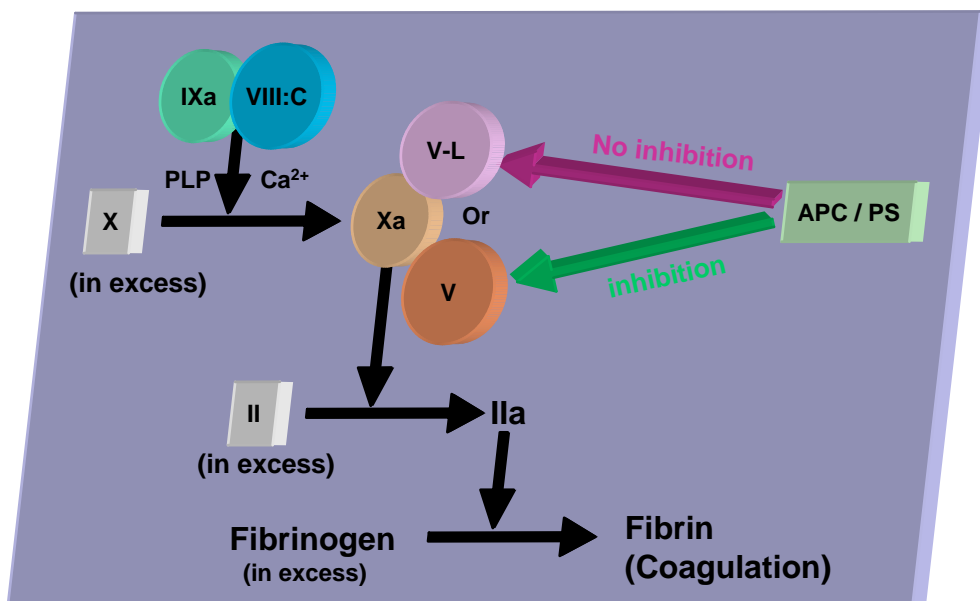


Measuring the APC Resistance associated with Factor V-L

HEMOCLOT Factor V-L kit is a clotting method proposed for testing the presence of Factor V-L (Factor V Leiden) in citrated plasma, by its resistance to the action of Activated Protein C (APC). The assay is performed in the absence or presence of Activated Protein C. In presence of APC, the prolongation of clotting time is directly related to the concentration of Normal Factor V, and inversely related to the amount of Factor V-Leiden (mutation R506Q).

Clinical background

The Factor V-L is insensitive to the action of Activated Protein C. Its presence induces a prolonged coagulant activity in blood (due to a prolonged survival of Factor Va activity), following activation of coagulation. Patients with Factor V-L (mutation R506Q), are exposed to an increased thrombotic risk. This risk, already present in heterozygous patients, where the Normal Factor V and Factor V-L are both present, is strongly enhanced in homozygous patients, who only have Factor V-L.



Assay characteristics

- Quantitative reagent for measuring Factor V-L.
- No interference of plasma factor deficiencies (Other than that of Factor V).
- Excellent discrimination between Heterozygous, Homozygous and Normals.
- Two Clotting Times (with (CT2) or without (CT1) APC), and calculation of ratio (CT/CT1).

Adaptations on instruments available upon request

Apr 2006/REF 020

Form AH12
04-2007

HEMOCLOT Factor V-L

(Ref. ACK061K / ACK061L)

Kit composition

Reagent 1: R1

Clotting mixture:

Fibrinogen/Prothrombin/rec FVIII:C/PS

Reagents 2: R2A & R2B

Purified human FX (with and without APC)

Reagent 3: R3

Purified human FIXa

Assay principle

The **HEMOCLOT Factor V-L** kit is a clotting method, triggered by purified Factor IXa with Phospholipids and Calcium, performed on the diluted tested plasma, in presence or absence of Activated Protein C. In the first step, the diluted plasma is mixed with purified clotting Factors (Prothrombin and Fibrinogen), in a constant and optimised concentration. Then, the purified Factor X, also in a constant and optimised concentration, is added, without (Clotting Time 1) or with (Clotting Time 2) Activated Protein C (APC). Clotting is initiated by the addition of Factor IXa, in presence of Phospholipids (PLP) and Calcium (Ca^{2+}). Clotting times are then recorded. The ratio of Clotting Times without or with APC (Clotting Time 2 / Clotting Time 1) is calculated. If the plasma is normal, this ratio is ≥ 2.00 , if the plasma is from a patient with the R506Q mutation (Factor V Leiden), this ratio is lowered and ≤ 1.80 .

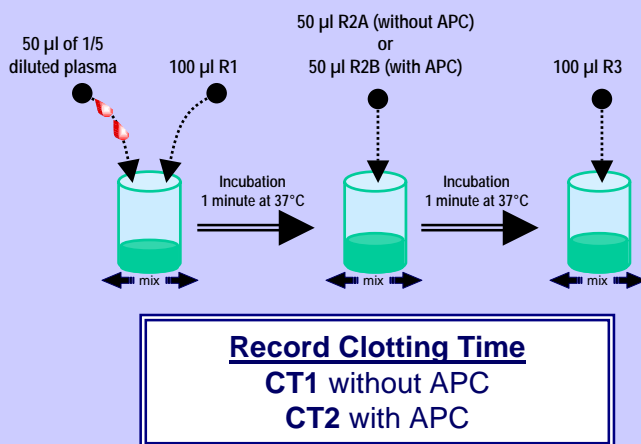
Results

The ratio of the Clotting Times obtained, with or without APC, allows measuring the sensitivity of Factor V, in the tested specimen, to the action of Activated Protein C.

- Normal plasma, containing **normal Factor V**, yields a ratio: **$\text{CT2}/\text{CT1} \geq 2.00$**
- Plasmas from patients carrying the R506Q mutation of Factor V, (**Factor V-L**) yield a ratio: **$\text{CT2}/\text{CT1} \leq 1.80$** .
- **$1.80 < \text{CT2}/\text{CT1} < 2.00 \Rightarrow ??$**

Molecular biology allows confirming the diagnosis, and classifying patients as heterozygous or homozygous.

Protocole



HEMOCLOT Factor V-L

an easy to use and reliable assay, fully automatisable,
designed with purified human factors.

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