The impact of the Factor V Leiden mutation on pregnancy

Vincenzo Spina¹,*, Vincenzo Aleandri¹ and Francesco Morini²

¹Institute of Obstetrics and Gynaecology, University of Rome ‘La Sapienza’, Viale del Policlinico, 155–00161 Rome, and ²Institute of Pediatrics, University of Rome ‘La Sapienza’, Viale del Policlinico, 155–00161 Rome, Italy

Received on October 8, 1999; accepted on February 14, 2000

A resistance to the anticoagulant activity of activated protein C (APC), most frequently due to a point mutation in the Factor V gene (the Leiden mutation), represents the most common genetic cause of thrombophilia. The Leiden mutation has been significantly related to pregnancy complications associated with hypercoagulation, e.g. deep vein thrombosis during pregnancy (8-fold increased risk), pre-eclampsia (prevalence of the mutation up to 26%), placental infarction extending to >10% of the placenta (10-fold increased risk), abruptio placentae (prevalence of the mutation up to 29.6%), and second- and third-trimester pregnancy failure (prevalence of the mutation up to 31.3%). An association of the maternal mutation with recurrent first-trimester miscarriage does not emerge from the literature, although fetal mutation (frequency higher than twice compared with that of the general population) has been related to early spontaneous miscarriage. Although some evidence suggests an association between APC resistance and intrauterine growth retardation, no significant relationship emerges currently from the literature. Screening for the Leiden mutation would seem advisable in women with previous pregnancy complications amongst those associated with APC resistance. Carriers of the mutation should be given appropriate counselling. The screening of asymptomatic women is not recommended at present.

Key words: activated protein C resistance/Factor V Leiden mutation/obstetric pathologies/pregnancy outcome/thrombophilia

TABLE OF CONTENTS

Introduction 301
Pathogenesis of thrombophilia by Factor V Leiden mutation 302
Impact of Factor V Leiden on pregnancy 302
Conclusions 305
References 305

Introduction

A condition of hypercoagulation has been described as a major factor in the pathogenesis of some severe obstetric pathologies, e.g. deep venous thrombosis, pre-eclampsia and intrauterine growth retardation (IUGR), as well as pregnancy failure. On the other hand, pregnancy itself represents a risk factor for venous thrombosis (Koster et al., 1993). Previously, known inherited thrombophilias included deficiencies of protein C, protein S and antithrombin; their relationship to the above mentioned pathologies and spontaneous miscarriage has been studied. However, taken together, such defects are responsible for only 15% of familial thrombophilias, and <5% of all venous thromboses (Cooper, 1994).

A hyperhomocysteinemia resulting from a mutation (677 C→T) in the methylene tetrahydrofolate reductase (MTHFR) gene, and a mutation (20210 G→C) in the Factor II (prothrombin) gene have been identified as further risk factors for thromboembolism (Guttormsen et al., 1996; Poort et al., 1996). Although scant data are available about their role in pathogenesis of obstetric pathologies (Grandone et al., 1998; Kutteh et al., 1999; Souza et al., 1999), knowledge of them may have therapeutic implications, e.g. in the case of a hyperhomocysteinemia, the use of folic acid and vitamin B6 has been suggested to enhance the chances of a physiological pregnancy (Quere et al., 1998).

A resistance to the anticoagulant activity of activated protein C (APC) has been recently described as a major cause of predisposition to thromboembolism (Dahlbäck et al., 1993), accounting for up to 40% of all thromboses (Dahlbäck, 1994). More than 95% of cases of APC resistance are due to a point mutation in the Factor V gene, i.e. guanine to adenine substitution at nucleotide 1691 (1691 G→A), known as the Leiden mutation (Zöller and Dahlbäck, 1994; Bertina et al., 1994), which is inherited as an autosomal dominant trait (Svensson and Dahlbäck, 1994). The risk of thrombosis is increased seven times in heterozygotes and 80 times in homozygotes. The distribution of Factor V Leiden varies greatly in ethnic groups (Rees et al., 1995). The estimated carrier rate is ~5% in Caucasians (European, Jews, Israeli Arabs, Indians) (De Stefano et al., 1998), reaching ~10–

*To whom correspondence should be addressed at: Iargo Messico 7, 00198 Roma, Italy. Phone: +39 06 855 21 96; Fax: +39 06 70 45 21 28.
15% in some European countries (Rees et al., 1995), while the allele is virtually absent in Africans and Asians (De Stefano et al., 1998). A carrier frequency of 20–60% has been reported in individuals with a personal or familial history of thrombosis (Bertina et al., 1994). Thus, it is clear that the Factor V Leiden mutation represents the most common hereditary cause of venous thrombosis, being at least 10 times more frequent than any of the anticoagulant protein deficiencies (Dahlbäck, 1994). The aim of this review is to update current knowledge on the association of Factor V Leiden with obstetric pathologies related to haemostatic disorders and fetal loss.

Pathogenesis of thrombophilia by Factor V Leiden mutation

Blood coagulation is under the control of anticoagulant proteins present in plasma or on the surface of endothelial cells (Davie et al., 1991). Protein C, a vitamin K-dependent plasma protein, plays a key role in natural anticoagulation. Once activated on the endothelial cells by the thrombin–thrombomodulin complex, protein C exerts a selective, proteolytic degradation of Factors Va and VIIIa (Dahlbäck and Stenflo, 1994). Protein S, another vitamin K-dependent protein present in plasma as both free protein (the active form) and protein bound to complement C4b-binding protein (C4BP), acts as a cofactor to APC (Dahlbäck, 1991; Dahlbäck and Stenflo, 1994) (Figure 1).

The replacement of arginine with a glutamine at position 506 in the Factor Va protein molecule, encoded by the Leiden mutation, alters the cleavage site of this molecule by activated protein C. As a result, the mutated Factor Va, while maintaining its physiological procoagulant properties, becomes resistant to proteolytic inactivation by APC (Bertina et al., 1994). In addition, co-factor activity to APC, recently found to depend upon Factor V, becomes lost (Dahlbäck and Hildebrand, 1994). These changes, in turn, yield stabilization of the prothrombinase complex, rise in thrombin production, and feedback activation of Factors V and VIII (Figure 1), resulting in a predisposition to thromboembolism (Dahlbäck, 1994).

Impact of Factor V Leiden on pregnancy

It is well known that pregnancy involves important changes in the haemostatic system in a direction that promotes coagulation. Values of fibrinogen, prothrombin, Factors V, VII, VIII, IX, X and XII are increased, whereas a decrease in the antithrombin concentration takes place (Corash, 1986). Although physiological, such changes may predispose to thromboembolism. When pregnancy is associated with a thrombophilia of any origin, then the thromboembolic risk becomes highly increased. Thus, the course of gestation may also be complicated by either pathologies currently known to be related to haemostatic disorders or even fetal loss. The Factor V Leiden mutation shows a high prevalence in the thrombosis as well as in the general white population (Bertina et al., 1994; Rees et al., 1995), thereby attracting the interest of researchers in investigating its role in such pregnancy complications.

Factor V Leiden and deep venous thrombosis in pregnancy

Thromboembolism is a major cause of maternal mortality, accounting for 12–15% of maternal deaths in pregnancy (Högberg, 1986; Sachs et al., 1987). The incidence of thromboembolic events has been reported to range from 0.018 to 0.1% during gestation, versus 0.005% in non-pregnant women of reproductive age (Böttiger and Westerholm, 1971; Sipes and Weiner, 1990); this means that the thromboembolic risk increases ~3–20 times during pregnancy. During the puerperium, the risk of thrombosis is increased by a further 2–3 times (Sipes and Weiner, 1990), and, following a Caesarean section, it becomes 3–5 times higher than after vaginal delivery (Berqvist et al., 1979).

An interesting investigation found that the prevalence of APC resistance and the Leiden mutation in patients with thrombosis during pregnancy was 46% (Bokarewa et al., 1996); whereas another study (Hellgren et al., 1995) found that it reached ~60%, compared with ~10% amongst non-pregnant and pregnant healthy controls.

Another study reported that the carrier frequency of the Factor V Leiden mutation in a low-risk obstetric population was ~3% (14 of 407 women), consistent with the published carrier rate in the general population. The incidence of deep venous thrombosis in the carriers was found to be 28%, whereas it was <1% in the population not carrying the mutation (Dizon-Townson et al., 1997a).
A large prospective study found that the risk of venous thromboembolism was 8-fold higher in pregnant women with the Factor V:G506 mutation than in the pregnant ones without APC resistance (three out of 270 versus three out of 2210 respectively) (Lindhqvist et al., 1999).

It has been suggested that women with a thromboembolic event should be screened for this mutation (Hellgren et al., 1995; Dizon-Townson et al., 1997a). In addition, given the high prevalence of the Leiden mutation, the question has been raised with regard to the opportunity of including testing for this mutation in routine prenatal laboratory assessment (Dizon-Townson et al., 1997a).

**Factor V Leiden and pre-eclampsia**

Although the aetiology of pre-eclampsia still remains to be established, some of the pathogenetic mechanisms have been elucidated. Clinical evidence indicates that pre-eclampsia may be aetologically heterogeneous; a genetic predisposition is suggested by the higher incidence of the pathology in patients whose mother or sister were affected (O’Brien, 1990), while the association with disorders of either coagulation or the immune system has been described (Lockshin, 1990; Triplett, 1992).

The relationship with haemostatic disorders appears to be of particular importance. It has been demonstrated that pre-eclampsia is associated with endothelial injury, clotting and fibrinolysis (Saleh et al., 1987). There is convincing body of evidence suggesting that, mostly, endothelial injury is crucial in development of pre-eclampsia (Roberts and Redman, 1993). In fact, amongst markers of endothelial injury, clotting and fibrinolysis, fibronectin showed the most significant changes in relation to pre-eclampsia. In addition, the clinical resolution of pre-eclampsia after delivery was more closely related to fibronectin than other parameters of haemostasis (Saleh et al., 1987). The endothelial damage may trigger both clotting and fibrinolysis (Busch and Gerdin, 1981; de Groot and Taylor, 1993). Although pregnancy is physiologically characterized by increase in clotting and fibrinolysis, such changes have been reported to be exaggerated in pre-eclampsia (McKay, 1982; Redman, 1995).

It has been postulated that endothelial injury may be caused by placental ischaemia (Williams and de Swiet, 1997). Poor utero-placental perfusion is a common feature of pregnancies complicated by pre-eclampsia (Roberts and Redman, 1993). Altered utero-placental vasculature, resulting from impaired remodelling by invasive trophoblast, has been suggested to cause decreased utero-placental perfusion. In pregnancies with pre-eclampsia, cytotrophoblast invasion of the spiral arteries is incomplete and high resistance vessels with a muscular wall (instead of thin-walled, low resistance vessels) persist until term (Khong et al., 1986; Zhou et al., 1993).

The low-pressure intervillus perfusion, in the presence of a hypercoagulable state, may determine fibrin deposition in the placenta (Dizon-Townson et al., 1996). Indeed, placental lesions related to coagulation are common findings in pre-eclampsia; they include utero-placental vessel thrombosis, intervillous thrombi, perivillous fibrin deposition, avascular villi, fetal stem vessel thrombi, etc. Such lesions have also been observed independently of utero-placental vascular pathologic features; i.e. coagulation may be involved in the pathogenesis of pre-eclampsia, independent of utero-placental vascular lesions (Salafia et al., 1995).

Systemic fibrin deposition and platelet thrombi in tissue samples have been demonstrated in pre-eclampsia patients, indicating a stimulation of the haemostatic system (McKay, 1972).

A thrombophilia may therefore play a significant role in the pathophysiology of pre-eclampsia. Deficiencies of protein S, protein C and antithrombin III, and antiphospholipid antibodies syndrome had been previously demonstrated in association with pre-eclampsia (Aznar et al., 1986; Dekker et al., 1995).

More recently, APC resistance has been observed in 16–22% of white population with previous pre-eclampsia (Dekker et al., 1995; Lindoff et al., 1997). Another study demonstrated a statistically significant difference in the carrier rate of the Factor V Leiden between patients with severe pre-eclampsia and normotensive pregnant controls (8.9 versus 4.2% respectively) (Dizon-Townson et al., 1996). These results have been confirmed by Mimuro et al. (1998), who reported a significantly higher prevalence of both APC resistance and Factor V Leiden mutation in patients with pre-eclampsia compared with normal pregnant controls (22 versus 2.7%, $P < 0.01$; and respectively, 8 versus 0.07%, $P < 0.01$). In addition, the study by Kupferminc et al. (1999) on 110 patients with severe obstetric complications and 110 normal pregnant controls demonstrated the Leiden mutation in 26% (nine out of 34) of the women with severe pre-eclampsia (OR 5.3; 95% CI: 1.8–15.6).

These results suggest a correlation between the Leiden mutation and pre-eclampsia, and raise once again the question of whether the test for Factor V Leiden should be included in genetic screening for various obstetric pathologies, e.g. pre-eclampsia.

**Factor V Leiden and fetal loss**

The outcome of pregnancy is highly affected by the placental development and function, which, in turn, depend upon the creation of an adequate maternal–fetal circulation. A thrombophilia may result in impaired maternal–fetal circulation by compromising the vascular system. Maternal floor infarction, which is actually characterized by a massive deposition of fibrin in the decidua underneath the placenta, has been associated with significant fetal morbidity and mortality (Naeye, 1985; Laurini et al., 1994).

Placental thrombi have been associated with maternal haemostatic disorders including deficiencies of proteins C and S, and Lupus anticoagulant (Rayne and Kraus, 1993). Interestingly, in animal experiments provide evidence that the protein C/protein S pathway is important for successful pregnancy outcome (Healy et al., 1995). Furthermore, a European multicentre study, European Prospective Cohort on Thrombophilia (EPCOT), found a significant risk in the risk of fetal loss (especially stillbirth), which was correlated with inherited deficiencies of antithrombin, protein C and protein S (Preston et al., 1996). Therefore, available data lend support to the hypothesis that the increased risk of fetal loss caused by such defects may result from utero-placental insufficiency.

A positive correlation between the Leiden mutation and recurrent first-trimester pregnancy loss did not emerge from the current literature. An investigation reported that none of 40 women with idiopathic recurrent miscarriage (22 of which were first-trimester aborters) carried the mutation (Dizon-Townson et al., 1997b). Similar results have been documented by a
Japanese study on 52 women with recurrent first trimester abortion, 41 of their partners and 55 controls with a previous uneventful pregnancy: the mutation was found in none of the abortors, partners and controls, although it must be taken into account that the prevalence of the mutation is extremely low in the Japanese population (Hashimoto et al., 1999). Another investigation on 50 women with three or more pregnancy losses (91.7% of which occurred during the first trimester) documented a heterozygosity for the Leiden mutation in 2% of patients versus 4% of controls (OR 0.49; 95% CI 0.04–5.58) (Kutteh et al., 1999). The study by Tal et al. (1999) on 125 patients and 125 controls found a significantly decreased rate of clinical first-trimester miscarriages amongst patients carrying the Leiden mutation compared with patients with no APC resistance (21/48 versus 177/214 abortions respectively; P < 0.001); interestingly, a significant increase in the frequency of preclinical miscarriages has been shown in the former patients, compared with the latter, by this study (17/48 versus 25/214 respectively; P < 0.001).

Conversely, most of the recent investigations support a positive correlation between the Leiden mutation and the second and third-trimester pregnancy loss. An increased risk for second-trimester pregnancy loss has been found in patients with an APC resistance (Brenner et al., 1997). Rai et al. (1995) reported a frequency of Leiden mutation carriers significantly higher amongst second-trimester abortors than amongst controls (20 versus 4.3% respectively). Another investigation (43 caucasian women and 118 controls) found a 31.3% frequency of the mutation amongst patients with second and third-trimester abortions, compared with 7.4% amongst first-trimester aborters and 4.2% in the control group (Grando et al., 1997). Tal et al. (1999) reported a rate of second-trimester pregnancy losses significantly higher in patients with the Leiden mutation than in patients with no APC resistance (10/48 versus 14/214; P < 0.01). The EPCOT study on 1384 women (843 patients and 541 controls) appears in agreement with the above data; an OR of 2.0 (95% CI 0.5–7.7) has been reported for stillbirth (fetal loss after 28 weeks gestation), suggesting an association between the Leiden mutation and late pregnancy loss (Preston et al., 1996).

Importantly, the frequency of fetal carriers of the Factor V Leiden amongst miscarried fetuses (mean gestational age: 12 weeks) has been documented to be as high as twice that of general population (Dizon-Townson et al., 1997c). Thus, fetal expression of the Leiden mutation, and therefore hypercoagulable state, presenting early in gestation seems to predispose the fetus to spontaneous miscarriage. Nevertheless, large prospective studies are needed to evaluate the opportunity of testing couples with recurrent miscarriage for this mutation (Dizon-Townson et al., 1997c).

Factor V Leiden and placental lesions

Placental infarctions are frequent features in pathologies potentially related to Factor V Leiden, e.g. pre-eclampsia and fetal loss. An interesting study investigating the relationship between adverse pregnancy outcome, placental thrombotic lesions and laboratory abnormalities consistent with a thrombophilia, found a 53.8% frequency of the Leiden mutation amongst 13 patients (Arias et al., 1998).

In addition, true placental infarctions due to thrombotic events have been reported to be significantly associated with the fetal genotype of the Factor V Leiden mutation. Importantly, a 10-fold increase in the carrier frequency of the mutation has been demonstrated in fetuses with placentas presenting infarction of >10% in extent. Interestingly, such infarctions were characterized by localization in the distribution of the fetal vessels coursing along the surface of the fetal side of the placenta (Dizon-Townson et al., 1997c). These data suggest that fetal expression of the Leiden mutation presenting late during pregnancy predisposes to placental infarction in the distribution of the fetal vessels (Dizon-Townson et al., 1997c).

Abruptio placentae is another placental lesion, which has been associated to APC resistance and the Factor V Leiden mutation. Wiener-Megnay et al. (1998) found an APC resistance in 17 of 27 patients compared with five of 29 control subjects (OR 8.3; 95% CI 3.6–21.75; P = 0.00125), and the Leiden mutation in 29.6% of the patients (eight of 27). Such a correlation has been more recently confirmed by a study, which detected the Factor V Leiden mutation in five of 20 patients (25%) with abruptio placentae (Kupferminc et al., 1999).

Factor V Leiden and IUGR

There is increasing agreement that altered utero–placental circulation may have an important role in the pathogenesis of IUGR (Brosens et al., 1987). Histological features of utero–placental insufficiency have been documented with a frequency significantly higher in pregnancies with IUGR than in normal ones (Rayburn et al., 1989).

Massive intervillous fibrin deposition (MIFD) is characterized histologically by a plaque composed of villi entrapped in fibrin, which obliterates the whole intervillous space (Fox, 1978). Such a lesion is secondary to intravascular coagulation and thrombosis resulting from disturbances of utero–placental perfusion and abnormal platelet behaviour. In addition to recurrent pregnancy failure (Perrin, 1984), MIFD has also been related to IUGR (Fuke et al., 1994). In fact, since the intervillous space is entirely obliterated by fibrin, placental ischaemia and inhibition of nutritional absorption may follow, causing IUGR eventually. The small-for-date birth rate in pregnancies with MIFD placentas has been reported to be ~8 times greater than in controls (62.9 versus 8.3%). Furthermore, the recurrence rate of MIFD appeared to be high. Interestingly, MIFD did not recur after therapy with aspirin, dipyridamole and/or heparin, and the incidence of approximate-for-date in the treated group was significantly higher than in the untreated one (87.5 versus 33.3%) (Fuke et al., 1994).

A significant correlation has also been documented between placental infarction and the incidence of small-for-gestational-age fetuses as well as Doppler findings of an abnormal blood flow in the descending aorta and the umbilical artery (Laurini et al., 1994). Finally, fetal thrombotic vasculopathy resulting in avascular villi within the placenta has been reported to be associated with increased rates of IUGR (Redline and Pappin, 1995). All of the above data indicate a relationship between ischaemic pathology of the placenta and restriction of fetal growth. In addition, maternal haemostasis disorders have been related to placental ischaemic lesions and IUGR (Redline and Pappin, 1995).

Since Factor V Leiden has also been documented as predisposing to placental ischaemic lesions and infarction (Dizon-Townson et al., 1997c), then it may be reasonably speculated that...
this mutation might result in utero-placental insufficiency and restriction of fetal growth. However, no significant correlation between the mutation and IUGR has been demonstrated so far. In fact, although IUGR has been observed in association with APC resistance and pre-eclampsia, its prevalence did not show a statistical difference between those patients with, and those patients without, APC resistance (Lindoff et al., 1997; Lindqvist et al., 1998; Kupferminc et al., 1999).

Conclusions

Evidence from research suggests that APC resistance due to the Factor V Leiden may be involved (probably through different mechanisms and interactions with other aetiological factors) in pathogenesis of serious obstetric complications, e.g., gestational deep venous thrombosis, pre-eclampsia, placental infarction, abruptio placenta and recurrent late pregnancy failure. A significant association with IUGR has not been demonstrated.

We agree with Rouse et al. (1997) and Walker (1997) that screening for the Factor V Leiden mutation of asymptomatic pregnant women is not recommended at present. A prospective study in Sweden on 2480 women in early pregnancy has not found in the women carrying the Leiden mutation a pregnancy outcome significantly different from the ones with no APC resistance; however, an 8-fold higher risk of venous thromboembolism has been found in the former, and conclusions are not drawn regarding the association of the mutation with perinatal death (Lindqvist et al., 1999). The high prevalence of Factor V Leiden suggests that further large prospective studies are needed to assess definitively the opportunity of both utilizing the APC resistance test (or the test for the Leiden mutation) as a screening analysis for multiple obstetric pathologies and addressing the indication, in carriers of the mutation, to adequate thromboprophylaxis.

Currently, screening for thrombophilia (including the test for the Leiden mutation) would seem to be advisable in women with previous pregnancy complications, amongst those associated with the mutation, for their tendency to recur. Ideally, testing should be performed between pregnancies, since the concentrations and activity of anticoagulant proteins change physiologically (decrease in protein S and antithrombin values, increase in APC resistance) during pregnancy (Nelson-Piercy, 1999). Although there exists no definitive evidence indicating the possibility of improving the outcome in future pregnancies, preliminary studies suggest a favourable effect of low molecular weight heparin on the pregnancy outcome of women with a history of pregnancy complications (pre-eclampsia, pregnancy loss, fetal growth restriction) and documented APC resistance (Riyazi et al., 1998; Tal et al., 1999); however, such results must be urgently confirmed by prospective randomized trials. Meanwhile, the patients should be informed on the potential risks (osteoarthritis, in the case of heparin) of untested but logical therapeutic treatments (Nelson-Piercy, 1999).

In any case, counselling should be provided to carriers of the Leiden mutation, with regard to their own and their family members thromboembolic risk. In particular, women of reproductive age should be given correct information concerning the risk of both thromboembolic complications related to pregnancy, oral contraception and surgery, and obstetric complications related to such a thrombophilia.

References


Received on October 8, 1999; accepted on February 14, 2000.