

TEST: FACTOR V LEIDEN MUTATION AT POSITION 506 (FV)

PRINCIPLE:

Several genetic mutations are associated with an increased risk of thromboembolic complications. Certain patients with recurrent venous thrombosis can not be anti-coagulated with activated protein C (APC) in spite of the fact that they have normal levels of protein S and no other detectable abnormalities in the coagulation cascade. This was due to a mutation in the substrate for APC, factor V (FV). The APC resistant phenotype is associated with heterozygosity or homozygosity for a single point mutation at nucleotide position 1691, G/A substitution, in the FV gene (the factor V Leiden mutation). This mutation occurs in the putative APC binding site and predicts the replacement of Arg506 (CGA) by Gln (CAA) resulting in FVQ506 or FV Leiden. This FV Leiden mutation is the most common defect associated with increased risk of recurrent venous thrombosis and can be detected by molecular analysis. Specific amplification of a 267-bp DNA fragment of the factor V gene surrounding nucleotide 1691 by PCR preceded by digestion with Mnl I yields three fragments of 163-, 67-, and 37-bp for normal alleles and two fragments of 200- and 67-bp for mutant alleles which can be visualized by gel electrophoresis. The FV Leiden mutation predisposes individuals to thrombosis and may be an important risk factor for obstetrical complications associated with abnormalities in maternal-fetal circulation. The factor V Leiden mutation is more prevalent in women with severe pre-eclampsia, abruptio placentae, fetal growth retardation, and stillbirth.

SPECIMEN REQUIREMENTS:

Collect two to three tubes of blood in EDTA (lavender top) tubes. Heparinized blood cannot be used. Specimen should be delivered to the laboratory immediately, within 24 hours. Store or send at room temperature. Peripheral blood specimens that are clotted, have not been collected in EDTA, or frozen are not acceptable.

METHOD: Polymerase Chain Reaction (PCR)

REFERENCES:

1. Bertina, R. M., B. P. C. Koeleman, T. Koster, F. R. Rosendaal, R. J. Dirven, H. de Ronde, P. A. van der Velden, and P. H. Reitsma. 1994 Mutation in blood coagulation factor V associated with resistance to activated protein C. *Nature* 369:64-67.
2. Brenner, B., A. Zivelin, N. Lanir, J. S. Greengard, J. H. Griffin, and U. Seligsohn. 1996 Venous thromboembolism associated with double heterozygosity for R506Q mutation of factor V and for T298M mutation of protein C in a large family of a previously described homozygous protein C-deficient newborn with massive thrombosis. *Blood* 88:877-880.
3. Kupferminc, M. J., A. Eldor, N. Steinman, A. Many, A. BarAm, A. Jaffa, G. Fait, J. B. Lessing. 1999 Increased frequency of genetic thrombophilia in women with complications of pregnancy. *N. Engl. J. Med.* 340:9-13.

Normal Range: Reported as FV Normal, FV Heterozygous, or FV Homozygous

FV Normal – 163-, 67-, & 37-bp fragments

FV Heterozygous – 200-, 163-, 67-, & 37-bp fragments

FV Homozygous – 200- & 67-bp fragments

Turnaround time: Two weeks