Factor V Leiden thrombophilia

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Abstract: Factor V Leiden is a genetic disorder characterized by a poor anticoagulant response to activated Protein C and an increased risk for venous thromboembolism. Deep venous thrombosis and pulmonary embolism are the most common manifestations, but thrombosis in unusual locations also occurs. The current evidence suggests that the mutation has at most a modest effect on recurrence risk after initial treatment of a first venous thromboembolism. Factor V Leiden is also associated with a 2- to 3-fold increased relative risk for pregnancy loss and possibly other obstetric complications, although the probability of a successful pregnancy outcome is high. The clinical expression of Factor V Leiden is influenced by the number of Factor V Leiden alleles, coexisting genetic and acquired thrombophilic disorders, and circumstantial risk factors. Diagnosis requires the activated Protein C resistance assay (a coagulation screening test) or DNA analysis of the *F5* gene, which encodes the Factor V protein. The first acute thrombosis is treated according to standard guidelines. Decisions regarding the optimal duration of anticoagulation are based on an individualized assessment of the risks for venous thromboembolism recurrence and anticoagulant-related bleeding. In the absence of a history of thrombosis, long-term anticoagulation is not routinely recommended for asymptomatic Factor V Leiden heterozygotes, although prophylactic anticoagulation may be considered in high-risk clinical settings. In the absence of evidence that early diagnosis reduces morbidity or mortality, decisions regarding testing at-risk family members should be made on an individual basis. ***Genet Med* 2011:13(1):1–16.**

Key Words: *Factor V Leiden, thrombophilia, APC resistance, venous thrombosis, anticoagulation*

Venous thromboembolism (VTE; deep venous thrombosis [DVT] and/or pulmonary embolism) is a major medical problem, which affects approximately 1 in 1000 persons per year. VTE is a multicausal disease that results from multiple interactions between genetic, acquired, and circumstantial risk factors. Thrombophilic disorders can be identified in half of patients presenting with venous thrombosis. Inherited thrombophilia is a genetically determined tendency to develop VTE, which is often recurrent. Inherited deficiencies of antithrombin, Protein C, and Protein S are relatively rare and together account for $<10\%$ of patients with venous thrombosis. In 1993, Dahlbäck et al.[1] reported an inherited poor response to the anticoagulant effect of activated Protein C (APC) in several thrombosis prone families. The APC-resistant phenotype was prevalent among patients with venous thrombosis and subsequently shown to result from a single-point mutation in the Factor V gene.[2]

Factor V Leiden is the most common genetic risk factor for VTE, found in 20–25% of patients with VTE and 50% of patients with familial thrombophilia.[3,4] Testing for Factor V Leiden is now one of the most frequently ordered molecular genetic tests. This review focuses on current data defining the thrombotic risks and complications associated with Factor V Leiden and its interaction with other genetic and acquired risk

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Disclosure: The author declares no conflict of interest.

Submitted for publication August 12, 2010.

Accepted for publication September 1, 2010.

Published online ahead of print November 24, 2010.

DOI: 10.1097/GIM.0b013e3181faa0f2

factors. It also summarizes indications for testing and guidelines for management and genetic counseling.

PATHOGENIC MECHANISMS AND MOLECULAR BASIS

Factor V Leiden is a genetic disorder characterized by a poor anticoagulant response to APC. APC is a natural anticoagulant protein that cleaves and inactivates procoagulant Factors Va and VIIIa, thereby downregulating further thrombin generation.[5,6] APC inactivates Factor Va by cleavage at three different amino acid positions: R (arginine) 306, R 506, and R 679. The term "Factor V Leiden" refers to the specific guanine to adenine substitution at nucleotide 1691 in the Factor V gene, which predicts the substitution of glutamine for arginine at the Arg 506 APC cleavage site. Because of this single amino acid substitution, Factor Va is resistant to APC and is inactivated at a 10-fold slower rate than normal, resulting in increased thrombin generation. Factor V cleaved at position 506 also functions as a cofactor (along with Protein S) for APC-mediated inactivation of Factor VIIIa. Loss of this anticoagulant activity of Factor V may contribute to thrombin generation. The resulting mild prothrombotic state is reflected by elevated levels of D-dimer, prothrombin fragment F1 + 2, and other activated coagulation markers.[7–9] The mutant Factor V is referred to as Factor V Leiden because Dutch investigators from the city of Leiden were the first to report the mutation.[2]

PREVALENCE

Factor V Leiden is the most common inherited form of inherited thrombophilia, accounting for 40–50% of cases. The prevalence varies by population (Table 1). Heterozygosity for Factor V Leiden occurs in 3–8% of the general US and European populations. The highest heterozygosity rate is found in Europe; the mutation is extremely rare in Asian, African, and indigenous Australian populations. Within Europe, the prevalence varies from 10 to 15% in southern Sweden and Greece to 2–3% in Italy and Spain.[10] The mutation is found in 3.8% of individuals in France, but the frequency ranges from 1.3% in southwestern regions to 7.1% in northeastern France.[9] In the United States, the prevalence reflects the world distribution of the mutation.[11] It is present in:

- 5.2% of white Americans.
- 2.2% of Hispanic Americans.
- 1.2% of African Americans.
- 0.45% of Asian Americans.
- 1.25% of native Americans.

The frequency of homozygosity for Factor V Leiden in white populations is approximately 1 in 5000. Haplotype analysis of the Factor V gene strongly suggests that the mutation was a single event that occurred 20,000–30,000 years ago, after the evolutionary separation of whites from Asians and Africans.[18] The high prevalence of Factor V Leiden among whites suggests a balanced polymorphism with some type of survival advantage associated with the heterozygous state. Some investigators speculate that the mild prothrombotic state conferred by the mutation could have reduced mortality from bleeding associated with childbirth or trauma in premodern times.[5,18] In several studies, Factor V Leiden heterozygotes had significantly less blood loss during menses, childbirth, and cardiac surgery.[19–21] Some evidence suggests that severe hemophiliacs who are heterozygous for Factor V Leiden have less severe bleeding and reduced clotting factor concentrate utilization, also consistent with this hypothesis.[22,23] However, an evolutionary survival benefit of Factor V Leiden has not been confirmed.

Table 1 Worldwide prevalence of Factor V Leiden[a,b]

Population	Prevalence (%)[9–11,12–17]
European whites	3–15
Spain	3.3
France	3.8
Germany	4
Iceland	5.2
United Kingdom	8.8
Greece	15
Sweden	11
Africa	Absent
Southeast Asia	Absent
Asia minor	1.2
Australia (indigenous)	Absent
Japan	Absent
Jordanian Arabs	12.2
Lebanon	14
Western Iran	2.97
Canada	5.3
United States	
Whites	5.2
Hispanic Americans	2.2
African Americans	1.2
Asian Americans	0.45
Native Americans	1.25

[a]Healthy individuals with no history of venous thromboembolism.
[b]Includes heterozygous and homozygous individuals.

DIAGNOSIS

Clinical diagnosis

No clinical features are specific for Factor V Leiden thrombophilia. The diagnosis is suspected in individuals with a history of VTE manifest as DVT or pulmonary emboli, especially in women with VTE during pregnancy or in association with estrogen contraception, and in individuals with a personal or family history of recurrent thrombosis.

Testing

The diagnosis of Factor V Leiden requires the APC resistance assay, a coagulation screening test, or DNA analysis of *F5*, the gene encoding Factor V, to identify the Leiden mutation, a specific G-to-A substitution at nucleotide 1691 that predicts a single-amino acid replacement (R506Q).

The APC resistance assay involves performing an activated partial thromboplastin time (aPTT) on the individual's plasma in the presence and absence of a standardized amount of exogenous APC; the two results are expressed as a ratio (aPTT +

APC/aPTT – APC). This assay is based on the principle that when added to normal plasma, APC inactivates Factors Va and VIIIa, which slows coagulation and prolongs the aPTT. The APC-resistant phenotype is characterized by a minimal prolongation of the aPTT in response to APC and a correspondingly low ratio. Modified ("second generation") assays include predilution in Factor V-deficient plasma to normalize factors that might affect the APC resistance ratio (Protein S, other Vitamin-K-dependent clotting factors lowered by oral anticoagulation, etc.). The modified assay can be used to test plasma from patients receiving anticoagulation. This test has a sensitivity and specificity for Factor V Leiden approaching 100%.[24]

Targeted mutation analysis for Factor V Leiden is performed by a variety of comparable methods using genomic DNA in peripheral blood mononuclear cells.[25,26] Molecular genetic tests are reliable in individuals on warfarin or heparin anticoagulation and during acute thrombotic episodes. When clinical care requires testing for Factor V Leiden, either direct DNA-based genotyping or a modified APC resistance assay is recommended. DNA-based testing is recommended in individuals with strong lupus inhibitors and a prolonged baseline aPTT and those with very low or borderline APC resistance assay values. Individuals with a positive screening assay should have the DNA test for confirmation and to distinguish heterozygotes, homozygotes, and "pseudohomozygotes" who are heterozygous for both Factor V Leiden and a second mutation causing Factor V deficiency. When relatives of individuals known to have Factor V Leiden are tested, the DNA method is recommended.[25]

Indications for testing

There is a general consensus that Factor V Leiden testing may have utility in certain circumstances.[25–31] However, guidelines differ on the specific indications for testing. The decision to test selected patients should be based on the likelihood that the results are likely to influence treatment.[31] There is no evidence that testing for thrombophilia reduces the risk for recurrent VTE or improves clinical outcomes in family members who undergo screening.[32,33] Recent consensus recommendations also differ on the indications for screening women with adverse pregnancy outcomes.[29–31] Testing is appropriate in selected individuals with the following:

- A first unprovoked VTE at any age (especially age $<$ 50 years).
- A history of recurrent VTE.
- Venous thrombosis at unusual sites (e.g., cerebral, mesenteric, hepatic, and portal veins).
- VTE during pregnancy or the puerperium.
- VTE associated with the use of estrogen contraception or hormone replacement therapy (HRT).
- A first VTE with a first-degree family member with VTE before the age of 50 years.

Other clinical circumstances in which testing may be appropriate include the following:

- Women with recurrent unexplained first-trimester pregnancy losses, or an unexplained fetal loss after 10 weeks gestation, or stillbirth.
- Selected women with unexplained severe preeclampsia, placental abruption, or a fetus with severe intrauterine growth restriction.
- A first VTE related to the use of tamoxifen or other selective estrogen receptor modulators (SERMs).
- Individuals younger than 50 years with a first provoked VTE in the absence of malignancy or an intravascular device.
- Asymptomatic adult family members of probands with a known Factor V Leiden mutation, especially those with a strong family history of VTE at a young age or female family members who are pregnant or are considering estrogen contraception use or pregnancy.
- Female smokers younger than 50 years with a myocardial infarction or stroke.
- Neonates and children with noncatheter-related idiopathic VTE or stroke.

Factor V Leiden testing is not routinely recommended during pregnancy or before the use of estrogen contraception, hormone replacement, or SERMs. Prenatal testing and screening of asymptomatic newborns, neonates, and children are also not recommended. Routine testing is not appropriate for patients with a personal or family history of arterial thrombosis (acute coronary syndrome or stroke). However, it may be considered in individuals younger than 50 years with unexplained arterial thrombosis.

Specific informed consent is not generally required for Factor V Leiden genetic testing, although it may be required in some states. However, before testing, individuals should be informed that genetic test results have implications regarding risk for other family members and the attendant issues of confidentiality. The results should be interpreted by a physician experienced in the management of thrombophilia and thrombosis.

NATURAL HISTORY AND CLINICAL MANIFESTATIONS

The clinical expression of Factor V Leiden thrombophilia is variable. Many individuals with a Factor V Leiden allele never develop thrombosis. Although most affected individuals do not experience their first thrombotic event until adulthood, some have recurrent VTE before the age of 30 years. Factor V Leiden homozygotes are more likely to develop their first VTE at a younger age. In a family study, 40% of homozygotes had a VTE by the age of 33 years, compared with 20% of heterozygotes and 8% of unaffected individuals.[34] Population studies suggest approximately 10% of Factor V Leiden heterozygotes develop VTE over their lifetime.[35] The lifetime risk of VTE is higher (25–40%) in heterozygotes from thrombophilic families.[36,37] Heterozygosity for Factor V Leiden is not associated with an increase in mortality or reduction in normal life expectancy.[38,39]

Venous thromboembolism

The primary clinical manifestation of Factor V Leiden is VTE. The mutation is found in 25% of patients with a first idiopathic VTE and up to 40–50% of those with recurrent VTE or an estrogen-related thrombosis[40,41] (Table 2). DVT most commonly develops in the legs, but upper extremity thrombosis also occurs. Some evidence suggests that pulmonary embolism is less common than DVT in individuals with a Factor V Leiden allele.[56,57] An analysis of pooled data from a large number of studies found that the prevalence of a Factor V Leiden allele in individuals with isolated pulmonary embolism was approximately 50% the prevalence in individuals with DVT.[58] The explanation for a differential effect of Factor V Leiden on the risk for DVT and pulmonary embolism is unclear.

Factor V Leiden was identified in 9–20% of individuals with upper extremity DVT including DVT not related to a central venous catheter.[43–45] Thrombosis in unusual locations including

Table 2 Estimated prevalence of Factor V Leiden among patients with thrombotic complications

Thrombotic complication	Prevalence (%)
First idiopathic VTE[3]	25
Recurrent VTE[40,42]	30–50
Upper extremity thrombosis[43–45]	9–20
Cerebral vein thrombosis[46,47]	10–20
Pregnancy-associated VTE[48–51]	20–46
Oral contraceptive-associated VTE[49,52]	20–60
Pregnancy loss[11,53–55]	8–30

VTE, venous thromboembolism.

cerebral, retinal, hepatic, ovarian, and renal veins may also occur less commonly. The risk of cerebral vein thrombosis is increased 3- to 5-fold in Factor V Leiden heterozygotes.[59,46] A Factor V Leiden allele is associated with a 6-fold increased risk for superficial vein thrombosis.[60]

Risk for a first VTE

The relative risk for VTE is increased 3- to 8-fold in Factor V Leiden heterozygotes (Table 3). Lower relative risks are reported in heterozygotes identified from general population screening.[35,72] The thrombotic risk is increased 10- to 80-fold in homozygotes[61,62] (Table 4). The risk for primary upper extremity thrombosis (not related to malignancy or a venous catheter) is increased 3- to 6-fold in Factor V Leiden heterozygotes.[43,45]

Recurrent VTE

Approximately 30% of individuals with an incident VTE develop recurrent thrombosis within the subsequent 8 years.[78] There are conflicting data on the risk of recurrent VTE associated with Factor V Leiden heterozygosity. The current evidence suggests that it has at most a modest effect on recurrence risk after initial treatment of a first VTE. Several prospective cohort studies of unselected individuals with a first VTE, did not find an increased risk for recurrence in Factor V Leiden heterozygotes.[79,80] Recently reported follow-up of participants in the Leiden Thrombophilia study also found no increase in recurrence risk.[81] A meta-analysis including 3104 individuals with a first VTE concluded that a heterozygous mutation is associated with a significantly increased risk for recurrent VTE after a first event with an odds ratio of 1.4.[42] Two recent systematic reviews found a similar modest but significant increase in recurrence risk (pooled odds ratio = 1.56 and 1.45, respectively).[33,68] Several studies showed that the reduction in risk during oral anticoagulation is similar in individuals with and without the factor Leiden mutation.[33,82]

The risk for recurrent VTE in Factor V Leiden homozygotes is not well defined but presumed to be higher than in heterozygotes. Prospective follow-up of the Leiden Thrombophilia study reported a 5-year cumulative recurrence rate of 12.5% in a small group of homozygotes not receiving long-term anticoagulation.[79] A systematic review found a 2- to 3-fold increased risk for recurrent VTE in Factor V Leiden homozygotes.[33] Individuals doubly heterozygous for both Factor V Leiden and the prothrombin 20210G>A mutation have a 3- to 9-fold higher risk for recurrence than those with neither mutation and a 3-fold higher risk than individuals heterozygous for Factor V Leiden alone.[33,83–85] However, not all studies found a high risk for recurrence in homozygotes and double heterozygotes. In a recent study of thrombophilic families, individuals homozygous for Factor V Leiden or doubly heterozygous for Factor V Leiden and the prothrombin 20210G>A mutations did not have an increased risk for recurrence, even when the analysis was restricted to those with a first idiopathic VTE.[86] Risk estimates were limited by the small number of homozygotes and double heterozygotes included in these studies.

The risk for recurrent VTE is 4- to 5-fold higher in Factor V Leiden heterozygotes with hyperhomocysteinemia than in individuals with a Factor V Leiden allele alone.[85] In contrast, Factor V Leiden heterozygotes with high Factor VIII levels did not have a higher risk for recurrent VTE than individuals without thrombophilia.[81]

Table 3 Estimated risk of thrombotic complications: Factor V Leiden heterozygotes

Thrombotic complication	Estimated risk (odds ratio)[a]
First VTE[61–64]	3–8
Cerebral vein thrombosis[46,59]	3–5
Primary upper extremity thrombosis[b][43,45]	3–6
CVC-associated thrombosis[65]	2–3
Superficial vein thrombosis[60]	6
Pregnancy-associated VTE[66,67]	8–52
Recurrent VTE[42,68]	1.4–1.6
Pregnancy loss[66,69–71]	2–4

[a]Risk relative to individuals without Factor V Leiden.
[b]Not related to malignancy or a central venous catheter.
VTE, venous thromboembolism; CVC, central venous catheter.

Table 4 Estimated risk of thrombotic complications: Factor V Leiden homozygotes

Thrombotic complication	Risk (odds ratio)[a]
First VTE[4,33,62]	10–80
Pregnancy-associated VTE[66,73,74]	20–40
Oral contraceptive-associated VTE[75]	100
Recurrent VTE[33]	2–3
Surgery-associated VTE[76]	20
Early fetal loss[66]	3
Late fetal loss[b][77]	11

[a]Risk relative to individuals without a Factor V Leiden allele.
[b]Occurring after 12 weeks gestation.
VTE, venous thromboembolism.

Pregnancy complications

The available data indicate that Factor V Leiden is associated with a 2- to 3-fold increased relative risk for pregnancy loss and possibly other complications such as preeclampsia, intrauterine growth restriction and placental abruption. However, a Factor V Leiden mutation is at most one of multiple predisposing factors contributing to these adverse outcomes. Other genetic and en-

vironmental triggers in addition to Factor V Leiden are likely necessary for the development of pregnancy complications. Overall, the probability of a successful pregnancy outcome is high, even in homozygous women.[77]

Pregnancy loss

A large number of case-control studies found a high prevalence of Factor V Leiden heterozygosity (up to 30%) in women with unexplained recurrent pregnancy loss, compared with 1–10% of controls, suggesting a 2- to 5-fold increase in risk.[11,53–55,87] In a prospective study, Factor V Leiden heterozygotes with a history of recurrent early miscarriage had a significantly lower live birth rate (38%) than women with a similar history of unsuccessful pregnancies but without the mutation (69%).[69] However, other studies found no association with pregnancy loss and suggested the mutation has no effect on the outcome of a subsequent pregnancy after a first fetal loss.[88–90] Several meta-analyses found a strong association with fetal loss.[66,70,91,92]

Some evidence suggests that women with Factor V Leiden have a higher risk for late pregnancy loss than early first-trimester loss.[70,71,91–94] A meta-analysis found that a heterozygous mutation conferred a 2-fold increased risk for a late unexplained fetal loss and a 4-fold higher risk for loss in the second trimester compared with the first trimester.[66] Factor V Leiden heterozygosity was associated with a 4-fold increased risk of unexplained stillbirth and an 11-fold increased risk of stillbirth associated with placental infarction.[95] A recent study found that homozygous women had a 5-fold higher risk of a late fetal loss (after 12 weeks gestation) than heterozygous women and an 11-fold higher risk than women without the mutation. In contrast, the frequency of early (first trimester) loss was similar in all three groups, suggesting the mutation has a stronger association with late pregnancy loss.[77] One possible explanation is that late pregnancy losses reflect thrombosis of the placental vessels, in contrast to first-trimester losses, which are more commonly attributable to other causes. However, other evidence suggests that Factor V Leiden also increases the risk for early first-trimester loss.[70,66,96]

Other obstetric complications

Although preeclampsia, intrauterine growth restriction, and placental abruption may also involve impaired placental perfusion, their association with Factor V Leiden is controversial. The conflicting results reported in different studies may reflect the varying diagnostic and selection criteria, different ethnic groups, and small number of cases included. Factor V heterozygosity is more likely to be present in women with unexplained severe and/or recurrent adverse pregnancy outcomes.[97]

Preeclampsia is a complex disorder, and it is unlikely that a single thrombophilic defect plays a major causal role. Multiple studies including several meta-analyses found an increased risk for preeclampsia in women with Factor V Leiden.[66,98–100] Other studies found no association.[88,101–103] The conflicting results of these studies suggest that Factor V Leiden heterozygosity has at most a weak effect on the risk for preeclampsia. Factor V Leiden has a stronger association with severe and early-onset preeclampsia than with mild forms of the disease.[94,104] Heterozygous women may have a higher risk for recurrent preeclampsia in a subsequent pregnancy.[105] They may also have a higher risk of serious maternal complications and adverse perinatal outcomes than women without thrombophilia.[98,105]

The data on the risk for intrauterine growth restriction and placental abruption are more limited and also conflicting. Many studies were retrospective and underpowered to detect a significant association.[97] Several meta-analyses found a 3- to 5-fold increased risk for fetal growth restriction with a stronger association with more severe growth restriction.[91,94,106] In contrast, multiple case-control, cohort and prospective studies found no association.[88,100,101,107]

CLINICAL EXPRESSION OF FACTOR V LEIDEN

The clinical expression of Factor V Leiden is influenced by coexisting genetic and acquired thrombophilic disorders and circumstantial risk factors (Table 5). The presence of a Factor V Leiden allele increases the risk associated with other inherited and acquired thrombophilic disorders. The combination of Factor V Leiden heterozygosity and most thrombophilic disorders has a supraadditive effect on overall thrombotic risk.

Coexisting thrombophilic disorders

There is an increased prevalence of Factor V Leiden among symptomatic patients with deficiencies of Protein C, Protein S, and antithrombin and the prothrombin 202010G>A mutation.[121] Individuals with two defects have a higher risk for thrombosis than family members with a single defect. In a pooled analysis of eight case-control studies, individuals with a single Factor V Leiden or prothrombin 20210G>A allele had a 5-fold and 4-fold increase in thrombotic risk, respectively. In contrast, individuals doubly heterozygous for both mutations had a 20-fold increase in relative risk, illustrating the multiplicative effect of these two mutations on overall thrombotic risk.[108] A prothrombin 20210G>A allele was 4- to 5-fold more common in symptomatic Factor V Leiden homozygotes with VTE than in controls with no thrombotic history.[76]

A recent study found that individuals with a Factor V Leiden mutation who had a first-degree relative with a history of thrombosis had a 3-fold higher risk for VTE than Factor V Leiden carriers with a negative family history. The risk was

Table 5 Estimated risk of venous thromboembolism in Factor V Leiden heterozygotes with coexisting risk factors

Coexisting risk factor	Risk of VTE (odds ratio)[a]
PG20210G>A double heterozygote[48,108]	20 100 (pregnancy-associated VTE)
Hyperhomocysteinemia[109]	22
Obesity[110]	8
Oral contraceptives[46,111–114]	11–41 30 (cerebral vein thrombosis)
Third generation oral contraceptives[b][114]	50
HRT[115–118]	7–16
Air travel[112,119]	14–16
Minor injury[57]	50
Malignancy[45,120]	12 20 (upper extremity thrombosis)

[a]Risk relative to risk of individuals without either risk factor.
[b]Oral contraceptives containing the third-generation progestagen desogestrel.
VTE, venous thromboembolism; PG 20210G>A, prothrombin 20210 G>A mutation; HRT, hormone replacement therapy.

increased 18-fold in those with two or more symptomatic relatives. The family history had additional value in predicting risk regardless of Factor V genotype, suggesting the presence of unknown genetic risk factors.[122]

Factor V Leiden heterozygotes with high Factor VIII levels (>150% of normal) have a 2- to 3-fold higher incidence of VTE than those with a heterozygous mutation alone.[123] The risk of thrombosis is also increased in patients with Factor V Leiden and hyperhomocysteinemia. In the Physicians' Health Study, individuals with either a Factor V Leiden allele or hyperhomocysteinemia had a 3- to 4-fold increased risk for idiopathic VTE compared with those with neither abnormality. The relative thrombotic risk was increased 22-fold in individuals with both abnormalities.[109]

It is still unclear to what extent Factor V Leiden increases the thrombotic risk associated with malignancy. A large population-based case-control study found that Factor V Leiden heterozygotes with malignancy had a 2-fold higher risk for VTE than patients with cancer without the mutation and a 12-fold higher risk than those with neither risk factor.[44] Individuals with cancer and heterozygosity for Factor V Leiden or the prothrombin 20210G>A mutation had a 20-fold higher risk of developing an upper extremity thrombosis than patients with cancer with neither prothrombotic mutation.[44] A Factor V Leiden mutation may also contribute to central venous catheter-related thrombosis in patients with cancer.[124] In contrast, Factor V heterozygosity did not increase the risk for VTE during adjuvant chemotherapy for breast or gastrointestinal malignancy.[125]

Circumstantial risk factors

Other predisposing factors for VTE include central venous catheter use, pregnancy, estrogen contraception, HRT, SERMs, travel, injury, age, obesity, and surgery. Factor V Leiden interacts with these environmental risk factors to increase the risk of VTE. At least 50% of thrombotic episodes in individuals with Factor V Leiden are provoked by additional predisposing factors.[37] In a large cohort of symptomatic Factor V Leiden homozygotes, the initial VTE was associated with circumstantial risk factors in 81% of women and 29% of men. Oral contraceptives and pregnancy were the most common predisposing factors in symptomatic women. Thirteen percent of major surgeries were complicated by VTE, suggesting an approximately 20-fold increase in risk.[76]

Central venous catheters

Factor V Leiden heterozygotes have a 2- to 3-fold increased risk for central venous catheter-related thrombosis.[65] The mutation increases the risk for catheter-related thrombosis in patients with advanced or metastatic breast cancer and those undergoing allogeneic bone marrow transplantation.[126,127]

Pregnancy

Factor V Leiden is found in 20–46% of women with pregnancy-associated VTE.[48–51] The mutation is associated with an 8- to 52-fold increase in thrombotic risk during pregnancy and the puerperium, when compared with nonpregnant women without thrombophilia.[66,67] The overall risk is higher in women with coexisting acquired or circumstantial risk factors, such as obesity and advanced maternal age.[51] Women with multiple or homozygous thrombophilic defects have the highest risk for pregnancy-associated VTE. The risk is increased 20- to 40-fold in women with homozygous Factor V Leiden.[66,73,74] In one study, the risk for pregnancy-associated VTE was increased 9-fold in Factor V Leiden heterozygotes, 15-fold in prothrombin 20210G>A heterozygotes, and more than 100-fold in double heterozygotes, illustrating the marked increase in overall risk when thrombophilic mutations are combined.[48] In studies of thrombophilic families, VTE complicated 4% of pregnancies in women doubly heterozygous for Factor V Leiden and the prothrombin 20210G>A mutation and 16% of pregnancies in Factor V Leiden homozygotes, compared with 0.5% of those of unaffected relatives.[74,128] However, another family study found a lower risk in doubly heterozygous women. There were no VTE events during pregnancy, and postpartum VTE complicated 1.8% of pregnancies.[129] The prevalence of pregnancy-related VTE was 9% in a series of unselected homozygous women.[130]

Although Factor V Leiden increases the relative risk for VTE during pregnancy and the pueperium, the absolute risk in asymptomatic heterozygotes is not well defined. The available evidence suggests that the absolute incidence of thrombosis during pregnancy is low. Two prospective studies of unselected pregnant women screened for Factor V Leiden both reported very low rates of VTE in heterozygotes (1.1% and 0%, respectively).[101,131] No VTE events occurred during pregnancy or postpartum among a cohort of 129 women with Factor V Leiden identified by population screening.[35] In several retrospective studies and meta-analyses, the estimated risk for pregnancy-associated VTE for heterozygous women was in the range of 1 in 125–400 pregnancies. Women with homozygous Factor V Leiden or combined thrombophilia have a much higher probability of VTE, in the range of 1 in 20 to 1 in 100 pregnancies.[66,48,73,74]

Oral contraceptives

The use of oral contraceptives substantially increases the risk for VTE in women heterozygous for Factor V Leiden. A heterozygous mutation is found in 20–60% of women with a history of VTE during oral contraceptive use.[49,52] In the Leiden Thrombophilia study, the risk for VTE was increased 4-fold in oral contraceptive users and 7-fold in women heterozygous for Factor V Leiden. However, the risk was increased 30-fold in heterozygous women who used oral contraceptives, indicating a multiplicative rather than additive effect on overall thrombotic risk.[75] The supraadditive effect of a Factor V Leiden allele and oral contraceptives was confirmed in other studies and a meta-analysis with odds ratios ranging from 11 to 41 for the combination of both risk factors.[111–113,115] Heterozygous women who use oral contraceptives have a 30-fold higher risk for cerebral vein thrombosis than nonusers without the mutation.[46]

The risk for VTE is increased more than 100-fold in women homozygous for Factor V Leiden who use oral contraceptives.[75] The risk is also markedly increased in oral contraceptive users who are doubly heterozygous for Factor V Leiden and the prothrombin 20210G>A mutation with reported odds ratios ranging from 17 to 110.[132] Women with inherited thrombophilic disorders such as Factor V Leiden tend to develop thrombotic complications sooner, with a much higher risk for thrombosis during the first year of oral contraceptive use.[133]

Oral contraceptives containing the third-generation progestagen desogestrel are associated with a 2-fold higher risk for VTE than second generation preparations, and the risk is especially high in Factor V Leiden heterozygotes. The risk was increased 50-fold in Factor V Leiden heterozygotes who used third-generation preparations, compared with women without the mutation who were not using oral contraceptives.[114] Despite the marked increase in relative risk, the absolute incidence of VTE may still be low because of the low baseline risk in young healthy women. For example, the combination of Factor V Leiden and oral contraceptives is estimated to result in an

additional 28 VTE events per 10,000 women per year.[75] In a prospective cohort study of asymptomatic women with Factor V Leiden, the risk of VTE was 1.8% per year of oral contraceptive use.[134] Long-term oral contraceptive use without thrombotic complications has been reported, underscoring the multifactorial etiology of VTE.[135]

Hormone replacement therapy

The relative risk for VTE is increased 2- to 4-fold in current users of HRT compared with nonusers.[136,137] The landmark Women's Health Initiative randomized trial of estrogen and progesterone HRT versus placebo in postmenopausal women found that HRT was associated with a 2-fold increased risk for VTE.[137] Women with Factor V Leiden who use HRT have a 7- to 15-fold increased risk for VTE compared with nonusers without the mutation.[115–118] In a prospective cohort study of asymptomatic women with Factor V Leiden, the risk of VTE was 2.9% per year of HRT use.[134] The 2- to 4-fold increase in thrombotic risk with HRT is similar to the risk observed with oral contraceptives. However, because of the higher baseline incidence of VTE, the absolute risk is much higher in older postmenopausal women with and without Factor V Leiden (Fig. 1). The estimated absolute incidence of VTE in postmenopausal women with Factor V Leiden who use HRT ranges from 8 to 15 VTE events per 1000 women per year, compared with two VTE events per 1000 women per year for nonusers without the mutation.[117,118]

There is increasing evidence that transdermal estrogen is associated with a lower thrombotic risk than the oral route in postmenopausal women.[138–140] Preliminary data suggest the use of transdermal estrogen may not confer additional thrombotic risk in women with thrombophilic mutations. In a case-control study, postmenopausal women with Factor V Leiden who used oral estrogen had a 16-fold higher risk for VTE than nonusers without the mutation. In contrast, women with Factor V Leiden who used transdermal estrogen had a lower thrombotic risk similar to that of women with the mutation who did not use estrogen. Among women with Factor V Leiden, the use of oral estrogen was associated with a 4-fold higher risk for VTE than transdermal estrogen.[141] However, there are no prospective randomized trials confirming the safety of transdermal HRT in women with thrombophilia.

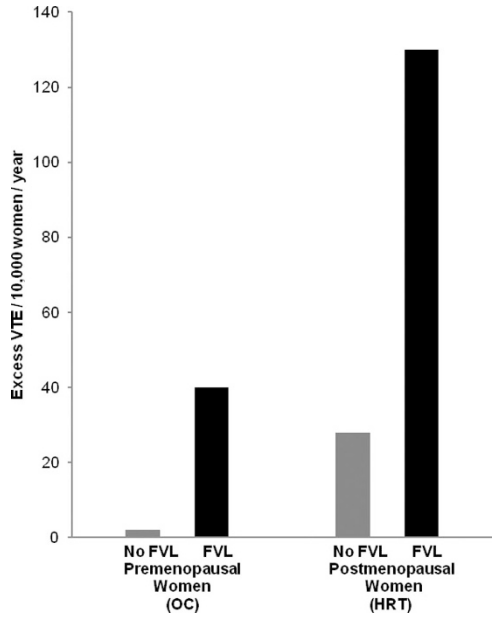

Fig. 1. Estimated excess venous thromboembolism (VTE) events attributable to oral contraceptive (OC) use in premenopausal women with and without Factor V Leiden (FVL) and to hormone replacement therapy (HRT) in postmenopausal women with coronary heart disease with and without FVL. Estimates of excess VTE events assume a baseline incidence of 1 VTE/10,000 women/year in premenopausal women and 23 VTE/10,000 women/year in postmenopausal women without either risk factor.[75,117,136]

Selective estrogen receptor modulators

SERMs such as tamoxifen and raloxifene are associated with a 2- to 3-fold increased risk of VTE, similar to the risk of HRT. The risk for VTE in women with Factor V Leiden who use SERMs is not well defined but likely higher than that associated with SERM use alone, in light of the interaction of the mutation with HRT. Factor V Leiden had no significant effect on the risk of VTE associated with tamoxifen in two case-control studies of high-risk healthy women enrolled in breast cancer prevention trials.[142,143] However, both studies were limited by the small number of cases included. The risk for thromboembolic complications during adjuvant tamoxifen for early-stage breast cancer was approximately 5-fold higher in Factor V Leiden heterozygotes than in women without the mutation receiving the same therapy. Factor V Leiden heterozygosity was found in 19% of women who developed thromboembolism compared with 5% of women without these complications.[144]

Travel

Extended travel is a risk factor for VTE, and the risk is higher in individuals with Factor V Leiden. The combination of air travel and thrombophilia (including Factor V Leiden) is associated with a 14- to 16-fold increased risk for VTE.[112,119]

Age

The risk and incidence of VTE increases with age. The risk increases at a greater rate with advancing age in individuals with a Factor V Leiden mutation, suggesting that thrombosis involves acquired and genetic predisposing factors.[145] In the Physicians' Health Study, the mutation was found in approximately one third of men older than 60 years with an initial spontaneous thrombotic event.

In a population-based cohort study, the risk for VTE was significantly increased only among Factor V Leiden heterozygotes older than 60 years (relative risk 3.6).[35] Another prospective study found that the absolute risk for VTE in unselected individuals with Factor V Leiden increased with age, body mass index (BMI), and smoking. The 10-year risk for VTE among heterozygotes was 10% in smokers older than 60 years with a BMI >30 kg/m^2, in contrast to a $<1\%$ risk in nonsmokers younger than 40 years who were not overweight. The corresponding absolute 10-year thrombotic risks for Factor V Leiden homozygotes with and without these risk factors were 51% and 3%, respectively.[72]

Minor injury

A large population-based study found that minor leg injuries are associated with a 5-fold increased risk for VTE. Factor V Leiden carriers with a minor leg injury had a 50-fold higher thrombotic risk than individuals without these risk factors.[57]

Obesity

Obesity increases the risk of VTE in Factor V Leiden heterozygotes. In one study, obesity (BMI >30 kg/m^2) was associated with a 2.5-fold increased risk of VTE. The risk was increased 8-fold in obese individuals with Factor V Leiden and approximately 6-fold in overweight (BMI ≥ 25 and <30 kg/m^2) individuals with the mutation.[110]

Surgery

It is still unclear to what extent Factor V Leiden adds to the overall thrombotic risk associated with surgery. In a study of unselected families with Factor V Leiden, surgery was a common predisposing factor for VTE in relatives with the mutation.[37] Several studies suggested the mutation was associated with a 5-fold increased risk of symptomatic VTE after orthopedic and other surgery.[41,146,147] Factor V Leiden heterozygotes undergoing surgery had an approximately 13-fold higher risk for upper extremity DVT than controls with neither risk factor.[44] In contrast, other studies found no significant association.[148,149] Factor V Leiden homozygotes had an approximately 20-fold increased risk for VTE after surgery, especially urologic and orthopedic procedures.[76]

THROMBOSIS NOT CONVINCINGLY ASSOCIATED WITH FACTOR V LEIDEN

Arterial thrombosis

The role of Factor V Leiden in arterial disease is controversial with conflicting results from different studies. The available evidence suggests that Factor V Leiden is not a major risk factor for arterial thrombosis. The majority of myocardial infarctions and strokes occur in the presence of established cardiovascular risk factors such as hypertension, hyperlipidemia, diabetes, and smoking. The contribution of a single prothrombotic mutation to these complex diseases is likely to be small. However, it is possible that Factor V Leiden may interact with other genetic and environmental risk factors to promote arterial thrombosis. Most studies of unselected adult populations found no association between Factor V Leiden and an increased risk for myocardial infarction or stroke.[150–152] In contrast, a large meta-analysis found that a Factor V Leiden allele conferred a moderately increased risk for coronary disease and myocardial infarction.[153] There is also data suggesting the mutation may contribute to the risk for arterial thrombotic events in specific subgroups of individuals. The risk for arterial thrombosis in Factor V Leiden homozygotes is unknown, as very few were included in the available studies.

Myocardial infarction

There is some evidence that Factor V Leiden may contribute to myocardial infarction in younger individuals and in those with other traditional cardiovascular risk factors. One study found an increased risk for myocardial infarction in young women with other cardiovascular risk factors, particularly smoking. Factor V Leiden heterozygotes who smoked had a 30-fold increased risk for myocardial infarction compared with women with neither risk factor.[154] In another recent study, a Factor V Leiden mutation was associated with an approximately 2-fold increased risk of premature myocardial infarction (before the age of 45 years). The combination of hypercholesterolemia and a Factor V Leiden allele increased the risk approximately 4-fold.[155] Other studies also found the combination of Factor V Leiden and one or more established cardiovascular risk factors substantially increased the risk of acute myocardial infarction. The combination of a thrombophilic mutation and smoking conferred the highest risk, with reported odds ratios ranging from 6 to 18.[156,157]

Stroke

Most studies of unselected adult populations did not find a significant association between Factor V Leiden and ischemic stroke.[150,158] The prevalence of Factor V Leiden was similar in unselected individuals with severe carotid atherosclerosis and healthy controls.[159] Although Factor V Leiden is not a major risk factor for stroke, it may contribute in certain populations. Factor V Leiden was associated with a 3-fold increased risk for stroke in individuals younger than 45–50 years. The risk was even higher among women in this age group (odds ratio range: 4–6).[160,161]

The interaction of Factor V Leiden with other cardiovascular risk factors may increase the risk for ischemic stroke. Young women with a Factor V Leiden allele who use oral contraceptives have a 9- to 13-fold increased risk for stroke, compared with women without either risk factor.[162,163] Women younger than 60 years with Factor V Leiden who smoked had a approximately 9-fold higher risk of stroke than nonsmokers without the mutation.[158] In these studies, Factor V Leiden increased the risk of stroke only in women with these other risk factors. The combination of a Factor V Leiden allele and one or more traditional cardiovascular risk factors was associated with a approximately 11-fold increase in stroke risk.[160] A recent study found that Factor V Leiden heterozygotes with a first ischemic stroke were significantly more likely to have widespread atherosclerotic vascular disease and multiple silent infarctions than stroke patients without the mutation.[164] A high prevalence of Factor V Leiden or the prothrombin 20210G$>$A mutation was found in patients with a cryptogeneic stroke and a patent foramen ovale (PFO), suggesting the possibility of paradoxical embolism.[165] A meta-analysis found that individuals with a Factor V Leiden or prothrombin 20210G$>$A mutation had a approximately 2-fold increased risk for PFO-related stroke compared with both control and non-PFO-associated stroke patients.[166]

GENOTYPE-PHENOTYPE CORRELATIONS

Individuals homozygous for Factor V Leiden have a higher risk for thrombosis than heterozygotes (Table 4). However, the clinical course of an acute thrombotic episode is not more severe or resistant to anticoagulation in homozygotes than in heterozygotes. The thrombotic risk in homozygotes is also substantially lower than the risk of homozygous Protein C or S deficiency.

Factor V Leiden and Factor V deficiency

"Pseudohomozygous" Factor V Leiden occurs in individuals who are doubly heterozygous for Factor V Leiden and a Factor V null mutation. Coinheritance of a Factor V null allele occurs in approximately 1:1000 individuals heterozygous for Factor V Leiden. Rather than attenuating the effect of a Factor V Leiden allele, a coexisting Factor V deficiency enhances it, producing a more severe APC-resistant phenotype, reflected by an extremely low APC resistance ratio, indistinguishable from that of a homozygous mutation.[167] The diagnosis of pseudohomozygous Factor V Leiden is based on the combination of a heterozygous Factor V Leiden mutation, low-plasma Factor V activity level (approximately 50% of normal), and a low APC resistance ratio in the range typical for a homozygous mutation. Factor V Leiden pseudohomozygotes seem to have an increased thrombotic risk and clinical phenotype similar to that of Factor V

Leiden homozygotes.[168] Pseudohomozygous APC resistance has also been reported in individuals doubly heterozygous for Factor V Leiden and Factor V Cambridge.[169]

In rare case, both a null allele and Factor V Leiden mutation occur on the same chromosome in *cis* configuration. In these individuals, the resulting quantitative Factor V deficiency prevents expression of the Factor V Leiden mutation.[170]

HR2 haplotype

A Factor V gene haplotype (HR2) defined by the R2 polymorphism (4070A>G) is in complete linkage disequilibrium with Factor V Leiden and may confer mild APC resistance[6] (Table 6). It may interact with the Factor V Leiden mutation to produce a more severe APC resistance phenotype.[171,177] Coinheritance of the HR2 haplotype increased the risk for VTE associated with Factor V Leiden by approximately 3-fold.[172] It is controversial whether the HR2 haplotype is associated with an increased thrombotic risk in the absence of Factor V Leiden.[62,171,178,179]

GENETICALLY RELATED (ALLELIC) DISORDERS

Two different mutations at the Arg306 APC cleavage site in Factor V have been reported, only one of which is associated with APC resistance (Table 6). A G-to-C point mutation, which predicts the replacement of Arg with Thr at position 306 (Factor V Cambridge), was identified in a British individual with a history of thrombosis and APC resistance in the absence of a Factor V Leiden mutation.[173] However, the mutation was not found in several series of individuals with VTE or healthy controls, suggesting it is a very rare Factor V variant.[173,180]

A different mutation in the same codon predicting an Arg-to-Gly substitution was identified in two Chinese individuals with a history of thrombosis. The Arg306Gly mutation (Factor V Hong Kong) was not associated with APC resistance in the single individual tested with a coagulation screening test.[174] The clinical significance of the Arg306Gly mutation is unclear because it was found in a similar percentage of healthy Hong Kong Chinese blood donors (4.5%) and individuals with a history of thrombosis (4.7%).[174,175]

A missense mutation in Factor V, Ile359Thr (Factor V Liverpool) was identified in several family members with recurrent thrombosis.[181] Factor V Liverpool showed resistance to APC and impaired cofactor activity for APC similar to that of Factor V Leiden.[176] Although the available evidence suggests that the Arg306Thr, Arg306Gly, and Ile359Thr mutations are not major risk factors for thrombosis, they may contribute when combined with other genetic or acquired risk factors. There are anecdotal reports of double heterozygosity for Factor V Leiden and Factor V Cambridge.[169]

PENETRANCE

Factor V Leiden heterozygotes identified by general population screening have a low absolute incidence of VTE of approximately two VTE events/1000 persons/year and a cumulative incidence of VTE of 6.5% at 65 years. The absolute incidence in homozygotes is in the range of 15 VTE events/1000 persons/year.[35,72]

The risk for thrombosis is higher in studies of asymptomatic Factor V Leiden heterozygotes from thrombophilic families than in unselected individuals identified by population screening. Multiple retrospective studies of relatives of unselected symptomatic and asymptomatic Factor V Leiden heterozygotes consistently found a low thrombotic risk, even during high-risk circumstances such as pregnancy surgery and immobilization. The absolute incidence of VTE ranged from 0.19%/year to 0.49%/year, compared with 0.05%/year to 0.10%/year in individuals without the mutation.[37,63,182,183] VTE occurred in 7–12% of relatives with Factor V Leiden heterozygosity, compared with 2–3% of relatives without the mutation, consistent with other estimates that the lifetime risk for thrombosis is approximately 10%. Multiple prospective cohort studies found a similar low incidence of VTE in asymptomatic Factor V Leiden heterozygotes, in the range of 0.1%/year–0.67%/year. At least 50% of thrombotic events were associated with other risk factors.[134,184,185] The absolute risk for VTE is much higher in relatives with homozygous Factor V Leiden (1.30%/year) and those with Factor V Leiden in combination with other thrombophilic defects (0.62%/year–1.54%/year).[63]

DIFFERENTIAL DIAGNOSIS

The differential diagnosis of VTE includes other inherited and acquired thrombophilic disorders. Because these disorders are not clinically distinguishable, laboratory testing is required for diagnosis in each case.

Inherited disorders

- The **prothrombin 20210G>A** mutation in the 3′ untranslated region of the gene encoding prothrombin is found in 2% of the general population, 6% of individuals presenting with a first VTE, and up to 18% of individuals with a personal and family history of thrombosis.[186] Double heterozygosity for both Factor V Leiden and the prothrombin 20210G>A mutation occurs in approximately 1 in 1000 in the general population and 1–5% of individuals with VTE.[83,108]

Table 6 Selected Factor V allelic variants

Allelic variant	DNA nucleotide change	Protein amino acid change	Clinical significance
R2 polymorphism[171,172]	4070 A>G	His1299Arg	Increases APC resistance and VTE risk associated with Factor V Leiden
Factor V Cambridge[6,173]	1091 G>C	Arg306Thr	Uncertain; rare variant; confers APC resistance
Factor V Hong Kong[6,174,175]	1090 A>G	Arg306Gly	Uncertain, found in 4.7% Hong Kong Chinese with and 4.5% without VTE
Factor V Liverpool[6,176]	1250 T>C	Ile359Thr	Uncertain; confers APC resistance
Factor V Leiden[61,62]	1691 G>A	Arg506Gln	APC resistance and 3- to 8-fold increased risk of VTE

- Inherited deficiencies or abnormalities of the natural anticoagulant **Proteins C, Protein S, and antithrombin** are approximately 10-fold less common than Factor V Leiden, with a combined prevalence of <1% of the population.[187] Anticoagulant protein deficiencies are found in 1–3% of individuals with a first VTE.
- Elevated levels of **lipoprotein(a)** are associated with premature atherosclerosis and may also be a risk factor for venous thrombosis.
- Hereditary **dysfibrinogenemias** are rare and infrequently cause thrombophilia and thrombosis.
- A specific point mutation in the *MTHFR* **gene (677C>T)** encoding methylenetetrahydrofolate reductase results in a variant thermolabile enzyme with reduced activity for the remethylation of homocysteine. Homozygosity for the 677C>T mutation occurs in 10–20% of the general population and predisposes to mild hyperhomocysteinemia, usually in the setting of suboptimal folate levels. However, the *MTHFR* 677C>T mutation is not associated with an increased risk for VTE independent of plasma homocysteine levels.[188]

Acquired disorders

- **APC resistance:** Although Factor V Leiden accounts for 95% of cases of APC resistance, 5% of cases occur in the absence of a Factor V Leiden allele. Depending on the screening assay used, acquired APC resistance can be caused by high Factor VIII levels, pregnancy, use of oral contraceptives, or the presence of antiphospholipid antibodies. APC resistance in the absence of Factor V Leiden is also a risk factor for VTE.[189,190] In rare cases, other genetic abnormalities may produce an APC-resistant phenotype.[173]
- **Antiphospholipid antibodies** comprise a heterogeneous group of autoantibodies directed against proteins bound to phospholipid. Criteria for the diagnosis of the **antiphospholipid antibody syndrome** include the following:

  A history of arterial or VTE

  OR one or more unexplained fetal deaths (>10 weeks of gestation) or three or more unexplained miscarriages

  AND

  Persistent anticardiolipin antibodies OR antibeta2-glycoprotein 1 antibodies OR a lupus inhibitor on at least two occasions of testing 3 months apart.

Persistent lupus inhibitors, high-titer IgG anticardiolipin antibodies, and antibeta2 glycoprotein 1 antibodies are most strongly associated with arterial and VTE.[191]

- **A high level of homocysteine** occurs in 10% of individuals with a first VTE and is associated with a 2- to 3-fold increase in relative risk. The plasma level of homocysteine reflects genetic and environmental factors and is more directly associated with thrombotic risk than molecular genetic testing of the *MTHFR* gene.
- **High clotting factor levels:** An elevated **Factor VIII level** >150% of normal is associated with a 4- to 5-fold increase in risk of VTE and also increases the risk for recurrence.[192–194] High levels of both **Factor IX and Factor XI** are associated with a 2-fold increased risk of VTE.[195,196] Elevated levels of **both Factor VIII and Factor IX** confer an 8-fold increased risk of VTE.[195] A high **prothrombin** level >110–115% of normal is associated with a 2-fold increased risk for VTE in the absence of prothrombin 20210G>A heterozygosity.[111,186]
- Although abnormalities in other coagulation or fibrinolytic proteins including PAI-1, tissue factor pathway inhibitor, thrombin activatable fibrinolysis inhibitor, and Protein Z have been reported in patients with venous thrombosis, a causal association has not been established.

MANAGEMENT

Evaluation after initial diagnosis

Individuals heterozygous or homozygous for Factor V Leiden should be tested for other thrombophilic disorders to assess the risk for thrombosis. Factor V Leiden often coexists with other inherited and/or acquired disorders. Testing should include the following:

- DNA testing for the prothrombin 20210G>A mutation.
- Serologic assays for anticardiolipin antibodies and antibeta2glycoprotein 1 antibodies.
- Multiple phospholipid-dependent coagulation assays for a lupus inhibitor.

The evaluation of high-risk individuals (i.e., those with a history of recurrent VTE, especially at a young age, or those with a strong family history of VTE at a young age) should also include assays of the following:

- Protein C activity.
- Antithrombin activity.
- Free Protein S antigen or Protein S activity.

Although routine measurement of Factor VIII levels is not recommended, testing may be useful in selected cases. It is still unclear whether clotting factor levels should be included in a thrombophilia evaluation.[197] Measurement of homocysteine levels is no longer routinely included because lowering homocysteine levels with vitamin supplementation has not been shown to reduce the risk of recurrent venous or arterial thrombosis.[198] There is also no clinical rationale for DNA testing for *MTHFR* polymorphisms.

Treatment of manifestations

The management of individuals with Factor V Leiden depends on the clinical circumstances.

The first acute VTE should be treated according to standard guidelines with a course of low molecular weight heparin (LMWH) or intravenous unfractionated heparin. Warfarin is started concurrently with LMWH (except during pregnancy) and monitored with the international normalized ratio (INR).[199] A target INR of 2.5 (therapeutic range: 2.0–3.0) provides effective anticoagulation, even in individuals with homozygous Factor V Leiden.[200] LMWH and warfarin should be overlapped for at least 5 days and until the INR has been within the therapeutic range on two consecutive measurements over 2 days.

Decisions regarding the duration of anticoagulation should be based on an assessment of the risks for VTE recurrence and anticoagulant-related bleeding. Approximately 30% of individuals with an incident VTE develop recurrent thrombosis within the subsequent 10 years.[201] Individuals with a spontaneous VTE and no identifiable provoking factors require a longer course of anticoagulation. Anticoagulation for at least 3 months is recommended for individuals with VTE associated with a transient (reversible) risk factor.[199] The optimal duration of anticoagulation for Factor V Leiden heterozygotes is debated. Factor V Leiden heterozygosity is generally not an indication for long-term anticoagulation in the absence of other indications. The presence of hereditary thrombophilia was not a major factor

determining the duration of anticoagulation in the 2008 American College of Chest Physicians Guidelines on Antithrombotic Therapy, based on evidence that these disorders are not major determinants of recurrence risk.

Long-term anticoagulation is recommended for individuals with a first or recurrent unprovoked VTE and no risk factors for bleeding with good anticoagulation monitoring. It should also be considered in individuals with homozygous Factor V Leiden or with multiple thrombophilic disorders.[199] In these individuals at high risk for recurrence, the potential benefits from long-term warfarin may outweigh the bleeding risks.

Prevention of primary manifestations

In the absence of a history of thrombosis, long-term anticoagulation is not recommended for asymptomatic Factor V Leiden heterozygotes, because the 1–3%/year risk of major bleeding from warfarin is greater than the estimated <1%/year risk for thrombosis. Because the initial VTE in Factor V Leiden heterozygotes occurs in association with other circumstantial risk factors in 50% of cases, a course of prophylactic anticoagulation during high-risk clinical settings such as surgery, pregnancy, or prolonged immobilization may prevent some of these episodes. However, there is no evidence confirming the benefit of primary prophylaxis for asymptomatic Factor V Leiden heterozygotes. Decisions regarding prophylactic anticoagulation should be based on a risk/benefit assessment in each individual case. Recommendations for prophylaxis at the time of surgery and other high-risk situations are available in consensus guidelines.[202]

Pregnancy

There is no consensus on the optimal management of women with Factor V Leiden during pregnancy. Prophylactic anticoagulation is not routinely recommended in asymptomatic heterozygous women with no history of thrombosis.[29,30] These women should be warned about potential thrombotic complications and counseled about the risks and benefits of anticoagulation during pregnancy. They should also be offered a 4- to 6-week course of anticoagulation after delivery, as the greatest thrombotic risk is in the initial postpartum period.[29] Prophylactic anticoagulation is recommended for women with Factor V Leiden and a history of prior unprovoked VTE. LMWH (or unfractionated heparin) is given during pregnancy, followed by a 4- to 6-week course of postpartum anticoagulation.[29,30] Prophylactic anticoagulation should also be considered for heterozygous women with a prior estrogen-related thrombosis (associated with oral contraceptive use or pregnancy) who are also at increased risk for recurrence.[29,203,204] It may also be appropriate for asymptomatic homozygotes or women doubly heterozygous for Factor V Leiden and prothrombin 20210G>A mutations or with other combined thrombophilic defects, especially those with coexisting circumstantial risk factors.[29,30]

Prevention of pregnancy complications

The results of observational studies and two randomized trials suggest that prophylaxis with LMWH may improve pregnancy outcome in women with thrombophilia and a history of unexplained recurrent miscarriage or late pregnancy loss.[205–207] In contrast, other studies found no benefit of LMWH on pregnancy outcome.[208,209] There are no prospective randomized trials including an untreated control group confirming the benefit of LMWH in preventing pregnancy loss in women with inherited thrombophilia. American College of Chest Physicians 2008 and recent obstetric consensus guidelines and expert opinion do not routinely recommend antithrombotic therapy for women with Factor V Leiden and pregnancy loss because of a lack of sufficient evidence confirming benefit.[29–31,97] Antithrombotic prophylaxis may be considered in selected women with Factor V Leiden and unexplained recurrent or late pregnancy loss after an informed discussion of the risks and the limited data suggesting benefit. Assessment of the maternal thrombotic risk during pregnancy should be incorporated into the decision regarding prophylaxis.

There is currently no evidence that antithrombotic therapy reduces the risk of preeclampsia or other pregnancy complications in women with thrombophilia including Factor V Leiden. LMWH is not routinely recommended for thrombophilic women with a history of preeclampsia or other adverse pregnancy outcomes.[29,30]

GENETIC COUNSELING

Because of the high prevalence of Factor V Leiden in the general population, the genetic status of both parents and/or the reproductive partner of an affected individual must be determined before information regarding potential risks to siblings or offspring can be provided.

Proband heterozygous for Factor V Leiden

Heterozygosity for Factor V Leiden and the associated risk for VTE are inherited in an autosomal dominant manner. If one parent is heterozygous, each sibling is at 50% risk of being heterozygous for the Factor V Leiden allele. If one parent is homozygous, each sibling has 100% chance of inheriting a heterozygous mutation. If both parents are heterozygous, each sibling of the proband has a 25% chance of being homozygous, a 50% chance of being heterozygous, and a 25% chance of inheriting both normal Factor V alleles.

Each offspring of a Factor V Leiden heterozygote has a 50% chance of inheriting the mutation. If the proband's reproductive partner is heterozygous, each offspring has a 25% chance of being homozygous, a 50% chance of being heterozygous, and a 25% chance of inheriting both normal Factor V alleles.

Proband homozygous for Factor V Leiden

Homozygosity for Factor V Leiden and the associated much greater thrombotic risk are inherited in an autosomal recessive manner. If both parents are heterozygotes, the siblings of the proband have a 25% chance of being homozygous, a 50% chance of being heterozygous, and a 25% chance of inheriting both normal Factor V alleles. If one parent is homozygous and the other parent is heterozygous, each sibling has a 50% chance of being homozygous for Factor V Leiden and a 50% chance of being heterozygous.

Each offspring of a Factor V Leiden homozygote has 100% chance of inheriting one Factor V Leiden allele. If the proband's reproductive partner is heterozygous, each offspring has a 50% chance of being homozygous and a 50% chance of inheriting a heterozygous mutation.

Testing at-risk family members

The genetic status of asymptomatic at-risk family members can be established using molecular testing; however, the indications for family testing are unresolved. Because heterozygosity for Factor V Leiden confers only a mildly increased risk for thrombosis, routine testing of at-risk family members is not recommended. In the absence of evidence that early diagnosis of Factor V Leiden reduces morbidity or mortality, decisions regarding testing should be made on an individual basis. Clarification of Factor V Leiden allele status may be useful in women considering hormonal contraception or pregnancy, and

in families with other known thrombophilic disorders and/or a strong history of recurrent venous thrombosis at young age. At-risk family members often request testing before exposure to recognized risk factors or from a desire to know their status. Asymptomatic relatives younger than 18 years are not usually tested because thrombosis rarely occurs before young adulthood, even in homozygotes.

Individuals requesting testing for Factor V Leiden and those identified as heterozygotes or homozygotes should be counseled on the signs and symptoms of VTE that require immediate medical attention, and the potential need for prophylactic anticoagulation in high-risk circumstances. They should be informed that although Factor V Leiden is an established risk factor, it does not predict thrombosis with certainty because the clinical course is variable even within the same family.

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