

# RhCG siRNA (h): sc-90218

## BACKGROUND

RhCG (Rhesus blood group family type C glycoprotein), also known as RHGK (Rh glycoprotein kidney), PDRC2 (tumor-related protein DRC2) or SLC42A3, is a multi-pass membrane protein with three potential N-glycosylation sites that belongs to the Rh subfamily of the ammonium transporter family. Expressed in a wide variety of tissues with predominant expression in adult and fetal kidney, RhCG localizes to the apical and/or basolateral cell membrane (depending on the species) and is believed to function as a non-erythroid, bidirectional, electroneutral ammonium transporter. Specifically, RhCG is found throughout the kidney in sites, such as the CNT (connecting segment), the DCT (distal convoluted tubule), the collecting duct and the ICT (initial collecting tubule), that are major contributors to renal ammonia secretion. This suggests that RhCG plays an important role in renal ammonia metabolism.

## REFERENCES

1. Liu, Z., et al. 2000. Characterization of human RhCG and mouse Rhcg as novel nonerythroid Rh glycoprotein homologues predominantly expressed in kidney and testis. *J. Biol. Chem.* 275: 25641-25651.
2. Heitman, J. and Agre, P. 2000. A new face of the Rhesus antigen. *Nat. Genet.* 26: 258-259.
3. Marini, A.M., et al. 2000. The human Rhesus-associated RhAG protein and a kidney homologue promote ammonium transport in yeast. *Nat. Genet.* 26: 341-344.
4. Online Mendelian Inheritance in Man, OMIM<sup>™</sup>. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 605381. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
5. Khademi, S., et al. 2004. Mechanism of ammonia transport by Amt/MEP/Rh: structure of AmtB at 1.35 Å. *Science* 305: 1587-1594.
6. Han, K.H., et al. 2006. Expression of the ammonia transporter, rh C glycoprotein, in normal and neoplastic human kidney. *J. Am. Soc. Nephrol.* 17: 2670-2679.
7. Callebaut, I., et al. 2006. Hydrophobic cluster analysis and modeling of the human Rh protein three-dimensional structures. *Transfus. Clin. Biol.* 13: 70-84.

## CHROMOSOMAL LOCATION

Genetic locus: RHCG (human) mapping to 15q26.1.

## PRODUCT

RhCG 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 RhCG shRNA Plasmid (h): sc-90218-SH and RhCG shRNA (h) Lentiviral Particles: sc-90218-V as alternate gene silencing products.

For independent verification of RhCG (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-90218A, sc-90218B and sc-90218C.

## 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

RhCG siRNA (h) is recommended for the inhibition of RhCG 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

RhCG (CB-62): sc-100287 is recommended as a control antibody for monitoring of RhCG 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<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> 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<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor RhCG gene expression knockdown using RT-PCR Primer: RhCG (h)-PR: sc-90218-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.