

# Factor IX siRNA (h): sc-40403

## 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 IX (plasma thromboplastic component, F9, F.IX, HEMB) is a vitamin K-dependent, single chain serine protease that is synthesized in the liver and circulates as an inactive precursor. Factor XI A-mediated proteolytic cleavage of Factor IX generates Factor IX A, an active serine protease composed of a 145 amino acid light chain and a 236 amino acid catalytic heavy chain, linked through disulfide bonds. Genetic alterations at the Factor IX locus such as point mutations, insertions and deletions, can lead to hemophilia B, also known as Christmas disease.

## REFERENCES

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3. Jaye, M., de la Salle, H., Schamber, F., Bolland, A., Kohli, V., Findeli, A., Tolstoshev, P. and Lecocq, J.P. 1983. Isolation of a human antihemophilic Factor IX cDNA clone using a unique 52-base synthetic oligonucleotide probe deduced from the amino acid sequence of bovine Factor IX. *Nucleic Acids Res.* 11: 2325-2335.
4. Davie, E.W., Fujikawa, K. and Kiesel, W. 1991. The coagulation cascade: initiation, maintenance and regulation. *Biochemistry* 30: 10363-10370.
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6. Online Mendelian Inheritance in Man, OMIM<sup>™</sup>. 2001. Johns Hopkins University, Baltimore, MD. MIM Number: 306900. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>

## CHROMOSOMAL LOCATION

Genetic locus: F9 (human) mapping to Xq27.1.

## PRODUCT

Factor IX 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 Factor IX shRNA Plasmid (h): sc-40403-SH and Factor IX shRNA (h) Lentiviral Particles: sc-40403-V as alternate gene silencing products.

For independent verification of Factor IX (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-40403A, sc-40403B and sc-40403C.

## 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

Factor IX siRNA (h) is recommended for the inhibition of Factor IX 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

Factor IX (B-3): sc-377187 is recommended as a control antibody for monitoring of Factor IX 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 IX gene expression knockdown using RT-PCR Primer: Factor IX (h)-PR: sc-40403-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.