HYPHEN BioMed
ZAC Neuville Université – 155 rue d'Eragny
95000 Neuville-sur-Oise (France)
Tel : +33.1.34.40.65.10
Fax : +33.1.34.48.72.36
www.hyphen-biomed.com

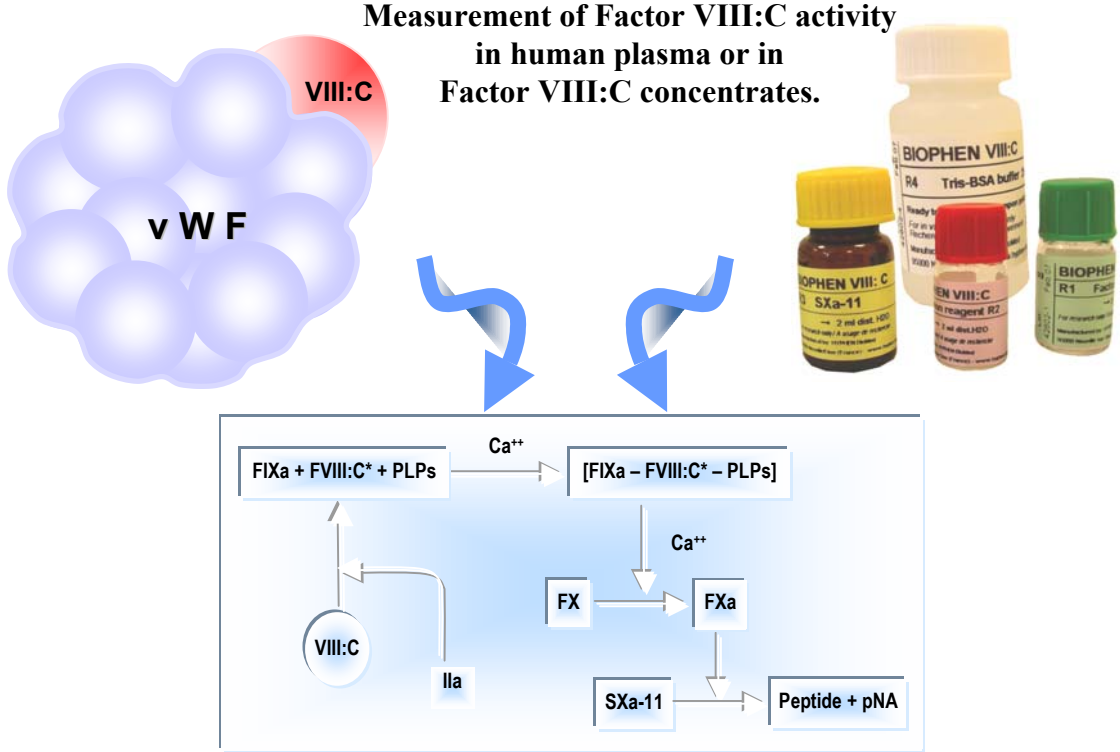
BIOPHEN VIII:C

Factor VIII:C Chromogenic assay

Ref. 221402

A reference method, which can be used with manual or automated techniques.

Measurement of Factor VIII:C activity in human plasma or in Factor VIII:C concentrates.



Nota : FVIII:C*: Thrombin activated FVIII:C

- **A fully homogeneous assay, using only highly purified human proteins.**
- **Designed with highly characterized synthetic phospholipids.**
- **Complies with European Pharmacopea Recommendations.**
- **Large excess of Factor X (< 40 % converted to Xa during the assay time).**
- **Offers a high (concentrates and high factor VIII:C concentrations in plasma) and a low working range (Von Willebrand disease and haemophilia A).**

BIOPHEN VIII:c
(Ref. 221402)

Chromogenic assay of Factor VIII:C activity in plasma or concentrates

Assay principle

- BIOPHEN Factor VIII:C is a chromogenic assay for testing the cofactor activity of Factor VIII:C.
- When activated by thrombin, Factor VIII:C forms an enzymatic complex with Factor IXa, phospholipids and Calcium, which activates Factor X to Factor Xa.
- In presence of a constant amount of Factor IXa, Phospholipids (PLPs) and Calcium, thrombin activated Factor VIII:C forms an enzymatic complex, which activates Factor X, supplied in the assay at a constant concentration and in excess, to Factor Xa. This activity is directly related to the amount of Factor VIII:C, which is the limiting factor in presence of a constant and in excess amount of Factor IXa. Generated Factor Xa is then exactly measured by its activity on a specific Factor Xa chromogenic substrate (SXa-11). Factor Xa cleaves the substrate and releases pNA. The amount of pNA generated is directly proportional to the Factor Xa activity.
- Finally, there is a direct relationship between the amount of Factor VIII:C in the assayed sample and the Factor Xa activity generated, measured by the amount of pNA released, determined by colour development at 405nm.

Kit presentation

R1: Reagent 1: Human Factor X
Human Factor X, lyophilised in presence of a fibrin polymerisation inhibitor; 2 vials**.**

R2: Reagent 2: Activation Reagent
Factor IXa (human), containing human thrombin, calcium and synthetic phospholipids, lyophilised; 2 vials.

R3: Reagent 3: SXa-11
Chomogenic substrate, specific for Factor Xa (SXa-11), lyophilised.
2 vials containing 6 mg of SXa-11 with a thrombin inhibitor.

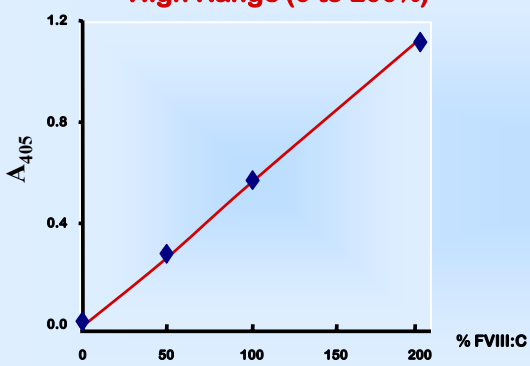
R4 : Reagent 4: Tris-BSA Buffer
Tris-BSA Buffer, ready to use, containing sodium azide (NaN3).
4 vials of 25 mL.

Procedure

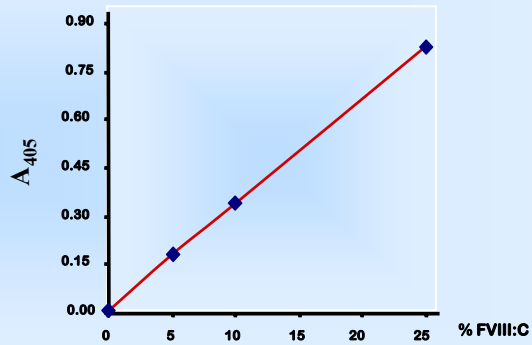
- Specimen: citrated human plasma, or factor VIII:C concentrates.
- Plasma Dilution: High range → 1:40; Low range → 1:10.
- Concentrates' Dilution: predilution to 50 – 200 %, then → 1 : 40.
- Calibration: factor VIII:C calibrator or standard.
- End-point method or kinetics protocols.

Calibration Curves

High Range (0 to 200%)



Low Range (0 to 25%)



Assay Characteristics

- Total assay time : **10 minutes** or below
- Assay range : **0 to 200 %** for high range
  0 to 25 % for low range
- Reproducibility: **< 2 %**
- Detection limit : **about 0.5 %** for low range
- Specificity : Factor VIII: C deficient plasma **< 1 %**
- Adaptations to automatic instruments available
- Can be used with microplate methods

Other Products

- Factor VIII deficient plasma (# DP040A)
- VWF deficient plasma (# DP150A)
- ZYMUTEST vWF (# RK030A)

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