. Transglutaminase 5 is acetylated at the N-terminal end. *Amino Acids* 26: 425-430.
7. Cassidy, A.J., et al. 2005. A homozygous missense mutation in TGM5 abolishes epidermal transglutaminase 5 activity and causes acral peeling skin syndrome. *Am. J. Hum. Genet.* 77: 909-917.
8. Thibaut, S., et al. 2005. Transglutaminase 5 expression in human hair follicle. *J. Invest. Dermatol.* 125: 581-585.
9. Pietroni, V., et al. 2008. Inactive and highly active, proteolytically processed transglutaminase-5 in epithelial cells. *J. Invest. Dermatol.* 128: 2760-2766.

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) for detailed protocols and support products.

## CHROMOSOMAL LOCATION

Genetic locus: TGM5 (human) mapping to 15q15.2.

## PRODUCT

TGase5 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  $\mu\text{M}$  solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see TGase5 shRNA Plasmid (h): sc-90196-SH and TGase5 shRNA (h) Lentiviral Particles: sc-90196-V as alternate gene silencing products.

For independent verification of TGase5 (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-90196A, sc-90196B and sc-90196C.

## STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at  $-20^{\circ}\text{C}$  with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at  $-20^{\circ}\text{C}$ , avoid contact with RNases and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330  $\mu\text{l}$  of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu\text{l}$  of RNase-free water makes a 10  $\mu\text{M}$  solution in a 10  $\mu\text{M}$  Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

TGase5 siRNA (h) is recommended for the inhibition of TGase5 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  $\mu\text{M}$  in 66  $\mu\text{l}$ . 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.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor TGase5 gene expression knockdown using RT-PCR Primer: TGase5 (h)-PR: sc-90196-PR (20  $\mu\text{l}$ ). Annealing temperature for the primers should be  $55-60^{\circ}\text{C}$  and the extension temperature should be  $68-72^{\circ}\text{C}$ .

## RESEARCH USE

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