

# TRIM5 siRNA (h): sc-61718

## BACKGROUND

TRIM5 is a 493 amino acid member of the large tripartite motif protein (TRIM) family. TRIM proteins are composed of three zinc-binding domains, a RING, a B-box 2 and a coiled-coil domain, and they use homomultimerization to identify different cell compartments. Some TRIM proteins, such as TRIM5, also possess a carboxy-terminal B30.2 (SPRY) domain and localize to the cytoplasm. Isoform alpha of TRIM5 mediates innate intracellular retroviral resistance, which is dependent on its carboxy-terminal domain. The three variable regions of the B30.2 domain form loops on one side of the B30.2 core structure of TRIM5 which may form a binding surface for the virus. Isoform  $\alpha$  TRIM5 trimerization plays a major role in its affinity for the retroviral capsid, and in its ability to inhibit virus infection. The linker region between the coiled-coil and B30.2 domains of TRIM5 $\alpha$  is required for this trimerization. TRIM5 $\alpha$  blocks infection after the virus has entered the cell.

## REFERENCES

1. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 608487. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
2. Song, B., et al. 2005. The B30.2 (SPRY) domain of the retroviral restriction factor TRIM5 $\alpha$  exhibits lineage-specific length and sequence variation in primates. *J. Virol.* 79: 6111-6121.
3. Berthou, L., et al. 2005. Disruption of human TRIM5 $\alpha$  antiviral activity by nonhuman primate orthologues. *J. Virol.* 79: 7883-7888.
4. Javanbakht, H., et al. 2005. The contribution of RING and B-box 2 domains to retroviral restriction mediated by monkey TRIM5 $\alpha$ . *J. Biol. Chem.* 280: 26933-26940.
5. Nakayama, E.E., et al. 2005. A specific region of 37 amino acid residues in the SPRY (B30.2) domain of African green monkey TRIM5 $\alpha$  determines species-specific restriction of simian immunodeficiency virus SIVmac infection. *J. Virol.* 79: 8870-8877.
6. Perez-Caballero, D., et al. 2005. Human tripartite motif 5 $\alpha$  domains responsible for retrovirus restriction activity and specificity. *J. Virol.* 79: 8969-8978.

## CHROMOSOMAL LOCATION

Genetic locus: TRIM5 (human) mapping to 11p15.4.

## PRODUCT

TRIM5 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see TRIM5 shRNA Plasmid (h): sc-61718-SH and TRIM5 shRNA (h) Lentiviral Particles: sc-61718-V as alternate gene silencing products.

For independent verification of TRIM5 (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-61718A, sc-61718B and sc-61718C.

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

## APPLICATIONS

TRIM5 siRNA (h) is recommended for the inhibition of TRIM5 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  $\mu$ M in 66  $\mu$ 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

TRIM5 (D-6): sc-373864 is recommended as a control antibody for monitoring of TRIM5 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 $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  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 $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  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 TRIM5 gene expression knockdown using RT-PCR Primer: TRIM5 (h)-PR: sc-61718-PR (20  $\mu$ l). 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](http://www.scbt.com) for detailed protocols and support products.