

# Rapid thrombophilia genetic test facilitates improved prenatal care for mother and child

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## Abstract

Physiological changes in coagulation factors during pregnancy are important to minimise blood loss during gestation and delivery, but may also lead to a 4-6 fold increased risk of venous thromboembolism during pregnancy and after delivery. Approximately 25% of maternal mortality can be ascribed to thromboembolism if untreated, while this figure is reduced to less than 1% when diagnosed on time. Clinical diagnosis is complicated by the fact that the symptoms associated with venous thrombosis are relatively common complaints of pregnant women. A rapid genetic test has been developed for simultaneous detection of the most common genetic risk factors associated with thrombophilia, the factor V 1691GA (Leiden) and prothrombin 20210GA mutations. Mutation 677CT in the 5,10-methylenetetrahydrofolate reductase (MTHFR) gene, which increases homocysteine levels by 15-25% when two copies of the T-allele is present, is also included as part of this "prenatal care genetic test". Hyperhomocysteinaemia has been associated with an increased risk of thrombosis and neural tube defects. The importance of multi-gene testing is emphasised by the low predictive value of any single inherited marker and the significant increase in the probability of thrombosis when more than one risk factor is identified.

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## Introduction

In the overall scheme of prenatal care, genetic testing for single gene disorders forms an important component of disease prevention in high-risk populations or families with hereditary defects. This has been shown clearly in a South African family with familial hypercholesterolaemia (FH), where the application of DNA technology prevented the birth of a foetus with homozygous FH.<sup>1</sup> With the availability of a recently-developed multi-gene test for cardiovascular disease (CVD),<sup>2,3</sup> it has become possible to extend genetic testing for CVD risk factors from monogenic to multifactorial conditions. The CVD test consists of four components - dyslipidaemia, folate metabolism, coagulation and iron overload - that are evaluated in the context of family and medical history (e.g. hypertension, obesity, diabetes), biochemical parameters and relevant environmental risk factors. Testing for mutations involved in coagulation and folate metabolism may be particularly useful during pregnancy due to their association with an increased risk for venous thrombosis, recurrent pregnancy loss and foetal abnormalities.

## Inherited thrombophilia in pregnant woman

The risk of venous thromboembolism is at least five times higher in pregnant

women compared with non-pregnant women of similar age and the risk increases further in the presence of genetic risk factors.<sup>4</sup> Factor V Leiden (1691GA) is the most common inherited form of thrombophilia and occurs in approximately 5% of the Caucasian population. The prothrombin mutation (20210GA) is the second most common cause of inherited thrombophilia. The risk is further increased when these gene mutations are present in the same individual, or if one or more mutation occurs in combination with other genetic risk factors such as 677CT in the 5,10-methylenetetrahydrofolate reductase (MTHFR) which causes increased homocysteine levels.

## Inherited thrombophilia and pregnancy loss

Inherited and acquired thrombophilia are responsible for recurrent pregnancy loss of unknown cause in more than 50% of women.<sup>5</sup> Thrombophilic risk factors are also frequent in women with other vascular/placental pathologies such as placental abruption, preeclampsia, intrauterine growth retardation and late foetal loss. The greatest risk was documented in women with homozygous mutations for the factor V Leiden or prothrombin gene or combinations of these risk factors. In addition, genetic variation in the plasminogen activator inhibitor-1 (PAI-1) and coagulation factor

XIII genes appear to contribute synergistically to impaired fibrinolysis which could promote the development of early pregnancy loss.<sup>6</sup>

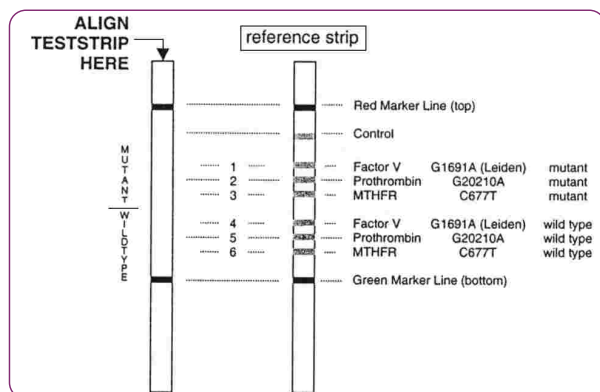
## MTHFR mutation and neural tube defects

The MTHFR 677 CT mutation both in children and their mothers is associated with increased risk of neural tube defects (NTD) and may explain up to half of all folate related neural tube defects.<sup>7</sup> This mutation is dependent on folate status for manifestation of NTD risk. Considering the relatively high frequency of the T-allele in the South African population,<sup>8</sup> maintenance of adequate folate and B-vitamin levels should be stressed for women who are considering pregnancy. The T-allele is also associated with an increased risk of severe diastolic hypertension during pregnancy.<sup>9</sup>

## Genetic testing

The development of a rapid strip-assay test allows simultaneous analysis of the factor V 1691GA (Leiden), prothrombin 20210GA and MTHFR 677CT mutations (Figure 1, ViennaLab, Austria) in a single cost-effective reaction. When deficiencies of proteins C, S and AT have been excluded, extended testing for the plasminogen activator inhibitor-1 (PAI-1) 4G/5G and the coagulation factor XIII Val34Leu polymorphisms<sup>6,10,11</sup> may be particularly useful as part of a

**Figure 1:** The multi-gene genetic test is performed by hybridising the DNA extracted from a cheek swab of the patient with the teststrip and comparing the pattern obtained with the reference strip containing the internal controls for the mutations being analysed.



comprehensive CVD risk reduction approach<sup>12</sup> that may benefit both mother and child.

### Who should be tested for inherited thrombophilia?

Any pregnant woman with a personal history of a previous thromboembolic event or a strong family history of thrombosis should be screened for hypercoagulability. Women with a history of adverse outcome of pregnancy and gestational vascular complications should also be tested. Thrombophilia screening is strongly recommended in the following cases: foetal loss including three or more first trimester, two or more second trimester or any stillbirth; early severe or recurrent pre-eclampsia or severe intrauterine growth restriction.<sup>13</sup>

### Treatment

Prophylactic antithrombotic therapy should not be given to pregnant women without due consideration to the potential harmful effects of such treatment. Due to the high recurrence rate of pregnancy complication in women with thrombophilia, therapy in the form of low molecular weight heparin subcutaneously should be considered for the following indications: history of recurrent pregnancy loss, previous severe pre-eclampsia and possibly for placental abruption, stillbirth and intrauterine fetal growth restriction. The use of prophylactic low dose aspirin or oral anticoagulants such as coumarins is not recommended in pregnancy.

Prophylactic treatment in high risk patients usually starts before pregnancy occurs and needs to be continued until 4-6 weeks after birth, especially in women with both the factor V Leiden and prothrombin mutations and with a

venous thromboembolism before pregnancy. In those without venous thromboembolism before pregnancy, prophylaxis might be decided for each individual case based on the presence of all risk factors identified, which may include multiple genetic risk factors.

Beneficial effects on coagulation status have been shown in pregnant women using folate supplements,<sup>14</sup> therefore folate, vitamins B6, B12 and riboflavin are indicated for patients with hyperhomocysteinaemia. To prevent birth defects such as cleft lip and palate, the recommended dietary allowance (RDA) for lactating and pregnant women is 800-1000 µg folate per day. Women with a previous neural tube defect (NTD) pregnancy may consider taking 4 mg supplemental folic acid per day when planning a subsequent pregnancy.<sup>15</sup> Wald et al.<sup>16</sup> furthermore reported that taking a 5 mg folic acid tablet daily would reduce the risk of NTDs by about 85%.

### Conclusions

Specialised laboratory tests can be requested to distinguish between the many different causes of thrombophilia in patients with early-onset, familial or recurrent venous thromboembolism. The results obtained would determine the intensity and duration of anticoagulant therapy. There is general support for presymptomatic family testing in antithrombin, protein C, or protein S deficiency and where the factor V Leiden or prothrombin mutation is strongly penetrant and expressed. Testing in families in which clotting factor polymorphisms are weakly expressed should be restricted to young women when they consider hormonal contraception or pregnancy, given that these acquired factors multiply the risk.<sup>17</sup>

The fruits of the human genome project will undoubtedly lead to expansion of the menu of possible prenatal testing options for disease prevention and reproductive choice. This can be expected to raise the level of complexity in both counselling and testing logistics for medical practitioners who refer patients for genetic testing. Support is available in the form of a

health professional network ([www.genecare.co.za](http://www.genecare.co.za)) that provides access to genetic counselling services on a national basis.

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