

HIGH RISK OF CEREBRAL-VEIN THROMBOSIS IN CARRIERS OF A PROTHROMBIN-GENE MUTATION AND IN USERS OF ORAL CONTRACEPTIVES

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ABSTRACT

Background Idiopathic cerebral-vein thrombosis can cause serious neurologic disability. We evaluated risk factors for this disorder, including genetic risk factors (mutations in the genes encoding factor V and prothrombin) and nongenetic risk factors (such as the use of oral contraceptive agents).

Methods We compared the prevalence of these risk factors in 40 patients with cerebral-vein thrombosis, 80 patients with deep-vein thrombosis of the lower extremities, and 120 healthy controls. The G1691A mutation in the factor V gene and the G20210A prothrombin-gene mutation, which are established genetic risk factors for venous thrombosis, were studied. We also assessed the use of oral contraceptives and other risk factors for thrombosis.

Results The prevalence of the prothrombin-gene mutation was higher in patients with cerebral-vein thrombosis (20 percent) than in healthy controls (3 percent; odds ratio, 10.2; 95 percent confidence interval, 2.3 to 31.0) and was similar to that in patients with deep-vein thrombosis (18 percent). Similar results were obtained for the mutation in the factor V gene. The use of oral contraceptives was more frequent among women with cerebral-vein thrombosis (96 percent) than among controls (32 percent; odds ratio, 22.1; 95 percent confidence interval, 5.9 to 84.2) and among those with deep-vein thrombosis (61 percent; odds ratio, 4.4; 95 percent confidence interval, 1.1 to 17.8). For women who were taking oral contraceptives and who also had the prothrombin-gene mutation (seven patients with cerebral-vein thrombosis but only one control), the odds ratio for cerebral-vein thrombosis rose to 149.3 (95 percent confidence interval, 31.0 to 711.0).

Conclusions Mutations in the prothrombin gene and the factor V gene are associated with cerebral-vein thrombosis. The use of oral contraceptives is also strongly and independently associated with the disorder. The presence of both the prothrombin-gene mutation and oral-contraceptive use raises the risk of cerebral-vein thrombosis further. (N Engl J Med 1998;338:1793-7.)

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THE risk of venous thrombosis of the lower extremities is increased by factors that cause hypercoagulability or venous stasis, such as the use of oral contraceptives, pregnancy or the postpartum state, surgery, trauma, and prolonged immobilization. The risk of venous thrombosis is also increased by hypercoagulable states due to inherited abnormalities of the coagulation system, such as the G1691A mutation in the

factor V gene, which causes resistance to activated protein C, and deficiencies of antithrombin, protein C, or protein S. Acquired abnormalities such as the presence of antiphospholipid antibodies are also associated with an increased risk of venous thrombosis.¹ The recent discovery of a transition from guanine to adenine at position 20210 in the sequence of the 3' untranslated region of the prothrombin gene has widened the spectrum of inherited thrombophilia.² Next to the mutation in the factor V gene,^{3,4} the prothrombin-gene mutation is the most common genetic determinant of deep-vein thrombosis of the lower extremities.²

Cerebral-vein thrombosis is a frightening event because of the severity of the clinical manifestations and the high mortality rate, estimated to be 5 to 30 percent.⁵⁻⁷ Clinically, cerebral-vein thrombosis presents with a wide range of symptoms, including headache, focal deficits (motor or sensory), dysphasia, seizures, and impaired consciousness. Idiopathic cerebral-vein thrombosis (i.e., that occurring in the absence of infection, trauma, tumors, or autoimmune disease) represents a large proportion of cases (approximately 30 percent).⁶ Although it is known that the mutation of the factor V gene,⁸⁻¹³ the use of oral contraceptives, and pregnancy^{6,8,10,11} are risk factors for cerebral-vein thrombosis, no information about the relation between the disorder and the prothrombin-gene mutation is yet available. The main objective of this study was to evaluate this risk factor and potential interactions with other risk factors for cerebral-vein thrombosis.

METHODS**Patients with Cerebral-Vein Thrombosis**

Forty unrelated patients with a first episode of idiopathic cerebral-vein thrombosis were studied retrospectively (9 men and 31 women; median age, 31 years; range, 15 to 64). They were referred to our thrombosis center between April 1991 and May 1997 for screening for abnormalities of the coagulation system. None of them had overt evidence of autoimmune or neoplastic disease. Two additional patients were excluded from the study because cerebral-vein thrombosis was secondary to infection (acute otitis) or to a post-traumatic cerebral arteriovenous fistula. The clinical records and the objective documentation of cerebral-vein

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thrombosis were reviewed by a neurologist to confirm the diagnosis. The diagnosis was made by intraarterial angiography in 20 cases, intravenous digital angiographic imaging in 2 cases, magnetic resonance imaging in 16 cases, and computed tomography showing the typical delta and cord signs in 2 cases.¹⁴

Twelve patients had symptoms of chronic isolated intracranial hypertension (headache and papilledema in all 12 and a sixth-nerve palsy in 2). The remaining 28 patients had various combinations of focal neurologic deficits, seizures, and impaired consciousness. Nineteen patients had radiologic evidence of thrombosis in a single dural sinus, and 21 had evidence of thromboses in two or more sinuses. Six patients had concomitant cortical-vein thromboses (five with a single dural-sinus thrombosis and one with multiple sinus thromboses).

Patients with Deep-Vein Thrombosis of the Lower Extremities

Four hundred eighty-nine patients with first documented episodes of proximal deep-vein thrombosis of the lower extremities were referred to our thrombosis center for the same screening for abnormalities of the coagulation system during the same period as the patients with cerebral-vein thrombosis. We randomly selected 80 of these patients (18 men and 62 women; median age, 30 years; range, 13 to 62) who were matched to the patients with cerebral-vein thrombosis according to sex, age (± 5 years), geographic origin, and level of education (less than high school, high school, or college). They had no evidence of cancer or autoimmune disease.

Healthy Controls

One hundred twenty healthy persons (27 men and 93 women; median age, 32 years; range, 18 to 64) were matched to the patients with cerebral-vein thrombosis according to sex, age, geographic origin, and level of education. These controls came from a population of biologically unrelated friends or partners of the patients who agreed to accompany the patients to the thrombosis center and to participate in the study. Persons who had had thrombosis were excluded with use of a structured questionnaire validated for the retrospective diagnosis of thrombosis.^{15,16}

The presence of known risk factors for thrombosis, such as the use of oral contraceptives, pregnancy or the postpartum state (the three months after childbirth), surgery, trauma, and prolonged immobilization, was recorded. The period of oral-contraceptive use was recorded. Women were considered to be using oral contraceptives if they had taken them during the two weeks preceding the episode of thrombosis. Since smoking may increase the risk of thrombosis in carriers of genetic coagulation defects,^{17,18} information on smoking was also recorded. Persons who smoked at least five cigarettes daily were considered smokers.

Laboratory Tests

Blood was collected in vacuum tubes containing 3.8 percent (wt/vol) sodium citrate as an anticoagulant. DNA analysis to identify a transition from guanine to adenine at position 20210 in the 3' untranslated region of the prothrombin gene was carried out according to the method of Poort et al.¹⁹ DNA analysis to identify a transition from guanine to adenine at position 1691 in the coagulation factor V gene was carried out as described by de Ronde and Bertina.²⁰ Heparin cofactor activity, an indicator of the antithrombin level, was measured by an amidolytic assay (Coamate AT, Chromogenix, Mölndal, Sweden). When antithrombin levels were low, the defect was further characterized by measuring antigen levels by immunoelectrophoresis with a polyclonal antibody (Stago, Asnieres, France), measuring functional activity in the absence of heparin, and performing crossed immunoelectrophoresis with and without heparin.²¹

Protein C activity was assayed by a clotting assay (ProClot, Instrumentation Laboratory, Milan, Italy). When plasma levels of protein C activity were low, antigen levels were also measured by

enzyme-linked immunosorbent assay (ELISA) with polyclonal antibodies (Dako, Glostrup, Denmark). Total protein S antigen was measured by ELISA with polyclonal antibodies (Dako). Free protein S was measured in the same way after precipitation of the C4b-binding protein-protein S complex with polyethylene glycol 6000 (final concentration, 3.5 percent)²² or directly by ELISA with a commercial kit that uses a specific monoclonal antibody (Asserachrom Free Protein S, Stago). The diagnosis of antiphospholipid-antibody syndrome was made when lupus anticoagulant, anticardiolipin antibodies, or both were present. Protein C and protein S levels and lupus anticoagulant could not be evaluated in one patient with cerebral-vein thrombosis and in seven patients with deep-vein thrombosis who were receiving oral anticoagulant therapy at the time of blood sampling, since measurements of vitamin K-dependent proteins are unreliable during such therapy.

Statistical Analysis

We compared patients with cerebral-vein thrombosis with those with deep-vein thrombosis with respect to the listed risk factors. Because patients with deep-vein thrombosis were matched with patients with cerebral-vein thrombosis, they could not be directly compared with healthy controls. The estimated risks for patients with cerebral-vein thrombosis are given as odds ratios and 95 percent confidence intervals, with the approximation of Woolf.²³ Because using oral contraceptives is incompatible with being pregnant or in the postpartum state, the proportion of women taking oral contraceptives was calculated after excluding pregnant or postpartum women, and the proportion of pregnant women was calculated after excluding women taking oral contraceptives. These proportions were also calculated after the exclusion of postmenopausal women (two with cerebral-vein thrombosis, three with deep-vein thrombosis, and six controls). Adjustment for matching factors and potential confounding factors was performed by unconditional logistic-regression analysis with the SAS software package.²⁴

RESULTS

Coagulation Abnormalities

The main characteristics of the patients and the prevalence of coagulation defects are listed in Table 1. Twenty percent of the patients with cerebral-vein thrombosis (7 women and 1 man), 18 percent of those with deep-vein thrombosis (11 women and 3 men), and 3 percent of the controls (2 women and 1 man) were carriers of the prothrombin-gene mutation. The prevalence of the defect was significantly higher in patients with cerebral-vein thrombosis than in controls (odds ratio, 10.2; 95 percent confidence interval, 2.3 to 31.0) and was similar to that found in patients with deep-vein thrombosis (18 percent) (Table 2). Similarly, the prevalence of the factor V mutation was significantly higher in patients with cerebral-vein thrombosis (15 percent) than in controls (3 percent; odds ratio, 7.8; 95 percent confidence interval, 1.8 to 34.1) (Table 2). In one patient with cerebral-vein thrombosis and in two with deep-vein thrombosis, both the prothrombin-gene mutation and the factor V mutation were present.

The risk of cerebral-vein thrombosis associated with the prothrombin-gene mutation was independent of the presence of the factor V mutation, since the exclusion of carriers of both mutations did not appreciably change the results (odds ratio, 10.0; 95

TABLE 1. CHARACTERISTICS OF THE PATIENTS AND CONTROLS.

CHARACTERISTIC	PATIENTS WITH CEREBRAL-VEIN THROMBOSIS (N=40)	PATIENTS WITH DEEP-VEIN THROMBOSIS OF THE LOWER EXTREMITIES (N=80)	HEALTHY CONTROLS (N=120)
Sex — M/F	9/31	18/62	27/93
Median age (range) at the time of thrombosis — yr	31 (15–64)	30 (13–62)	—
Men	32 (15–64)	30 (13–62)	—
Women	30 (19–54)	30 (15–53)	—
Median age (range) at the time of blood sampling — yr	34 (17–67)	36 (16–72)	32 (18–64)
Men	34 (17–67)	48 (20–72)	32 (18–64)
Women	34 (22–57)	35 (16–58)	32 (20–62)
Subjects with coagulation defects — no. (%)			
G20210A prothrombin-gene mutation	8 (20)	14 (18)	3 (3)
G1691A factor V mutation	6 (15)	15 (19)	3 (3)
Other defects*	1 (3)	13 (16)	4 (3)
Subjects with nongenetic risk factors for thrombosis — no. (%)			
Surgery, trauma, or immobilization	1 (3)	14 (18)	0
Pregnancy or postpartum state†	4 (80)	10 (34)	0
Oral-contraceptive use‡	24 (96)	30 (61)	28 (32)

*Other defects are a deficiency of antithrombin, protein C, or protein S and the presence of antiphospholipid antibodies.

†Women taking oral contraceptives and postmenopausal women were excluded.

‡Pregnant, postpartum, and postmenopausal women were excluded.

percent confidence interval, 2.4 to 42.1). All carriers of the prothrombin-gene mutation were heterozygous, whereas four patients with deep-vein thrombosis were homozygous for the factor V mutation. The results did not substantially change after exclusion of the four homozygotes from the analysis.

The prevalence of other causes of thrombophilia (deficiency of antithrombin, protein C, or protein S or the presence of antiphospholipid antibodies) was 3 percent both in patients with cerebral-vein thrombosis and in controls, lower than the prevalence in patients with deep-vein thrombosis (16 percent). One patient with cerebral-vein thrombosis had both the mutation in the factor V gene and antithrombin deficiency. Of the patients with deep-vein thrombosis, two (2 percent) had antithrombin deficiency, three (4 percent) had protein C deficiency, one (1 percent) had protein S deficiency, and seven (9 percent) had antiphospholipid antibodies. Four controls (3 percent) had coagulation defects other than prothrombin and factor V mutations (three had antithrombin deficiency, and one had protein S deficiency).

Other Risk Factors for Thrombosis

The prevalence of other risk factors for thrombosis is shown in Table 1. Apart from the use of oral contraceptives, none of the controls had had any of the risk factors for thrombosis in the month before

TABLE 2. RISK OF CEREBRAL-VEIN THROMBOSIS IN THE PRESENCE OF GENETIC COAGULATION DEFECTS AND NONGENETIC RISK FACTORS.*

RISK FACTOR	ODDS RATIO (95% CONFIDENCE INTERVAL)
Genetic coagulation defects	
G20210A prothrombin-gene mutation	10.2 (2.3–31.0)
G1691A factor V mutation	7.8 (1.8–34.1)
Other risk factors for thrombosis	
Surgery, trauma, or immobilization	—
Pregnancy or postpartum state	—
Oral-contraceptive use†	22.1 (5.9–84.2)

*Patients with cerebral-vein thrombosis were compared with healthy controls.

†The odds ratio was calculated for women without other risk factors for thrombosis.

the visit. A smaller proportion of patients with cerebral-vein thrombosis than of patients with deep-vein thrombosis had had surgery, trauma, or prolonged immobilization (3 percent vs. 18 percent). Pregnancy or the postpartum state was frequently associated with both disorders (80 percent of patients with cerebral-vein thrombosis and 34 percent of those with deep-vein thrombosis). The most prevalent nongenetic risk factor for both disorders was

oral-contraceptive use. Eight patients with cerebral-vein thrombosis (20 percent), 20 with deep-vein thrombosis (25 percent), and 35 controls (29 percent) were smokers. Smoking was not associated with an increased risk of cerebral-vein thrombosis, either alone or in combination with the prothrombin-gene mutation or the factor V mutation (data not shown). Because of the high frequency of oral-contraceptive use among the female patients, we analyzed in detail the effect of oral contraceptives and their interaction with the mutations in the prothrombin and factor V genes in determining the risk of thrombosis.

Oral Contraceptives and Thrombosis

The proportion of oral-contraceptive users was higher among women with cerebral-vein thrombosis (96 percent) than among controls (32 percent; odds ratio, 22.1; 95 percent confidence interval, 5.9 to 84.2) (Table 2) and among patients with deep-vein thrombosis (61 percent; odds ratio, 4.4; 95 percent confidence interval, 1.1 to 17.8). The median period of oral-contraceptive use was 15 months for women with either cerebral-vein thrombosis or deep-vein thrombosis (range, 1 to 161 and 1 to 168, respectively) and 26 months for controls (range, 1 to 189).

The odds ratio for cerebral-vein thrombosis according to the combined presence of the prothrombin-gene mutation or the factor V mutation and the use of oral contraceptives at the time of thrombosis (or at the time of blood sampling, for controls) is shown in Table 3. Women using oral contraceptives who did not have the prothrombin-gene mutation had a higher risk of cerebral-vein thrombosis than controls (63 percent of the women with cerebral-vein thrombosis had these characteristics, as compared with 29 percent of the controls; odds ratio, 13.4; 95 percent confidence interval, 3.5 to 51.3). This odds ratio was similar to that calculated for oral-contraceptive users who did not have the factor V mutation (78 percent of the women with cerebral-vein thrombosis had these characteristics, as compared with 30 percent of the controls; odds ratio, 15.8; 95 percent confidence interval, 4.3 to 57.2). For women who were taking oral contraceptives and who also had the prothrombin-gene mutation (seven patients with cerebral-vein thrombosis but only one control), the odds ratio for cerebral-vein thrombosis increased to nearly 150. The combined effect of the use of oral contraceptives and the factor V mutation could not be evaluated because none of the controls had both risk factors.

DISCUSSION

This study shows that there is a hypercoagulable state in 35 percent of patients with idiopathic cerebral-vein thrombosis. The most frequent coagulation abnormality is the G20210A mutation in the

TABLE 3. RISK OF CEREBRAL-VEIN THROMBOSIS IN WOMEN ACCORDING TO WHETHER THEY HAD THE G20210A PROTHROMBIN-GENE MUTATION OR THE G1691A FACTOR V MUTATION AND USED ORAL CONTRACEPTIVES.*

COMBINATION†	PATIENTS WITH CEREBRAL-VEIN THROMBOSIS (N=27)	HEALTHY CONTROLS (N=93)	ODDS RATIO (95% CONFIDENCE INTERVAL)
	no. (%)		
No G20210A, no OC	3 (11)	64 (69)	1 (reference group)
No G20210A, OC	17 (63)	27 (29)	13.4 (3.5–51.3)
G20210A, no OC	0	1 (1)	—
G20210A, OC	7 (26)	1 (1)	149.3 (31.0–711.0)
No G1691A, no OC	3 (11)	63 (68)	1 (reference group)
No G1691A, OC	21 (78)	28 (30)	15.8 (4.3–57.2)
G1691A, no OC	0	1 (1)	—
G1691A, OC	3 (11)	0	—

*Four women who had cerebral-vein thrombosis during pregnancy or post partum were excluded from the analysis.

†OC denotes oral-contraceptive use.

prothrombin gene, which increases the risk of the disorder by a factor of 10. This relation was not affected by the concomitant presence of the G1691A mutation in the factor V gene, which is known to increase the risk of cerebral-vein thrombosis.⁸⁻¹³ This study also indicates that oral contraceptives are strongly associated with cerebral-vein thrombosis, increasing the risk by a factor of approximately 20. Although the number of women with both risk factors was small, the combined effect of the prothrombin-gene mutation and the use of oral contraceptives greatly increased the risk of cerebral-vein thrombosis. Since both the prothrombin-gene mutation and the use of oral contraceptives cause hypercoagulability,^{2,25} their combination probably enhances their individual effects on coagulation. In particular, both increase plasma levels of prothrombin,^{2,25} the zymogen responsible for thrombin formation in the coagulation system.

It has previously been observed that cerebral-vein thrombosis is associated with the factor V mutation and occurs more frequently in young women who are taking oral contraceptives or who are pregnant or in the postpartum state.^{6,8,10} In a recent small study, the relative risk associated with the use of oral contraceptives was estimated to be 4.2.²⁶ In the current study of twice as many women, there was an association between oral-contraceptive use and cerebral-vein thrombosis, but the magnitude of the risk, especially when oral-contraceptive use and the prothrombin-gene mutation were both present, was unexpected.

This finding raises two questions. One question is whether screening for the prothrombin-gene muta-

tion in young women before they are prescribed oral contraceptives would be useful. The other question is whether withholding oral contraceptives from carriers of the prothrombin-gene mutation would be worthwhile. Indiscriminate screening for the prothrombin-gene mutation would probably not be useful, since cerebral-vein thrombosis is a rare condition. The incidence of cerebral-vein thrombosis is not precisely known, but it is likely to be lower than the incidence of approximately 1 per 1000 persons per year reported for deep-vein thrombosis.²⁷ Vandenbroucke et al.^{28,29} suggested that indiscriminate screening would not be cost effective, even though the risk of deep-vein thrombosis in oral-contraceptive users who have the factor V mutation is greater than the risk in those with only one of these risk factors. Likewise, since the prevalence of the prothrombin-gene mutation is similar to or lower than that of the factor V mutation in the general population, one can reasonably draw the same conclusion for cerebral-vein thrombosis, even though its mortality rate is higher than that of deep-vein thrombosis.

With respect to the second question, withholding the most effective mode of contraception might lead to more pregnancies, which would also increase the risk of venous thromboembolism. Therefore, we recommend that carriers of the prothrombin-gene mutation who have had an episode of thrombosis discontinue taking oral contraceptives. For asymptomatic carriers, who are usually identified in family studies, counseling about alternative methods of contraception should be considered, taking into account whether other risk factors for thrombosis are present.

Another unresolved issue is the indication for anticoagulant treatment in patients with cerebral-vein thrombosis.^{5,6,30} Because of the design of our study, no definite opinion can be given about the efficacy and safety of this treatment. In the absence of objective evidence of cerebral hemorrhage, we give oral anticoagulant therapy for at least three months after the occurrence of cerebral-vein thrombosis to patients without coagulation defects, for up to one year to those with a coagulation defect, and for life to those who have had more than one thrombotic episode.

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