

POLYMORPHISMS IN THE FACTOR VII GENE AND THE RISK OF MYOCARDIAL INFARCTION IN PATIENTS WITH CORONARY ARTERY DISEASE

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ABSTRACT

Background High plasma levels of coagulation factor VII have been suggested to be predictors of death due to coronary artery disease. Since polymorphisms in the factor VII gene contribute to variations in factor VII levels, such polymorphisms may be associated with the risk of myocardial infarction, which is precipitated by thrombosis.

Methods We studied a total of 444 patients, 311 of whom had severe, angiographically documented coronary atherosclerosis. Of these 311 patients, 175 had documentation of a previous myocardial infarction. As a control group, 133 patients with normal coronary arteriograms were also included. We measured the levels of activated factor VII and assessed three polymorphisms in the factor VII gene, one involving the promoter (A1 and A2 alleles), one involving the catalytic region (R353Q), and one involving intron 7.

Results Each of the polymorphisms influenced factor VII levels. Patients with the A2A2 and QQ genotypes had the lowest levels of activated factor VII (66 percent and 72 percent lower, respectively, than the levels in patients with the wild-type genotypes). The frequencies of the various genotypes in the patients free of coronary artery disease were similar to those in the entire population of patients with coronary artery disease. In the latter group, there were significantly more heterozygotes and homozygotes for the A2 and Q alleles among those who had not had a myocardial infarction than among those who had had an infarction ($P=0.008$ for the presence of the promoter polymorphism and $P=0.01$ for the presence of the R353Q polymorphism by chi-square analysis). The adjusted odds ratio for myocardial infarction among the patients with the A1A2 or RQ genotype was 0.47 (95 percent confidence interval, 0.27 to 0.81).

Conclusions Our findings suggest that certain factor VII genotypes have a role in protection against myocardial infarction. This may explain why some patients do not have myocardial infarction despite the presence of severe coronary atherosclerosis. (N Engl J Med 2000;343:774-80.)

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THROMBOSIS underlies most acute manifestations of coronary atherosclerotic disease, including myocardial infarction.¹ Plaque disruption, with resulting exposure of tissue factor to blood and binding of tissue factor to circulating coagulation factor VII,^{2,3} is considered a major cause of thrombosis in myocardial infarction. During the past two decades, there has been substantial interest in investigating the role of factor VII levels in coronary disease. The Northwick Park Heart Study investigators reported that a high plasma level of factor VII was a predictor of death due to coronary disease.⁴ The trend was similar in some other studies,^{5,6} but not all.^{7,8} Plasma levels of factor VII are influenced by both environmental and genetic factors.⁹ Population studies have suggested that two common polymorphisms in the factor VII gene — the substitution of glutamine for arginine at position 353 in the catalytic domain (R353Q) and a 10-bp insertion in the promoter region (5'F7) — may be responsible for up to one third of the variation in factor VII levels.¹⁰ The factor VII gene is also characterized by a polymorphism involving a variable number of 37-bp repeats in intron 7 (IVS7).¹¹ The rare alleles of each polymorphism are generally associated with decreased levels of factor VII.

Interest in polymorphisms in the factor VII gene has recently been heightened by a case-control study¹² suggesting that the presence of certain alleles may significantly influence the risk of myocardial infarction. Other studies, however, failed to detect such an influence.^{13,14} Wang et al.¹⁵ found no association between the R353Q polymorphism and the angiographically documented severity of coronary atherosclerosis. On the other hand, it is biologically plausible that factor VII does not influence the development of coronary atherosclerosis, but only its thrombotic complication, myocardial infarction.

It is not uncommon in clinical practice to encounter patients who have angiographically documented severe coronary atherosclerosis but who have not had a myocardial infarction. Whereas there is much informa-

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tion on risk factors for myocardial infarction, the role of potentially protective factors has not been fully explored. We analyzed the prevalence of several polymorphisms in the factor VII gene in patients who had undergone coronary angiography. Our main objective was to determine whether there were different distributions of alleles among patients with severe coronary atherosclerosis according to whether they had a documented history of myocardial infarction. We also investigated whether there was an association between polymorphisms in the factor VII gene and the circulating levels of activated factor VII (factor VIIa).

METHODS

Study Population

Criteria for selection of the study population have been described in detail.¹⁶ In brief, we studied a total of 444 unrelated adult patients of both sexes who were recruited consecutively from those referred to the Institute of Cardiovascular Surgery of the University of Verona in Italy. Of these 444 patients, 311 had angiographically documented, severe multivessel coronary atherosclerosis and thus were candidates for coronary-artery bypass grafting. Classification into two groups according to the presence or absence of a history of myocardial infarction was done by combining data from the medical history with a thorough review of medical records for evidence of diagnostic electrocardiographic and enzyme changes or the typical sequelae of myocardial infarction on ventricular angiograms. Appropriate documentation was obtained for 285 of the 311 patients (91.6 percent); 175 were classified as having had a myocardial infarction, and 110 as not having had an infarction. The severity of coronary atherosclerosis was determined by the number of coronary arteries with stenosis of more than 50 percent of the luminal diameter. The angiograms were assessed by two cardiologists who were unaware that the patients were participating in the study. Most of the patients with coronary artery disease (76 percent) had severe disease involving all three major coronary arteries, 18 percent had two stenosed vessels, and 6 percent had one stenosed vessel.

As a control group, 133 patients who were examined for reasons other than possible coronary artery disease (in most cases valvular heart disease) were enrolled. They were required to have normal coronary arteries as documented by angiography and to have neither a history of atherosclerosis nor clinical or laboratory evidence of atherosclerosis in other vascular beds. This control group was included so that the atherosclerotic phenotype could be clearly defined and any association between polymorphisms in the factor VII gene and coronary atherosclerosis itself could be identified.

All the study participants came from northern Italy and had similar socioeconomic and ethnic backgrounds. At the time of blood sampling, a complete clinical history, including the presence or absence of cardiovascular risk factors such as smoking, hypertension, and diabetes mellitus, was obtained from all the patients. Patients who were taking an anticoagulant drug were excluded from the genotype-phenotype correlation studies. The study was approved by our institutional review boards. Either written or oral informed consent was obtained from all the patients.

Biochemical Analysis and Factor VIIa Assay

Samples of venous blood were taken from each patient after an overnight fast. For patients scheduled to undergo coronary-artery bypass grafting, these samples were obtained several days before surgery. Serum lipids and other predictors of the risk of cardiovascular events, including the level of homocysteine, were measured as previously described.¹⁶

Blood was drawn into vacuum tubes containing 0.1 part 0.129 M buffered sodium citrate per 10 parts blood. Factor VIIa was assayed in plasma with the use of soluble, recombinant, truncated tissue factor (Staclot VIIa-rTF, Diagnostica Stago, Asnières-sur-Seine,

France).¹⁷ The results were expressed in milliunits per milliliter, where 30 such units are equivalent to 1 ng of factor VIIa. The within-run coefficient of variation was 7.8 percent, and the between-run coefficient of variation was 6.4 percent.

Mutation Analysis and Nomenclature

DNA was extracted from peripheral-blood lymphocytes by the phenol-chloroform method. Analysis to detect the three polymorphisms in the factor VII gene was performed as previously described.¹⁸ The three polymorphisms were 5'F7, involving a decamer insertion at position -323 in the 5' promoter region, where allele *A1* corresponds to the absence of the decamer and allele *A2* to its insertion; R353Q, involving a substitution of adenine for guanine in the codon for residue 353 in exon 8, which results in the substitution of glutamine (Q) for arginine (R); and IVS7, involving a variable number of 37-bp tandem repeats (five to eight) in the hypervariable region 4 of intron 7, where allele *a* (also called *H7*) corresponds to the presence of seven monomers,¹² *b* (*H6*), the presence of six monomers, *c* (*H5*) the presence of five monomers, and *d* (*H8*) the presence of eight monomers.

Statistical Analysis

All calculations were performed with SPSS statistical software (version 7.5.21, SPSS, Chicago). Distributions of continuous variables in groups are expressed as means \pm SD. Logarithmic transformation was performed on all skewed variables, including the level of factor VIIa. Differences in quantitative variables were assessed for statistical significance with Student's unpaired *t*-test. Qualitative data were analyzed with the chi-square test. Factor VIIa levels were compared among individual patients with different polymorphisms by analysis of variance, with use of the Tukey procedure for post hoc multivariate comparison of the means. In each of the two groups of patients (those with coronary artery disease and those without it), the frequencies of the alleles and genotypes associated with each of the three polymorphisms were compared by chi-square analysis, with the values predicted on the basis of the Hardy-Weinberg equilibrium. Genotypes were analyzed with the number of possible values (three for the promoter and R353Q polymorphisms and six for the IVS7 polymorphism) taken into account. To assess the extent to which the various alleles and genotypes were associated with coronary atherosclerosis or myocardial infarction, odds ratios with 95 percent confidence intervals were estimated by univariate logistic-regression analysis. The effects of the *A2*, *Q*, and *a* alleles were weighted, with the wild-type alleles *A1*, *R*, and *b*, respectively, used as the references. To provide separate odds ratios for each genotype, two (or, in the case of the IVS7 polymorphism, more than two) dummy variables were used, with *A1A1*, *RR*, and *bb* used as the reference groups. Adjustment for the patients' sex and smoking status was performed by including these covariates in a second set of multivariate logistic-regression models.

RESULTS

The characteristics of the patients are summarized in Table 1. As expected, the patients with coronary artery disease had more conventional risk factors for cardiovascular events than did those free of coronary artery disease. In agreement with the results of a previous study,¹⁸ a strong linkage disequilibrium among the polymorphisms was found, particularly between 5'F7R and R353Q. All 12 of the patients with the *QQ* genotype also had the *A2A2* genotype; the remaining 6 patients with *A2A2* had the *RQ* genotype.

Genotype-phenotype correlation analysis was performed with data from the entire study population, after patients taking an anticoagulant drug at the time of blood sampling or during the few days before blood

TABLE 1. CHARACTERISTICS OF THE STUDY PATIENTS AND CONTROLS.*

CHARACTERISTIC	PATIENTS WITH CORONARY ARTERY DISEASE (N=311)	CONTROLS (N=133)	P VALUE	PATIENTS WITH CORONARY ARTERY DISEASE AND MYOCARDIAL INFARCTION† (N=175)	PATIENTS WITH CORONARY ARTERY DISEASE AND NO MYOCARDIAL INFARCTION† (N=110)	P VALUE
Age (yr)	61.3±8.8	57.5±13	0.002	60.3±8.7	61.6±8.4	0.36
Male sex (% of patients)	87.6	51.3	<0.001	89.5	77.0	0.007
Body-mass index‡	26.2±2.9	24.8±3.3	<0.001	26.4±3.1	26.3±3	0.80
Serum cholesterol (mmol/liter)						
Total	6.16±1.1	5.62±1.1	<0.001	6.2±1.1	6.0±1.2	0.16
HDL	1.25±0.3	1.49±0.4	<0.001	1.2±0.3	1.3±0.4	0.10
LDL	4.18±1.0	3.63±0.9	<0.001	4.3±0.9	4.0±1.1	0.10
Serum triglycerides (mmol/liter)	2.14±1.7	1.47±0.7	<0.001	2.1±1.8	2.2±1.7	0.66
Plasma fibrinogen (g/liter)	3.55±0.8	3.32±0.8	0.01	3.5±0.8	3.4±0.6	0.32
Plasma homocysteine (μmol/liter)	15.7±1.4	13.8±1.4	0.02	15.6±1.5	15.7±1.5	0.80
Smoking (% of patients)	71.0	44.0	<0.001	74.7	62.6	0.04
Hypertension (% of patients)	69.8	42.1	<0.001	61.7	58.2	0.70
Diabetes mellitus (% of patients)	13.2	5.9	0.04	11.8	16.2	0.30
Plasma factor VIIa (mU/ml)§	43.8 (41.0–46.5)	40.8 (36.2–46.0)	0.33	44 (40.0–47.9)	42.5 (38.0–47.0)	0.50
No. of stenosed vessels (% of patients)						0.89
One	6			6	6	
Two	18			20	17	
Three	76			74	77	

*Plus–minus values are means ±SD. HDL denotes high-density lipoprotein, and LDL low-density lipoprotein. To convert the values for cholesterol to milligrams per deciliter, divide by 0.02586. To convert the values for triglycerides to milligrams per deciliter, divide by 0.01129.

†Of the 311 patients with coronary artery disease, 285 had documentation sufficient to allow classification according to the presence or absence of a history of myocardial infarction.

‡Body-mass index is calculated as the weight in kilograms divided by the square of the height in meters.

§Values for the factor VIIa are geometric means with 95 percent confidence intervals in parentheses.

sampling had been excluded. The plasma levels of factor VIIa were significantly influenced by each polymorphism (Table 2). The mean level of factor VIIa was 66 percent lower in patients with the *A2A2* genotype than in those with the *A1A1* genotype and was 72 percent lower in patients with the *QQ* genotype than in those with the *RR* genotype; heterozygotes for these alleles had intermediate mean levels. With respect to the *IVS7* polymorphism, mean levels of factor VIIa were lowest in patients with the *aa* genotype and highest in those with *ac*, with intermediate values in patients with the other genotypes for these alleles. By analysis of variance, factor VIIa levels were significantly lower in *aa* homozygotes than in *bb* homozygotes (by an average of 34 percent). Because of the small number of patients with the very rare *c* and *d* alleles, the results of statistical analysis of the factor VIIa levels associated with the presence of these alleles were unreliable.

For each polymorphism, the distributions of genotype frequencies were not significantly different between the entire population of patients with coronary artery disease and the patients without coronary artery disease. The frequencies of *A1A1*, *A1A2*, and *A2A2* were 68.8, 27.0, and 4.2 percent, respectively, in the

patients with coronary artery disease and 71.4, 24.8, and 3.8 percent in the control patients ($P=0.86$). The frequencies of *RR*, *RQ*, and *QQ* were 69.1, 28.6, and 2.3 percent, respectively, in patients with coronary artery disease and 70.7, 25.6, and 3.8 percent in control patients ($P=0.57$). The frequencies of *aa*, *ab*, *bb*, *bc*, *ac*, and *bd* were 12.0, 41.2, 44.5, 1.3, 0.6, and 0.3 percent, respectively, in patients with coronary artery disease and 12.0, 39.1, 45.1, 2.3, 1.5, and 0 percent in control patients ($P=0.87$).

Among the patients with coronary artery disease, those with a history of myocardial infarction were similar to those without such a history with regard to all the conventional risk factors for cardiovascular events except for smoking status and male sex; there were more men and more smokers in the subgroup with a history of myocardial infarction (Table 1). However, there were no significant differences between smokers and nonsmokers or between men and women in the frequencies of factor VII genotypes (data not shown). The distribution of genotypes associated with the *R353Q* and *5'F7* polymorphisms differed significantly between patients who had had myocardial infarction and those who had not (Table 3). With regard to the *IVS7* polymorphism, the *aa* genotype was identified

TABLE 2. PLASMA LEVELS OF FACTOR VIIa IN THE STUDY POPULATION, ACCORDING TO THE GENOTYPES FOR POLYMORPHISMS IN THE FACTOR VII GENE.*

GENOTYPE	NO. OF PATIENTS	MEAN FACTOR VIIa LEVEL (95% CI)†	P VALUE‡
		mU/ml	
5'F7 (promoter)			<0.001
<i>ALA1</i>	252	49.8 (46.9–53.5)	
<i>ALA2</i>	101	33.4 (30.5–36.2)§	
<i>A2A2</i>	16	16.7 (12.4–22.9)§¶	
R353Q (exon 8)			<0.001
<i>RR</i>	252	50.9 (47.4–54.0)	
<i>RQ</i>	107	31.5 (28.7–34.4)§	
<i>QQ</i>	10	14.0 (9.1–21.3)§¶	
IVS7 (intron 7)			<0.001
<i>bb</i>	161	47.7 (43.8–51.4)	
<i>ab</i>	158	41.6 (38.0–45.6)	
<i>aa</i>	40	31.5 (25.5–38.8)§¶	
<i>bc</i>	5	43.3 (21.5–87.3)	
<i>ac</i>	3	52.4 (35.1–77.4)	
<i>bd</i>	1	66**	

*Genotype–phenotype correlation analysis was performed with data from 369 patients (the entire study population after patients taking an anticoagulant drug at the time of blood sampling or in the few days before blood sampling had been excluded).

†The values shown are the untransformed geometric means, but the statistical analysis (analysis of variance with the Tukey procedure for post hoc comparison of the means) was performed on log-transformed values. CI denotes confidence interval.

‡P values are for the overall comparison among patients with a given polymorphism and were calculated by analysis of variance.

§P<0.05 for the comparison with the factor VIIa level in patients with the most frequent genotype.

¶P<0.05 for the comparison with the factor VIIa level in patients with the heterozygous genotype.

||In one patient, determination of the IVS7 genotype was not possible, for technical reasons.

**Only one patient had this genotype.

less often in patients who had had myocardial infarction than in those who had not, but this difference did not reach statistical significance. The results did not change significantly after the exclusion of patients carrying the rare IVS7 alleles (*c* and *d*). Similarly, analysis of the various combinations of the IVS7 genotypes with the 5'F7 or R353Q genotypes showed that certain combinations occurred more often in the subgroup without a history of myocardial infarction than in the subgroup with such a history and hence could be considered protective (for example, the *A2A2aa* combination occurred in 2.7 percent of the patients without a history of myocardial infarction, vs. 1.7 percent of those with such a history), but the relatively limited number in each of these two subgroups did not allow a formal statistical evaluation.

Crude and adjusted odds ratios for myocardial infarction with respect to each genotype are shown in Table 3. The greatest protection from myocardial infarction was conferred by homozygosity for the *A2*

allele; the *A2A2* genotype was associated with a decrease in risk of about 70 percent as compared with the corresponding *ALA1* wild-type genotype (odds ratio, 0.29; 95 percent confidence interval, 0.08 to 1). The frequencies of the *A2* and *Q* alleles were significantly higher among the patients without a history of myocardial infarction than among those with a history of myocardial infarction (24.1 vs. 13.7 percent [P=0.002] for the *A2* allele and 22.3 percent vs. 13.1 percent [P=0.003] for the *Q* allele). The allele-specific adjusted odds ratios were 0.48 (95 percent confidence interval, 0.31 to 0.76) for the *A2* allele and 0.52 (95 percent confidence interval, 0.33 to 0.83) for the *Q* allele. In other words, each of these two alleles was associated with a decrease of about 50 percent in the risk of myocardial infarction.

DISCUSSION

Previous studies established an association between particular factor VII gene polymorphisms and the levels of factor VII antigen and factor VII coagulant activity.^{10,13,19} The results of our study, in which we examined three polymorphisms in the factor VII gene, confirm that there is also an association with the activated form of circulating factor VII, especially for the 5'F7 and R353Q polymorphisms. Moreover, we found that among patients with coronary atherosclerosis who were otherwise similar in terms of the severity of their disease on angiography, the distribution of the genotypes differed significantly between those who did and those who did not have a history of myocardial infarction.

Previous studies of the relation between genetic markers of factor VII and the risk of coronary artery disease yielded various results.^{12–15} Nevertheless, it should be kept in mind that in addition to the critical part that may have been played by population-specific factors (i.e., documented differences in the geographic distribution of carriers of the mutations^{18,20}), there was substantial heterogeneity within the populations studied. With respect to patients with coronary artery disease, most of the previous studies included only survivors of myocardial infarction.^{12–14} In addition, subjects from the general population were generally used as the control group, without a requirement for objective angiographic information about their coronary arteries. Because of this approach, the studies may have included controls who had substantial coronary artery disease, although it was not clinically evident.

Our study extends this previous work because all of our patients, including the control patients, had coronary angiograms and because we included patients with severe, usually multivessel, coronary atherosclerosis, who were classified according to whether or not they had a history of myocardial infarction. The first point is important to allow investigation of the potential association between polymorphisms in the factor VII gene and coronary atherosclerosis — that is, to

TABLE 3. GENOTYPE FREQUENCIES FOR POLYMORPHISMS IN THE FACTOR VII GENE IN PATIENTS WITH CORONARY ARTERY DISEASE, ACCORDING TO THE PRESENCE OR ABSENCE OF A HISTORY OF MYOCARDIAL INFARCTION, AND THE ASSOCIATED RISK OF MYOCARDIAL INFARCTION.*

GENOTYPE	NO MYOCARDIAL INFARCTION		P VALUE†	ODDS RATIO (95% CI)	
	MYOCARDIAL INFARCTION (N=175)	MYOCARDIAL INFARCTION (N=110)		UNIVARIATE ANALYSIS	MULTIVARIATE‡
no. of patients (%)					
5'F7 (promoter)			0.008		
<i>ALA1</i>	132 (75.4)	64 (58.2)		1.0	1.0
<i>ALA2</i>	38 (21.7)	39 (35.5)		0.47 (0.27–0.81)	0.47 (0.27–0.81)
<i>A2A2</i>	5 (2.9)	7 (6.4)		0.34 (0.10–1.10)	0.29 (0.08–1.00)
R353Q (exon 8)			0.01		
<i>RR</i>	132 (75.4)	65 (59.1)		1.0	1.0
<i>RQ</i>	40 (22.9)	41 (37.3)		0.48 (0.28–0.81)	0.47 (0.27–0.81)
<i>QQ</i>	3 (1.7)	4 (3.6)		0.36 (0.08–1.70)	0.46 (0.09–2.20)
IVS7 (intron 7)§			0.8		
<i>aa</i>	19 (11.0)	15 (13.6)		0.73 (0.33–1.60)	0.67 (0.29–1.50)
<i>ab</i>	70 (40.7)	48 (43.6)		0.84 (0.50–1.41)	0.8 (0.47–1.37)
<i>bb</i>	78 (45.3)	45 (40.9)		1.0	1.0
<i>bc</i>	3 (1.7)	1 (0.9)		1.73 (0.17–17.10)	1.65 (0.16–16.70)
<i>ac</i>	1 (0.6)	1 (0.9)		0.57 (0.03–9.44)	0.72 (0.04–12.50)
<i>bd</i>	1 (0.6)	0			

*CI denotes confidence interval. Because of rounding, not all percentages total 100.

†P values are for the overall comparison among patients with a given polymorphism and were calculated by chi-square analysis.

‡The multivariate logistic-regression analysis was adjusted for sex and smoking status.

§Data on IVS7 were missing for three patients with myocardial infarction.

define their potential value as predictors of the risk of atherogenesis. Our clear definitions of phenotypes should have reduced the chance of spurious results, a problem inherent in studies of allelic association.²¹ The genotype frequencies in the entire population of patients with coronary artery disease did not differ significantly from those in patients without coronary artery disease, suggesting that these genetic markers do not influence the development of the atheromatous lesions. This finding is in accordance with those of previous studies that identified no association between the R353Q polymorphism and either the severity of coronary disease on angiography or the occurrence of subclinical carotid atherosclerosis.^{15,22} The frequencies of each allele in our control patients were similar to those reported in patients in other studies from northern Italy (for example, with respect to the R353Q polymorphism, 16.2 percent of our controls carried the *Q* allele, as compared with 16.5 percent of 200 healthy controls in the study by Ardissino et al.²³), so there is no reason to doubt their suitability.

On the other hand, the key finding of our study deals with the risk of thrombotic events associated with polymorphisms in the factor VII gene. The significantly higher prevalence of the *A2* and *Q* alleles in patients with coronary artery disease who did not have a history of myocardial infarction suggests that these

genetic markers correspond to protection from thrombosis. The patients who had had myocardial infarction were similar to those who had not had myocardial infarction not only with respect to the degree of coronary atherosclerosis but also with respect to all classic risk factors for cardiovascular events except smoking and male sex. Nevertheless, adjustment for smoking by logistic regression confirmed the independent protective role of the two alleles. It is well accepted that most myocardial infarctions result from thrombotic occlusion after plaque disruption and exposure of tissue factor to blood.¹ Thus, it is biologically plausible that in persons with low levels of factor VII because of genetic factors, a lesser degree of thrombus formation and a shorter-lasting thrombus could account for a lower risk of myocardial infarction. Our results agree with those of a study by Iacoviello et al.,¹² who found that the frequency of the *Q* and *a* alleles in Italian patients with familial myocardial infarction was lower than that in age-matched controls who were hospitalized for other reasons. Taken together, these two differently designed studies underscore the role of polymorphisms in the factor VII gene as markers of a low susceptibility to myocardial infarction, although the results may be generalizable only to the Italian population. A gradient in the distribution of the factor VII alleles associated with low levels of factor VII, with an

increase from north to south, has been described in the European population.¹⁸ Thus, the inconclusiveness of the results of some previous studies^{13,14} may be due to geographic factors, since they included subjects from populations with a low prevalence of the protective alleles. Also, the predictive power of individual polymorphisms may vary among populations according to differences in the overall prevalence of risk factors and differences in gene–environment interactions.

Among the patients with coronary artery disease in our study, the mean levels of factor VIIa did not differ significantly between those with a history of myocardial infarction and those without it, despite the different distributions of genotypes in these two groups. Several factors may account for this finding. First, homozygotes for the protective alleles were a minority in each group. Second, whereas genotype analysis was performed for all the patients, data on factor VIIa levels from a substantial number of patients could not be included because these patients were receiving concomitant anticoagulant therapy. Moreover, it is well accepted that precise genetic markers may provide a better measure of individual lifelong exposure to a putative risk factor than related plasma measurements, which may vary over time.²⁴ This may be particularly true of factor VIIa levels. Whereas genetic markers are probably the strongest determinants of these levels,¹⁸ a number of well-known, transient, environmental influences^{25,26} may obscure the relation with myocardial infarction when a single measurement is made.

In addition, it is important to recognize that none of the polymorphisms we investigated have been definitely proved to be functional. Expression studies have indicated that the R353Q polymorphism may modulate the secretion of factor VII.²⁷ Others have shown that the polymorphism involving the A1 and A2 alleles, in the promoter, may reduce the rate of transcription, with ensuing reductions in the synthesis of factor VII.²⁸ Indeed, there is linkage disequilibrium between the two polymorphisms, with a degree of allelic association of more than 80 percent, as confirmed in the current study. Recently, van 't Hooft et al.²⁹ described the functional effect of another polymorphism in the factor VII promoter (a substitution of thymine for guanine at position –401), which exhibited complete association with the polymorphism involving A1 and A2. This polymorphism at position –401 strongly influenced the binding of nuclear proteins and was associated with a reduced rate of transcription. The slightly stronger relation we found for the 5'F7 polymorphism as compared with the R353Q polymorphism may be in accordance with this finding.

A limitation of our study is the case–control design; the results need to be confirmed in prospective cohort studies. The Thrombosis Prevention Trial recently showed that low-dose regimens of oral anti-

coagulants independently reduce the rate of death due to coronary heart disease in men at high risk for cardiovascular events.³⁰ Remarkably, the low-normal factor VII levels resulting from low-dose warfarin treatment substantially overlap those associated with the protective factor VII genotypes.³¹ Such a pharmacologic approach could be effectively restricted to persons with “unfavorable” genotypes; those with the protective genotypes could be excluded because of the low probability of benefit and increased risk of bleeding.

In conclusion, our results add evidence of the role of factor VII genotypes in modulating the risk of myocardial infarction. In particular, they may help explain why some patients are at low risk for myocardial infarction, despite the presence of severe, angiographically documented coronary artery disease. In the future, genotyping for factor VII genetic markers may help identify subgroups of patients with coronary artery disease who might benefit from various therapies.

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