

# Factor V siRNA (h): sc-40399

## BACKGROUND

Hemostasis following tissue injury involves the deployment of essential plasma procoagulants (prothrombin, and factors X, IX, V, and VIII), which are involved in a blood coagulation cascade that leads to the formation of insoluble fibrin clots and the promotion of platelet aggregation. Coagulation Factor V (Factor V, FV, proaccelerin, labile factor) is a 2,196 amino acid, single chain glycoprotein that is cleaved by thrombin to yield an active,  $\text{Ca}^{2+}$  dependent dimer. This heterodimer is essential to the blood coagulation cascade. Together with catalytic Factor Xa and  $\text{Ca}^{2+}$  on the surface of platelets or endothelial cells, Factor Va coordinates in a prothrombinase complex, which mediates proteolysis of prothrombin into active thrombin. Due to both the procoagulant properties of Factor V in coordinating proteolytic activation of thrombin, and anticoagulant properties as a cofactor to activated protein C (APC), which selectively destroys FVa and FXa, alterations at the Factor V locus can contribute to hemorrhagic diathesis or thrombosis, respectively.

## REFERENCES

1. Davie, E.W. and Fujikawa, K. 1975. Basic mechanisms in blood coagulation. *Annu. Rev. Biochem.* 44: 799-829.
2. Kane, W.H. and Davie, E.W. 1986. Cloning of a cDNA coding for human Factor V, a blood coagulation Factor homologous to Factor VIII and ceruloplasmin. *Proc. Natl. Acad. Sci. USA* 83: 6800-6804.
3. Jenny, R.J., et al. 1987. Complete cDNA and derived amino acid sequence of human Factor V. *Proc. Natl. Acad. Sci. USA* 84: 4846-4850.
4. Davie, E.W., et al. 1991. The coagulation cascade: initiation, maintenance and regulation. *Biochemistry* 30: 10363-10370.
5. Rand, M.D., et al. 1994. Platelet coagulation Factor Va: the major secretory platelet phosphoprotein. *Blood* 83: 2180-2190.
6. Macedo-Ribeiro, S., et al. 1999. Crystal structures of the membrane-binding C2 domain of human coagulation Factor V. *Nature* 402: 434-439.
7. Online Mendelian Inheritance in Man, OMIM<sup>™</sup>. 2001. Johns Hopkins University, Baltimore, MD. MIM Number: 227400. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>

## CHROMOSOMAL LOCATION

Genetic locus: F5 (human) mapping to 1q24.2.

## PRODUCT

Factor V 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 Factor V shRNA Plasmid (h): sc-40399-SH and Factor V shRNA (h) Lentiviral Particles: sc-40399-V as alternate gene silencing products.

For independent verification of Factor V (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-40399A, sc-40399B and sc-40399C.

## 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

Factor V siRNA (h) is recommended for the inhibition of Factor V 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.

## GENE EXPRESSION MONITORING

Factor V (6A5): sc-13512 is recommended as a control antibody for monitoring of Factor V 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 Factor V gene expression knockdown using RT-PCR Primer: Factor V (h)-PR: sc-40399-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.

## PROTOCOLS

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