

# Factor VIII (D-15): sc-27651

## BACKGROUND

Factor VIII is a glycoprotein cofactor that serves as a critical component in the blood coagulation pathway. Insufficient expression levels or expression of nonfunctional Factor VIII results in hemophilia A, a common severe hereditary bleeding disorder. In the liver, the main site of factor VIII synthesis, the mature polypeptide chain of 2,332 amino acids is secreted into the lumen of the endoplasmic reticulum, where it interacts with various chaperone proteins, including Calreticulin, Calnexin and IgG-binding protein. From the lumen, a portion of Factor VIII translocates to the Golgi and undergoes activation via proteolysis of both the heavy and light chain portions of the protein into three fragments. Finally, proteolysis of activated Factor VIII by Factor Xa, Protein C or Thrombin results in inactivation of Factor VIII. Survival of Factor VIII in the bloodstream requires binding to von Willebrand factor (VWF) at both the amino- and carboxy-termini of the light chain. Point mutations occurring in those binding domains as well as at other active sites of Factor VIII likely underly 90-95% of disease cases.

## REFERENCES

1. Fulcher, C.A., et al. 1983. Thrombin proteolysis of purified Factor VIII: Correlation of activation with generation of a specific polypeptide. *Blood* 61: 807-811.
2. Eaton, D., et al. 1986. Proteolytic processing of human Factor VIII: Correlation of specific cleavages by Thrombin, Factor Xa, and activated protein C with activation and inactivation of Factor VIII coagulant activity. *Biochemistry* 25: 505-512.
3. Foster, P.A., et al. 1989. Factor VIII structure and function. *Blood Rev.* 3:180-191.
4. Kaufman, R.J. 1992. Biological regulation of Factor VIII activity. *Annu. Rev. Med.* 43: 325-339.
5. Saenko, E.L., et al. 1997. The acidic region of the Factor VIII light chain and the C2 domain together form the high affinity binding site for von Willebrand Factor. *J. Biol. Chem.* 272: 18007-18014.
6. Bhopale, G.M., et al. 2003. Blood coagulation Factor VIII: An overview. *J. Biosci.* 28: 783-789.

## CHROMOSOMAL LOCATION

Genetic locus: F8 (human) mapping to Xq28; F8 (mouse) mapping to X A7.3.

## SOURCE

Factor VIII (D-15) is an affinity purified goat polyclonal antibody raised against a peptide mapping near the C-terminus of Factor VIII of human origin.

## PRODUCT

Each vial contains 200 µg IgG in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

Blocking peptide available for competition studies, sc-27651 P, (100 µg peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% BSA).

## APPLICATIONS

Factor VIII (D-15) is recommended for detection of precursor and mature light chain Factor VIII of mouse, rat and human origin by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000), immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000); non cross-reactive with heavy chain Factor VIII.

Factor VIII (D-15) is also recommended for detection of precursor and mature light chain Factor VIII in additional species, including equine, canine, bovine, porcine and avian.

Suitable for use as control antibody for Factor VIII siRNA (h): sc-43756, Factor VIII siRNA (m): sc-44757, Factor VIII shRNA Plasmid (h): sc-43756-SH, Factor VIII shRNA Plasmid (m): sc-44757-SH, Factor VIII shRNA (h) Lentiviral Particles: sc-43756-V and Factor VIII shRNA (m) Lentiviral Particles: sc-44757-V.

Molecular Weight of Factor VIII heavy chain: 200 kDa.

Molecular Weight of Factor VIII light chain: 80 kDa.

Molecular Weight of Factor VIII cleaved fragments: 50/43/73 kDa.

Positive Controls: rat liver extract: sc-2395, mouse liver extract: sc-2256 or mouse kidney extract: sc-2255.

## RECOMMENDED SECONDARY REAGENTS

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

## STORAGE

Store at 4° C, \*\*DO NOT FREEZE\*\*. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

## 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) or our catalog for detailed protocols and support products.



Try **Factor VIII light chain (RFFVIII C/5): sc-59512** or **Factor VIII (R8B12): sc-73597**, our highly recommended monoclonal alternatives to Factor VIII (D-15). Also, for AC, HRP, FITC, PE, Alexa Fluor® 488 and Alexa Fluor® 647 conjugates, see **Factor VIII light chain (RFFVIII C/5): sc-59512**.