Incidence of venous thromboembolism in first-degree relatives of patients with venous thromboembolism who have factor V Leiden

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Summary

The factor V Leiden (FVL) mutation, a genetic abnormality with an autosomal mode of inheritance, is associated with an increased risk of venous thromboembolism (VTE). We aimed to determine the annual incidence of VTE in first-degree relatives of patients with VTE and FVL and to identify factors in patients and the relatives that influence this incidence. In this retrospective and prospective cohort study, the incidence of objectively diagnosed first episodes of VTE was assessed in 553 first-degree relatives of 161 patients with acute VTE and FVL. The annual incidence of VTE was 0.43% (95% CI, 0.3 to 0.56) with FVL and 0.17 % (95% CI, 0.07 to 0.27) without FVL (relative risk of 2.5, 95% CI, 1.3 to 4.7). A majority (70%) of episodes of VTE were provoked, and this proportion was similar with and without FVL. A larger proportion of VTE was provoked in women (83%) that in men (33%), with the difference accounted for by pregnancy and use of oral contraceptives. The proportion of pregnancies complicated by VTE was 3.9% (95% CI, 2.0–5.8) with FVL and 1.4% (95% CI, 0.04–2.7) without FVL. FVL is associated with a two- to three-fold increase in VTE in first-degree relatives of patients with VTE. No subgroup of relatives was identified who require more than routine prophylaxis because of a particularly high risk of VTE.

Keywords

Familial thrombosis, venous thrombosis, factor V Leiden

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Introduction

The presence of factor V Leiden (FVL) (1, 2) in patients who have venous thromboembolism (VTE) may have two important clinical implications. First, after completing anticoagulation for a first episode of acute VTE, FVL might be associated with an increased risk of recurrence, which could favour more prolonged treatment of such patients. However, current evidence does not suggest that FVL, particularly in its heterozygous state, is a clinically important risk factor for recurrence (3–10). Second, as FVL is a genetic abnormality with an autosomal mode of inheritance, half of patient's first-degree relatives are expected to also have FVL and, consequently, to have an associated increase in their risk of VTE. Consistent with the association between FVL and VTE in the general population (2, 11–13), an increase in the incidence of VTE among relatives who have FVL compared with relatives who do not have FVL has been observed (14–19). However, estimates of the risk of VTE in family members of patients with FVL are uncertain as previous studies that evaluated this risk were small. Furthermore, previous studies were performed in either The Netherlands (14, 16, 17) or Italy (15, 18) and their findings can perhaps not be generalized to other populations.

We performed a study, with retrospective and prospective components, to assess the incidence of venous thrombosis in first-degree relatives of patients with a first episode of VTE who had FVL. Secondary objectives of this study were: i) to explore whether the occurrence of VTE increased similarly with age in relatives with, and without, FVL; ii) to estimate the absolute frequency of VTE in relatives during and after a pregnancy; and iii) to examine whether clinical features of the index case of thrombosis influenced the incidence of VTE in the relatives.

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Methods

Population

Study subjects were the first-degree relatives (i.e. parents, siblings, children) of consecutive patients (index cases) who were diagnosed with an acute episode of VTE (first or recurrent) at the University Hospital in Brest, France, and who had FVL. Index cases were prospectively recruited and had to meet all of the following eligibility criteria: age 16 years or older; acute VTE documented by objective testing (see diagnostic criteria below); presence of at least one living first-degree relative; and willingness to provide written informed consent to participate in the study and to allow one or more of their first-degree relatives to be approached for the study. First-degree relatives had to meet the following eligibility criteria: age 16 years or older; willingness to provide a blood sample to determine if they had FVL; and willingness to provide written informed consent. The study was approved by the research ethics committee of Brest Hospital.

Diagnosis of VTE

VTE in index cases or first-degree relatives was defined as acute deep vein thrombosis (DVT) and/or pulmonary embolism (PE). DVT of the proximal and/or distal deep veins was diagnosed by lack of full compression on compression ultrasonography, or an intraluminal filling defect on venography. PE was diagnosed by a "high probability" ventilation-perfusion lung scan, an intraluminal filling defect in a segmental or more proximal pulmonary artery on CT pulmonary angiography, an intraluminal filling defect on pulmonary angiography, or diagnostic criteria for DVT in patients with suspected PE who had a non-high probability lung scan (20–22).

In the first-degree relatives, in the absence of previous objective testing, VTE was also diagnosed in the retrospective component of the study if the relative was treated for DVT or PE with anticoagulant therapy for more than two months.

Episodes of VTE in index cases and first-degree relatives were classified as "secondary" when the following risk factors were present: surgical intervention, bed rest for more than three days, plaster cast of a lower limb, all within three months of VTE (23); airtravel for more than two hours in the last two months (24); pregnancy in the past two months; current use of oral contraception or hormonal replacement therapy (23); or cancer that was active within the past six months (25). In the absence of one these risk factors, VTE was classified as "unprovoked".

Study personnel who were not aware if the first-degree relatives had FVL recorded all data in a standardized manner. In the retrospective component of the study, as there was no routine screening for FVL and prothrombin gene mutation among relatives in the Brest district, the high majority of first-degree relatives did not know if they had FVL when they were included. In the prospective component of the study, first-degree relatives had previously been informed if FVL was present or absent; those with FVL had been given written recommendations about appropriate VTE prophylaxis during periods of increased risk, and were cautioned about the risks of receiving oestrogen-containing oral contraceptive or hormonal replacement therapy.

Period of observation

The period of observation for first-degree relatives was from 16 years of age to the date of the first VTE or, in the absence of VTE, to the date of death or last contact (applicable to the prospective component of the study). For each relative, the observation period was divided into a retrospective component and a prospective component that started at enrolment in the study. First-degree relatives who were too young to be eligible when initially assessed for the study were not added to the study cohort when they subsequently reached 16 years of age.

Genetic testing for FVL and the prothrombin gene mutation

Blood from index cases and first-degree relatives was collected in 0.05 M EDTA for DNA analysis. DNA was extracted according to standard procedures and analysed for FVL and the prothrombin G20210A mutation as previously described (13); results were classified as normal, heterozygous, or homozygous, for each mutation.

Statistical analysis

The annual incidence of VTE in all first-degree relatives, and in subgroups, was calculated by dividing the number of first episodes of VTE that occurred in that group by the total number of years of observation in that group (i.e. probability, expressed as a percentage, of VTE per subject per year). Age of relatives during follow-up was divided into the following categories: 16 to 29 years, 30 to 44 years, 45 to 59 years, 60 to 74 years, and 75 years and older. 95% confidence intervals (CI) were calculated according to the normal approximation of the binomial distribution or using Cox proportional-hazards modelling. Statistical analyses were performed using SPSS software, version 12.0 (SPSS, Inc., Chicago, IL, USA). Relative risk of a first VTE in association with FVL was calculated by dividing the incidence of VTE in relatives with FVL by the incidence of VTE in those without FVL. Relative risk of VTE in association with FVL, adjusted for demographic variables (e.g. age, gender), were calculated using the Mantel-Haenszel test. VTE-free survival in relatives with and without FVL was analysed according to the Kaplan-Meier method, and curves were compared using the Log-Rank test. The influence of a number of predefined variables on the risk of VTE in relatives was determined: firstly by univariate analysis as a relative risk and, secondly, by multivariate analysis as a hazard ratio using Cox proportional-hazards modelling. An analysis for interaction between FVL and age was pre-planned; for this analysis, age of relatives was categorized as less than 70 years, or 70 years or older, based on the findings of a previous study by our group in an unrelated population (26).

Results

Between September 1993 and September 2000, 161 patients were eligible as index cases and consented to participate. The mean (± SD) age of index cases was 51.3 (± 18.7) years. VTE was a first episode in 98 patients and a recurrent episode in 63 cases. Twelve of the index cases (7.5%) were homozygous for FVL and 11 were heterozygous for both the prothrombin G20210A and FVL mutation. The 161 index cases had a total of

1,002 first-degree relatives: 449 of these were excluded as the presence or the absence of FVL could not be determined (269 relatives were dead and 180 relatives were unable or unwilling to participate). Characteristics of the 553 first-degree relatives who were enrolled in the study are shown in Table 1.

Incidence of VTE according to FVL, gender and status of relatives

During a total of 17,532 years of observation, 57 first-degree relatives developed a first episode of VTE, corresponding to an annual incidence of 0.32% (95%CI, 0.24 to 0.41) (Table 1). The annual incidence was 0.43% (95% CI, 0.07 to 0.27) among those

Table 1: Incidence of venous thromboembolism in first-degree relatives according to presence of factor V Leiden, gender and the status of the relatives.

	Absence of factor V Leiden	Presence of factor V Leiden
All family members, n	231	322
Age – years (± SD)	45.4 (± 17.3)	47.6 (± 18.2)
VTE events/ years of observation	12/7031	45/10,501
Incidence per person per year, [95%CI]	0.17 % [0.07 – 0.27]	0.43 % [0.3 – 0.56]
Relative risk, [95%CI]	2.5 [1.3 – 4.7]	
Men, n	94	133
Age – years (± SD)	46.0 (± 17.7)	47.8 (± 18.9)
VTE events/ years of observation	4/2782	11/4377
Incidence per person per year, [95% CI]	0.14 % [0.00 – 0.28]	0.25 % [0.1 – 0.40]
Relative risk, [95%CI]	1.7 [0.6 – 5.5]	
Women, n	137	189
Age – years (± SD)	44.6 (± 16.7)	47.9 (± 17.1)
VTE events/years of observation	8/4249	34/6124
Incidence per person per year, [95% CI]	0.19 % [0.06 – 0.32]	0.56 % [0.37 – 0.74]
Relative risk, [95%CI]	2.9 [1.4 – 6.4]	
Parents, n	32	71
Age – years (± SD)	60.6 (± 8.6)	62.4 (± 16.5)
VTE events/ years of observation	1/1460	17/3363
Incidence per person per year, [95%CI]	0.07 % [0.00 – 0.38]	0.51 % [0.29 – 0.81]
Relative risk, [95%CI]	7.4 [1.0 – 55.4]	
Sibling, n	107	127
Age – years (± SD)	49.5 (± 17.9)	49.1 (± 17.3)
VTE events/ years of observation	8/3695	15/4334
Incidence per person per year, [95% CI]	0.22 % [0.09 – 0.43]	0.35 % [0.13 – 0.57]
Relative risk, [95%CI]	1.6 [0.7 – 3.8]	
Children, n	92	124
Age – years (± SD)	35.4 (± 12.4)	37.6 (± 13.1)
Venous thromboembolism events/years of observation	3/1876	13/2804
Incidence per person per year, [95% CI]	0.16 % [0.03 – 0.47]	0.56 % [0.25 – 0.79]
Relative risk, [95%CI]	2.9 [0.8 – 10.2]	
Thrombophilia		
– No factor V Leiden, no PGM – n, (VTE events)	225 (12*)	-
– No factor V Leiden, heterozygous PGM – n, (VTE events)	6 (0†)	-
– Heterozygous factor V Leiden, no PGM – n (VTE events)	-	298 (35‡)
– Homozygous factor V Leiden, no PGM – n (VTE events)	-	14 (7§)
– Heterozygous factor V Leiden, heterozygous PGM – n (VTE events)	-	9 (2||)
– Homozygous factor V Leiden, heterozygous PGM- n (VTE events)	-	1 (1**)

PGM: prothrombin gene mutation; VTE: venous thromboembolism; * 0.17%/year [0.08 to 0.27]; † 0.0%/year [0.0 to 0.39]; ‡ 0.36%/year [0.24 to 0.49]; § 2.3%/year [0.6 to 3.9]; || 0.53%/year [0.0 to 1.26]; ** 2.33%/year; [0.0 to 12.3].

with FVL and 0.17% (95% CI, 0.07 to 0.27) among those without FVL; the corresponding relative risk of VTE with FVL was 2.5 (95% CI, 1.3 to 4.7) (Table 1). The annual incidence was 0.36% (95% CI, 0.24 to 0.49) for relatives with heterozygous FVL alone (relative risk of 2.1 [95% CI, 1.1 to 4.0]), 0.53% (95%CI, 0.0 to 1.26) for relatives who were compound heterozygotes for the FVL and prothrombin mutations (relative risk of 3.02 [95% CI, 0.68 – 13.46]) compared with no FVL) and 2.3% (95% CI, 0.6 to 3.9) for relatives with homozygous FVL (relative risk of 13.0 [95% CI, 5.2 to 32.8]) compared with no FVL (Table 1).

The annual incidence of VTE was higher in all women (0.40%; 95% CI, 0.28 to 0.53) than in men (0.21%; 95% CI, 0.10 to 0.32). The relative risk of VTE with FVL was similar in men and women (Table 1). The risks of VTE with FVL, and without FVL, in parents, siblings and children are reported in Table 1. The overall (both with and without FVL) annual incidence of VTE was similar in parents (0.37 [0.22 to 0.59]), siblings (0.29 [0.18 to 0.43]) and children (0.34 [0.20 to 0.55]). Finally, when adjusted on age, sex and status of relatives, the relative risk of VTE with FVL was 2.4 [1.26 to 4.58].

Incidence of VTE according to age and FVL

In both relatives with and without FVL, the annual incidence of a first episode of VTE did not increase until 60 years of age, and then increased markedly in the older age group (Fig. 1). The relative risk of VTE with FVL appeared to be greater when relatives were less than 60 years of age compared to when they were older (Fig. 1); however, when we tested our predefined hypothesis that the relative risk would be higher when relatives were less than 70 years of age (2.65; 95% CI, 1.33 to 5.31) compared to when they were older (1.11; 95% CI, 0.23 to 5.43), this difference was not statistically significant (p=0.36 for interaction test).

Provoking factors for VTE in first-degree relatives

Of the 57 episodes of VTE in first-degree relatives, 40 (70%; 95% CI, 57 to 82) were associated with an acquired provoking factor and 17 (30%; 95% CI, 18 to 43) were unprovoked. Of the 45 episodes of VTE in relatives with FVL, 33 (73%; 95% CI, 60 to 86) were provoked (surgery, trauma, or immobilization: 11 episodes; pregnancy or post-partum: 16 episodes; oestrogen-containing oral contraceptives or hormonal replacement therapy: 5 episodes; cancer: 1 episode) and of the 12 episodes in relatives without FVL, seven (58%; 95% CI, 28 to 84), were provoked (surgery, trauma, or immobilization: 2 episodes; pregnancy or post-partum: 4 episodes; oral contraceptives or hormonal replacement therapy: 0 episodes; cancer: 1 episode).

The proportion of VTE that was provoked differed between women and men; 35 of 42 (83%; 95% CI, 72 to 94) episodes of VTE were provoked in women and five of 15 (33%; 95% CI, 9 to 57) episodes of VTE were provoked in men. In women, 25 (60%) episodes of VTE were provoked by pregnancy or were post-partum (20 episodes) or were associated with oestrogen-containing oral contraceptive or hormonal replacement therapy (5 episodes). After 60 years of age, the proportion of episodes of VTE that was provoked was similar in women (2/6; 33% [95% CI, 4 to 77]) and in men (3/8; 38% [95% CI, 9 to 76]).

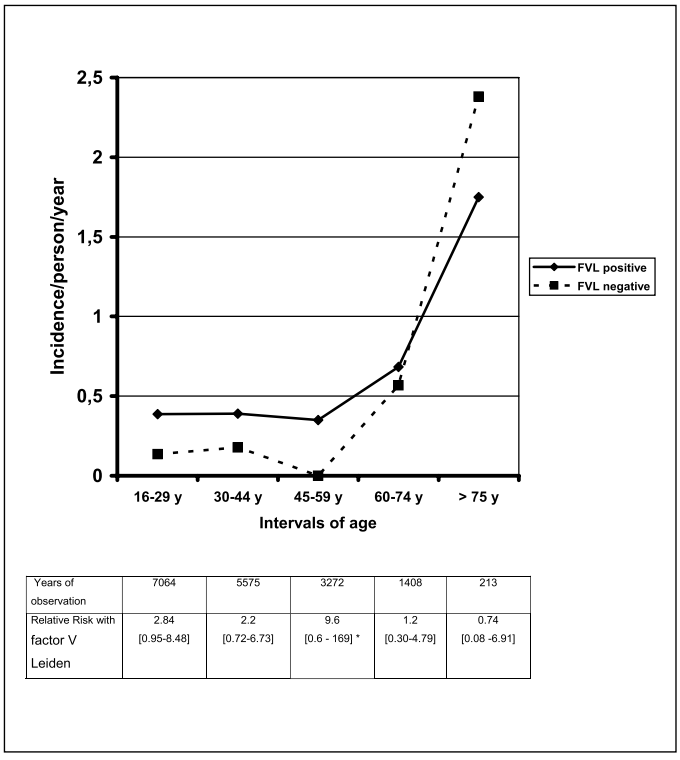
There were 407 pregnancies among relatives with FVL of which 16 were complicated by VTE (3.9%; 95%CI, 2.0 to 5.8) (14/16 with heterozygous FVL; 0/16 with homozygous FVL and 2/16 with heterozygous FVL and heterozygous prothrombin gene mutation), and 290 pregnancies among relatives without FVL of which four were complicated by VTE (1.4%; 95%CI, 0.04 to 2.7). The relative risk of VTE in association with FVL during pregnancy or the post-partum period was 2.9 (95% CI, 0.96 to 8.4, p= 0.06).

Characteristics of index cases as predictors of VTE in first-degree relatives

There was no evidence that the following clinical characteristics of the index cases influenced the risk of VTE in the first-degree relatives: female versus male (relative risk of 1.3; 95%CI, 0.8 to 2.3); age older versus younger than 60 years when the VTE was diagnosed (relative risk of 0.6; 95%CI, 0.3 to 1.2); unprovoked versus a provoked episode of VTE (relative risk of 0.9; 95%CI, 0.5 to 1.5); PE versus DVT (relative risk of 0.9; 95%CI, 0.5 to 1.5).

Factors influencing presentation of VTE as PE or DVT

Of the 57 episodes of VTE in first-degree relatives, 46 episodes were DVT (81%; 95% CI, 68 to 90) and 11 were PE with or without DVT (19%; 95% CI, 9 to 35). The proportion of episodes of VTE that presented as PE was similar among relatives with FVL (9/45; 20% [95% CI, 10 to 35]) and relatives without FVL (2/12; 17% [95% CI, 2 to 48]). Similarly, we found no convincing cor-



Years of observation	7064	5575	3272	1408	213
Relative Risk with factor V Leiden	2.84 [0.95-8.48]	2.2 [0.72-6.73]	9.6 [0.6 - 169] *	1.2 [0.30-4.79]	0.74 [0.08 -6.91]

Figure 1: Incidence of venous thromboembolism according to factor V Leiden and intervals of age. *Estimate with 0.5 recurrences added to the seven events in relatives with factor V Leiden and the zero events in those without factor V Leiden.

relation between the clinical presentation of VTE (i.e. PE or DVT) in the index cases and the clinical presentation in their relatives; in relatives of index cases with DVT, five of 32 episodes (16%; 95% CI, 5 to 33) presented as PE, whereas in relatives of index cases with PE, six of 25 episodes (24%; 95% CI, 9 to 45) presented as PE.

Incidence of VTE during the retrospective and prospective components of the study

Forty-five episodes of VTE occurred during 15,533 years of retrospective observation for an annual incidence of 0.29% (95%CI, 0.21 to 0.37), and 12 episodes of VTE occurred during 1,999 years of prospective observation for an annual incidence of 0.60% (95%CI, 0.26 to 0.90). Relatives were exposed to risk of VTE for 28.1 (± 17.7) years during the retrospective component (follow-up started at 16 years of age) and for 4.3 (± 2.2) years during the prospective component (follow-up started at enrolment).

Among first-degree relatives who had FVL, 37 episodes of VTE occurred during 9,401 years of retrospective observation for an incidence of 0.39% (95% CI, 0.26 to 0.52), and eight episodes of VTE occurred during 1,100 years of prospective observation for an incidence of 0.73% (95% CI, 0.23 to 1.23); 29 of 37 (78%; 95% CI 62 to 90) episodes of VTE were provoked during the retrospective component, and four of eight (50%; 95% CI, 16 to 84) episodes were provoked during the prospective component.

Finally, the hazard ratio for VTE in association with FVL was 2.9 (95% CI, 1.3 to 6.2) in the retrospective component and 1.7 (95% CI, 0.5 to 5.6) in the prospective component (after adjustment for age during follow-up).

Discussion

This study showed that heterozygous FVL was associated with a two- to three-fold, and homozygous FVL a 10- to 15-fold, increase in VTE in first-degree relatives of patients who have VTE and FVL. Two-thirds of episodes of VTE in the first-degree relatives were associated with an acquired risk factor. Among women, estrogen therapy and pregnancy accounted for more than half of such risk factors, and our findings support that the increase in VTE in association with these acquired risk factors and FVL is multiplicative. We failed to identify clinical situations or subgroups of patients with such a high risk of VTE that more aggressive prophylaxis than is usually recommended in the general population would be clearly indicated.

The absolute and relative risks of VTE associated with FVL in this study are consistent with the findings of previous studies of a similar design (14–18) (Table 2). Taken together, these studies suggest that relatives with FVL have a three- to four-fold higher risk of VTE than relatives without the mutation, and that they have an annual risk of thrombosis of about 0.4%. In the current study, VTE complicated about 4% of pregnancies in women with FVL, which is about twice as frequently as that reported by Middeldorp (14) and Simioni (15). Women without FVL also had a higher frequency of pregnancy-associated VTE in our study.

Consistent with previous studies (26–30), there was a marked increase in the risk of VTE with advancing age in both relatives with, and without, FVL. There was some evidence that the increase in VTE with age was less marked among relatives with FVL; however, this difference was not statistically significant.

This study has a number of potential limitations. First, most episodes of VTE occurred during the retrospective component of

Table 2: Relative risk of venous thromboembolism in association with factor V Leiden in first degree relatives of symptomatic probands with factor V Leiden.

	Middeldorp 1998*	Simioni 1999*	Lensen 2000*	Middeldorp 2000†	Simioni 2002†	Couturaud 2006‡	Combined
With factor V Leiden							
Observation period	6524	6114	4711	1564	1255	10501	30,669
Number of VTE	29	17	16	9	8	45	124
Incidence per person per year [95% CI]	0.44% [0.28–0.61]	0.28% [0.15–0.41]	0.34% [0.17–0.51]	0.58% [0.20–1.1]	0.64% [0.28–0.95]	0.43% [0.30–0.55]	0.40% [0.33–0.48]§
Without factor V Leiden							
Observation period	5716	4401	4049		984	7031	22,181
Number of VTE	5	4	5		1	12	27
Incidence per person per year [95% CI]	0.09% [0.01–0.16]	0.09% [0.0–0.18]	0.12% [0.02–0.23]		0.10% [0.0–0.57]	0.17% [0.07–0.27]	0.12% [0.08–0.17]§
Relative risk	4.2 [1.8–9.9]	2.8 [1.1–8.6]	2.9 [1.1–7.9]		6.6 [1.1–39.8]	2.5 [1.3–4.7]	3.2 [2.1–4.9]¶

* Retrospective studies. †Prospective studies. ‡ Retrospective for 90% of observation period. § Pooled estimates for annual incidences were obtained by directly combining data from the individual studies, and 95% confidence intervals for these estimates were calculated using the binomial distribution. ¶ A pooled estimate for the relative risk, with associated 95% confidence interval, was obtained by combining the rate ratios from the individual studies using a fixed effect model with the Mantel-Haenszel method ($P = 0.74$ for heterogeneity).

the study, and accuracy of diagnosis may have been lower during that phase of the study. Consistent with studies by Middeldorp (14, 17) and Simioni (15, 18), there was a trend to a lower frequency of VTE in the retrospective component compared with the prospective phase. Second, the frequency of VTE associated with an acquired provoking factor may have been reduced in the relatives with FVL in the prospective component of the study, because they were encouraged to use preventative measures during periods of high risk. Third, because we did not have information about the use of VTE prophylaxis during either phase of the study, we are unable to determine if the episodes of VTE that occurred after major risk factors, such as surgery, might have been prevented by use of routinely recommended methods of prophylaxis. Lastly, we were unable to assess the influence of other hereditary thrombophilias (e.g. deficiencies of protein C, protein S, antithrombin) on the incidence of VTE, and to assess the interaction of these abnormalities with FVL.

Strengths of the study include that assessment of previous VTE in the retrospective component of the study was performed without knowledge of FVL status. Consequently, the estimated relative risk of VTE in association with FVL during that phase of the study should be unbiased. A second strength is that, with over 17,000 person-years of observation and 57 episodes of VTE, this study is the largest of its kind, which increases the precision of its findings (Table 2).

In conclusion, the presence of FVL was shown to be associated with a two- to three-fold increased risk of VTE in first-degree relatives of patients with venous thrombosis and FVL. No clinical situations, or subgroups of patients with FVL, were identified that would clearly justify a more aggressive approach to prophylaxis than is currently recommended for the general population.

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