Factor II G20210A Genotyping Assay: Genetics and Clinical Overview

Test Information (v1_020112):
Factor II G20210A Genotyping Assay (CMGDL test code 4302)
For sample collection, transport and testing information refer to the CMGDL website www.medgen.med.miami.edu
See the link for additional test ordering information such as CPT codes, test methodology and limitations.

Background Information
Thrombosis is the leading cause of death worldwide affecting 1 in 1000 individuals annually. Venous thrombosis is triggered by venous stasis, endothelial abnormalities and susceptibility genetic factors. Thrombophilia is defined as the tendency in a disturbance of the plasma coagulation system, and categorized into acquired and inherited. In acquired thrombophilia the abnormal clotting is usually related to a condition that leads to an increased clotting tendency such as recent surgery, trauma, prolonged immobility, pregnancy, obesity, cancer, inflammatory bowel disease, presence of antiphospholipid antibodies, or the use of oral contraceptives or hormone replacement therapy. Inherited Thrombophilia is associated with congenital predisposition factors such as Factor V Leiden mutation, Prothrombin (Factor II) mutation, Methylene tetrahydrofolate Reductase (MTHFR) mutation, Protein C deficiency, Protein S deficiency, and Antithrombin III deficiency (Heit, 2007, Dahblak, 2008).

Factor V Leiden, Factor II 2010G>A, and Methylene tetrahydrofolate Reductase (MTHFR) (667C>T and 1298A>C) mutations are the most common cause of predisposition to inherited thrombophilia (Rosendaal, 1998).

Factor II related Thrombophilia

Background Information
The F2 gene encodes for the coagulation Factor II (prothrombin). The gene encompasses a genomic region of over 21 kb, located on chromosome 11p11-q12. The prothrombin (or Factor II) is a 70 kD glycoprotein activated to thrombin by factor Xa in combination with phospholipids, calcium, and factor Va. The activated thrombin plays an important role in homeostasis and thrombosis, especially by converting fibrinogen to fibrin (Lancellotti, 2009, Degen 1990, Bocchini, 2011).

The Factor II 2010 G>A mutation is located at the 3'-end of the F2 gene and leads to accumulation of prothrombin in plasma because of enhanced mRNA processing. About 25% higher than average levels of prothrombin are expressed by Factor II 2010 G>A heterozygote (Poort, 1996), leading to increased thrombin generation. The most common clinical manifestation of Factor II related Thrombophilia is venous thromboembolism (VTE).

Factor II Mutation Nomenclature Notes
The designation of 2010 seems to be based on a historical reference sequence. The F2 gene 2010G>A (coloquial nomenclature) should be described as g.21538G>A (AF478696.1), c.*97G>A being the mutation located 97 nucleotides downstream of the stop codon.

Factor II mutation frequency
Heterozygosity for Factor II 2010G>A is the second most common inherited thrombophilia (after Factor V Leiden). The prevalence of 2010G>A heterozygosity varies among populations with a frequency of 1.7%-3% of the general US and European populations. This allele is extremely rare in Asian, African, and Native American populations (Rosendaal, 1998).

Clinical Significance

Clinical Diagnosis
Prothrombin-related Thrombophilia is characterized by venous thromboembolism (VTE) but no clinical features are specific. Its diagnosis requires 2010G>A mutation analysis. Legs deep-vein thrombosis and pulmonary embolism are the most common manifestations in adults. Disease expressivity is variable and penetrance incomplete as many F2 2010G>A heterozygous or homozygous individuals may never develop thrombosis. Whereas most heterozygotes who develop thrombotic complications remain asymptomatic until adulthood, recurrent thromboembolism may present in some individuals before age 30 years.

Molecular Genetics and disease risk
Prothrombin-related Thrombophilia has an autosomal dominant inheritance. Relative risk for a first episode of VTE in asymptomatic individuals with a 2010G>A heterozygous allele is increased 2- to 5-fold. Homozygosity for the 2010G>A mutation carries a higher risk for thrombosis than heterozygosity although its entity is not well defined.
The risk is higher for individuals from families with a strong history of VTE than in unselected individuals identified by population screening, and substantially increased by the presence of additional inherited or acquired thrombophilic disorders or conditions such as coexistence of Factor V Leiden, Protein S deficiency (Tirado, 2001), antiphospholipid antibodies, hyperhomocysteinemia (De Stefano, 2001), malignancy, pregnancy, oral contraceptive use and hormone replacement therapy. Risk for VTE increases after travel in individuals heterozygote for 20210G>A. Risk for pregnancy loss may increase in presence of prothrombin-related Thrombophilia (Kujovich, 2011).

**Laboratory Diagnosis**

Measurement of plasma concentration of prothrombin is not reliable for diagnosis of prothrombin-related thrombophilia, since prothrombin concentrations in heterozygote individuals overlap significantly with the normal range. DNA analysis of the F2 gene to identify the common 20210G>A mutation is required to diagnose prothrombin-related Thrombophilia.

**Patient management**

Patient management depends on the clinical circumstances and overall risk for Thrombophilia. Long term anticoagulation is not routinely recommended for asymptomatic 20210G>A heterozygotes, in absence of history of thrombosis. Prophylactic anticoagulation should be considered in high-risk settings such as surgery, pregnancy, or prolonged immobilization, although no current evidence confirms a benefit in 20210G>A heterozygote individuals. Women that are either heterozygous or homozygous for the Factor II 20210G>A mutation with a history of VTE should avoid estrogen-based contraception and hormone replacement therapy.

Patients heterozygous for the 20210G>A mutation should be tested for other inherited or acquired Thrombophilia causes including
- Activated Protein C resistance test or Factor V Leiden DNA analysis;
- Anticardiolipin antibodies and anti-B2 glycoproteinI antibodies;
- Multiple phospholipid-dependent coagulation assays for a lupus inhibitor.

Evaluation of high risk individuals should also include
- Protein C activity;
- Antithrombin activity;
- Protein S activity or free protein S antigen.
(Kujovich, 2011).
- Homocysteinemia levels

**Indications for Testing**

**Introductory Considerations**

The diagnosis of prothrombin-related Thrombophilia requires 20210G>A mutation analysis. This condition is suspected in individuals with a history of VTE that manifested as deep vein thrombosis or pulmonary embolism, especially with a history of VTE during pregnancy or associated with use of oral contraceptives. It is also suspected in individuals with a personal or family history of recurrent thrombosis at young age.

**Appropriate 20210G>A testing circumstances**

- A first unprovoked VTE before age 50 years;
- A history of recurrent VTE;
- Venous thrombosis at unusual sites such as the cerebral, mesenteric, portal, or hepatic veins;
- VTE during pregnancy or the puerperium;
- VTE associated with the use of estrogen-containing oral contraceptives or hormone replacement therapy;
- A first VTE at any age in an individual with a first-degree family member with a VTE before age 50 years;

**Testing may also be considered in case of**

- Asymptomatic adult family members of probands with heterozygous or homozygous 20210G>A mutation, especially those with a strong family history of VTE at a young age;
- Asymptomatic female family members of probands with known prothrombin-related thrombophilia who are pregnant or considering estrogen contraception or pregnancy;

(For a comprehensive list of circumstances, see [www.genetests.org](http://www.genetests.org)).

**Factor II testing is not recommended as:**

- General population screening or routine test;
- Prenatal or newborn testing;
- Prenatal testing is not performed by the CMGDLP

**Related Tests** (visit our website at [www.medgen.med.miami.edu](http://www.medgen.med.miami.edu/))

- Thrombophilia Risk Genotyping Assay (CMGDLP test code 4002)
- Factor II G20210A and Factor V Leiden Genotyping Assay (CMGDLP test code 4102)
- Factor V Leiden Genotyping Assay (CMGDLP test code 4202)
- MTHFR Genotyping Assay (CMGDLP test code 4402)
References