Plasma levels of fibrinolytic proteins and the risk of myocardial infarction in men

Mirjam E. Meltzer¹,², Carine J.M. Doggen²,³, Philip G. de Groot¹, Frits R. Rosendaal, MD¹,⁴,⁵, and Ton Lisman¹,⁶

¹Department of Clinical Chemistry and Hematology, University Medical Center Utrecht; ²Department of Clinical Epidemiology, Leiden University Medical Center, Leiden; ³Department of Health Technology and Services Research, School for Management and Governance, University of Twente, Enschede; ⁴Department of Thrombosis and Hemostasis, Leiden University Medical Center, Leiden; ⁵Einthoven Laboratory for Experimental Vascular Medicine, Leiden University Medical Center, Leiden; ⁶Surgical Research Laboratory, Department of Surgery, University Medical Center Groningen, University of Groningen, the Netherlands

Correspondence: Frits R. Rosendaal, Department of Clinical Epidemiology, C7-P, Leiden University Medical Center, P.O. Box 9600, 2300 RC Leiden, The Netherlands; e-mail: f.r.rosendaal@lumc.nl
Hypofibrinolysis as measured with overall clot lysis assays is associated with risk of arterial thrombosis. Individual components of the fibrinolytic system, however, have not been studied extensively in relation to arterial disease or results of studies were inconsistent. The relation between plasminogen and α2-antiplasmin levels and cardiovascular risk factors and the association between plasminogen, α2-antiplasmin, tissue-plasminogen activator (t-PA), and plasminogen activator inhibitor-1 (PAI-1) and risk of myocardial infarction was investigated in the Study of Myocardial Infarctions Leiden (555 men with a first myocardial infarction and 635 controls). α2-Antiplasmin was associated with age and lipid levels while plasminogen correlated with lipids, C-reactive protein and smoking. Increased levels of all fibrinolytic factors were associated with myocardial infarction. Age-adjusted odds ratios (OR) (95% confidence interval) for quartile 4 compared with 1 were 1.7 (1.2-2.3) for plasminogen, 1.9 (1.3-2.6) for α2-antiplasmin, 1.7 (1.2-2.3) for t-PA, and 1.7 (1.2-2.4) for PAI-1. After adjusting for cardiovascular risk factors, only α2-antiplasmin levels remained associated with risk (OR 1.4 (1.0-2.0)). t-PA and PAI-1 levels predominantly reflected lipid levels whereas plasminogen reflected the inflammatory state. Concluding, elevated α2-antiplasmin levels are independently associated with risk of myocardial infarction. t-PA, PAI-1, and plasminogen levels appear to reflect other cardiovascular risk factors.
Introduction

Decreased fibrinolytic potential as measured with overall clot lysis assays has been found to be associated with increased risk of arterial thrombosis, especially in young individuals, in several studies.\textsuperscript{1-3} Surprisingly, plasma levels of individual components of the fibrinolytic system have either not been studied extensively in the context of arterial thrombosis or were not consistently associated with arterial thrombosis. In particular, population-based studies on the role of $\alpha$2-antiplasmin and plasminogen in risk of arterial thrombosis are scarce. In the European Concerted Action on Thrombosis and Disabilities (ECAT) study, a cohort study including patients with angina pectoris, no association was found between levels of $\alpha$2-antiplasmin and risk of myocardial infarction or sudden death.\textsuperscript{4} Unexpectedly, increased levels of plasminogen were associated with an increased risk. In the Atherosclerosis Risk in Communities (ARIC) study, a population-based cohort study on subjects between 44 and 65 years at baseline, a positive association between plasminogen levels and coronary heart disease was also found.\textsuperscript{5}

The results of studies on levels of tissue type plasminogen activator (t-PA) and plasminogen activator inhibitor-1 (PAI-1) have been conflicting. Both t-PA and PAI-1 levels were associated with arterial disease in multiple studies. However, whether increased levels of t-PA and PAI-1 independently increase the risk remained to be elucidated.\textsuperscript{6} PAI-1 is now recognized as a true component of the metabolic syndrome,\textsuperscript{7} which is strongly associated with arterial thrombosis. In several studies the predictive value of PAI-1 and t-PA disappeared after adjusting for cardiovascular risk factors such as body mass index (BMI), insulin resistance, inflammation and lipid levels (reviewed in Meltzer \textit{et al.} \textsuperscript{6}). This may indicate that levels of these fibrinolytic factors are rather a reflection of underlying disease than a direct cause of arterial thrombosis.
Results of studies on the association of plasma levels of Thrombin Activatable Fibrinolysis Inhibitor (TAFI) with risk of arterial thrombosis are also contradictory. Several studies have found increased levels of TAFI to be associated with an increased risk of arterial disease \(^8,9\) while others found no association.\(^10\) We have recently shown increased functional TAFI levels to be associated with a decreased risk of myocardial infarction in the Study of Myocardial Infarctions LEiden (SMILE), a large case-control study in men.\(^11\) In the present study, we investigate associations between levels of plasminogen, \(\alpha_2\)-antiplasmin, PAI-1, and t-PA and risk of myocardial infarction in the SMILE. Furthermore, as knowledge on determinants of plasma levels of \(\alpha_2\)-antiplasmin and plasminogen is scarce the association between established cardiovascular risk factors and these two fibrinolytic factors was also studied.

Methods and materials

Subjects

The design of the SMILE has been described previously.\(^12\) Patients were 560 men with a first myocardial infarction between 1990 and 1996, below the age of 70 at the onset of myocardial infarction. Two of the following three characteristics had to be identifiable in the discharge report or hospital care record to confirm acute myocardial infarction: typical chest pain, electrocardiographical changes indicative of evolving myocardial infarction or a transient rise in cardiac enzymes to more than twice the upper limit of normal.

The control group consisted of 646 men without a history of myocardial infarction. They had undergone a minor orthopedic intervention between January 1990 and May 1996 for which they had received prophylactic anticoagulants for a short period. They had not received anticoagulants in the 6-months prior to participation in the study. Control subjects were frequency matched on 10-year age groups to the patients. Every participant completed a
questionnaire on cardiovascular risk factors including questions on current and former smoking habits and alcohol use, presence of diabetes, and current use of medication. In addition, for patients, presence of diabetes prior to myocardial infarction was retrieved from discharge letters. A person was classified as hypertensive or hypercholesterolemic when he was taking prescription drugs for these conditions. Blood pressure was measured after a rest of at least 10 minutes with the person sitting in an upright position. BMI was derived by dividing weight (in kilograms) by squared height (in meters). All participants gave informed consent. Approval for this study was obtained from the Medical Ethics Committee of the Leiden University Medical Center, Leiden, the Netherlands.

Laboratory analysis
Fasting blood samples of patients and control subjects were drawn from the antecubital vein in Sarstedt Monovette tubes (Sarstedt, Nümbrecht, Germany) and were obtained between July 1994 and February 1997. Blood samples were primarily drawn in the morning (median 9.30 h, 95% before 11.00 h), without a systematic difference between patients and control subjects. Time between myocardial infarction and blood draw ranged from 88 days to 6 years with a median of 2.6 years.

Serum and plasma samples were aliquotted in multiple tubes and immediately stored at – 80°C. Plasma levels of von Willebrand factor (VWF) and C-reactive protein (CRP) and serum total cholesterol, high-density lipoprotein (HDL) cholesterol and triglyceride levels were measured as described previously.12,13

Plasma levels of fibrinolytic factors were measured in citrated plasma. α2-Antiplasmin and plasminogen activity was measured using chromogenic assays (STA Stachrom antiplasmin and STA Stachrom plasminogen from Diagnostica Stago, Asnières, France) and were performed on a STA-R coagulation analyzer using a commercial calibration standard (Diagnostica Stago, Asnières, France) and expressed as a percentage of normal. PAI-1 antigen
levels were measured with a Technozym PAI-1 enzyme-linked immunosorbent assay (ELISA) reagent kit (Kordia, Biopool, the Netherlands) and were expressed in ng/ml. Antigen levels of t-PA were assessed by ELISA using a commercially available mouse anti-t-PA antibody (Nuclilab BV, Ede, The Netherlands) as capture, and a biotin-labelled rabbit anti human t-PA antibody (Nuclilab BV, Ede, The Netherlands) as detecting antibody. Bound detecting antibody was visualised using biotin-labeled streptavidine, followed by Tetramethylbenzidine (TMB) staining. A calibration curve was constructed using purified t-PA (Nuclilab BV, Ede, The Netherlands), and results were expressed as ng/ml.

The intra-assay coefficients of variation were 1.7% for plasminogen, 4.8% for α2-antiplasmin, 7.6% for PAI-1 and 11.4% for t-PA and the inter-assay coefficients of variation were 1.6% for plasminogen, 4.6% for α2-antiplasmin, 5.0% for PAI-1 and 8.1% for t-PA. In 5 patients and 11 control subjects fibrinolytic protein levels were not measured as available plasma was not sufficient, leaving 555 patients and 635 control subjects in the analyses.

Statistical analysis

The association between cardiovascular risk factors and plasma levels of α2-antiplasmin and plasminogen were studied in the control group. Mean α2-antiplasmin and plasminogen levels were calculated with 5th and 95th percentiles for categories of cardiovascular risk factors. Quartiles of blood pressure, total cholesterol, HDL cholesterol, triglyceride, VWF and CRP were defined based on the distribution among control subjects. Multiple linear regression was used to investigate which factors were independently associated with levels of α2-antiplasmin and plasminogen. As the associations between triglycerides, VWF, and CRP with α2-antiplasmin and plasminogen were not linear these variables were entered in the model divided into quartiles resulting in a regression coefficient for a quartile increase of the independent variable. For reasons of comparability systolic blood pressure, HDL cholesterol and total cholesterol were also included in the model as quartiles as were levels of α2-
antiplasmin when studied as determinants of plasminogen and vice versa. Using 10log-transformation for the not normally distributed variables instead of quartiles did not considerably change the results. However, as a regression coefficient of a 10log-transformed variable is more difficult to interpret than a regression coefficient for each quartile increase we present the latter.

To study the effect of fibrinolytic factors on risk of myocardial infarction, levels of α2-antiplasmin, plasminogen, PAI-1, and t-PA were grouped into quartiles based on the distribution among the control subjects, taking the lowest quartile as the reference group for the odds ratio (OR). A 95% confidence interval (CI) was calculated according to the method of Woolf.14 Unconditional logistic regression was performed to adjust for age and other potential confounders (see below). In the logistic regression model age, BMI, blood pressure, VWF, CRP, and lipid levels were included as continuous variables. Triglycerides, VWF and CRP levels were included in the model after 10log-transformation as these variables were not normally distributed. Subgroups were made according to age. An arbitrary cut-off at age 50 years was chosen in accordance to previous publications on the SMILE study.1,15,16 SPSS 16.0 (SPSS, Chicago, IL, USA) was used for statistical analyses.

**Confounders**

Available literature on factors influencing plasma levels of α2-antiplasmin and plasminogen is limited. Associations between established cardiovascular risk factors and levels of α2-antiplasmin or plasminogen were studied in our own data and potential confounders were selected from these analyses and included in the statistical model.

Several studies investigated determinants of t-PA and PAI-1. Therefore, confounding variables in the association between t-PA or PAI-1 and myocardial infarction were chosen from these studies and included in the models, i.e. diabetes, BMI, lipid levels, plasma levels of VWF and CRP, and blood pressure.4,7 Indeed, these factors were also associated with t-PA
and PAI-1 levels in our own data, with the exception of VWF, which was not associated with PAI-1 in our data (data not shown). The analyses on PAI-1 and t-PA and risk of myocardial infarction were also mutually adjusted for each other.

Results
Mean age of the 555 patients with myocardial infarction was 56.3 years (5th-95th percentiles 40.0-68.8 years) and mean age of 635 control subjects was 57.4 (5th-95th percentiles 34.7-72.1 years). Risk factors for arterial disease such as smoking, obesity, diabetes, hypertension, and hypercholesterolemia were more prevalent in patients than in control subjects (Table 1).

Cardiovascular risk factors and plasma levels of α2-antiplasmin and plasminogen
The association between risk factors for myocardial infarction and α2-antiplasmin levels in control subjects is shown in Table 2. α2-Antiplasmin was negatively associated with age, HDL cholesterol, VWF, and systolic blood pressure level, and positively associated with total cholesterol, triglyceride and plasminogen levels. α2-Antiplasmin also increased with BMI although the small group of men (n=10) with BMI<20 kg/m² did not have low levels. To determine the independent effect of these factors on α2-antiplasmin levels, age, BMI, HDL cholesterol, VWF, systolic blood pressure, total cholesterol, triglyceride and plasminogen were simultaneously included in a multiple linear regression model. Age (β=-0.3%/year; 95%CI -0.4;-0.2) and HDL cholesterol (β=-1.2%/quartile; 95%CI -2.2;-0.3) were both negatively associated with α2-antiplasmin. BMI (β=0.3%/kg*m⁻²; 95%CI 0.0;0.6) and total cholesterol (0.8%/quartile; 95%CI -0.1;1.8) were positively associated with α2-antiplasmin. Plasminogen was strongly related to α2-antiplasmin (β=2.5%/quartile; 95%CI 1.8;3.4). Systolic blood pressure (β=-0.5%/quartile; 95%CI -1.5; 0.4), triglycerides (β=-0.2%/quartile; 95%CI -1.3;0.8), and VWF (β=0.0%/quartile) were not associated with α2-antiplasmin. So,
HDL cholesterol, total cholesterol and plasminogen were the strongest determinants of α2-antiplasmin.

Plasminogen levels increased with levels of triglycerides, total cholesterol, and CRP and was increased in smokers (Table 2). Also alcohol use was associated with plasminogen although not in a dose dependent manner as the occasional drinkers had the lowest levels of plasminogen. Similar to α2-antiplasmin, plasminogen increased with BMI but the small group of underweight subjects (BMI < 20 kg/m2) had high levels. We performed a multiple regression including age, BMI, triglycerides, total cholesterol, CRP, alcohol use, smoking, and α2-antiplasmin levels. Except for age, all variables were independently associated with plasminogen. The regression coefficients were 1.6%/quartile (95%CI 0.9;2.4) for total cholesterol and 1.2%/quartile (95%CI 0.2;1.9) for triglycerides. Plasminogen increased 2.9% (95%CI 2.2-3.6) with each quartile increase of CRP and 2.2% (95%CI 1.5-2.9) per quartile increase in α2-antiplasmin. Compared to occasional drinkers of alcohol, regular drinkers had 5.7% higher levels (95%CI 1.5-10.0), and abstainers had 3.4% higher levels (95%CI -1.4;8.2). Smoking increased plasminogen levels with 3.6% (95%CI 1.9-5.2) compared to not smoking. So, plasminogen was strongly associated with variables related to inflammation.

Plasma levels of fibrinolytic proteins and risk of myocardial infarction

α2-antiplasmin

Mean α2-antiplasmin level in patients was 99% (median 98%; 5th-95th percentile 81-119%) and 96% in controls (median 95%; 5th-95th percentile 77-116%). Levels of α2-antiplasmin were associated with risk of myocardial infarction in a dose dependent manner. In men with the highest levels of α2-antiplasmin, the age-adjusted risk was approximately two-fold increased (model 1: OR 1.9; 95%CI 1.3-2.6; 4th quartile (Q) compared with 1st) (Table 3). As lipid levels were the strongest determinants of α2-antiplasmin levels, apart from plasminogen levels, we first adjusted for HDL and total cholesterol (model 2). This reduced the OR but
high levels of α2-antiplasmin were still associated with an increased risk of myocardial infarction (OR 1.5; 95%CI 1.0-2.1). Further adjustment for BMI (model 3; OR 1.5; 95%CI 1.1-2.2) and additional adjustment for plasminogen did not reduce the OR further (model 4; OR 1.4; 95%CI 1.0-2.0). The same analysis in men below 50 years of age resulted in an age-adjusted OR of 2.6 (95%CI 1.2-5.9; Q4 vs. Q1) and 1.6 (95%CI 0.7-3.8) after adjusting for age, HDL and total cholesterol, BMI and plasminogen (model 4). In men older than 50 these age-adjusted ORs were 1.7 (95%CI 1.2-1.7) and 1.4 (95%CI 0.9-2.2) after extensive adjustment.

**Plasminogen**

Mean plasminogen level in patients was 96% (median 96%; 5th-95th percentile 79-115%) and 94% in controls (median 94%; 5th-95th percentile 77-113%). The risk of myocardial infarction increased with each increasing quartile of plasminogen (Table 4). The risk in men with the highest levels of plasminogen was 1.7-fold (95%CI 1.2-2.3; Q4 vs Q1) increased after adjusting for age (model 1). As plasminogen levels were strongly associated with variables related to inflammation, we adjusted for CRP and smoking (model 2) reducing the OR to no effect (OR 1.1 (95%CI 0.7-1.5). Adjusting for age and CRP or age and smoking separately yielded similar results as simultaneous adjustment for age and both CRP and smoking (data not shown). Adding triglycerides, total cholesterol and alcohol use marginally changed the odds ratio (OR 0.9; 95%CI 0.06-1.3; model 3) as did additional adjustment for α2-antiplasmin levels (OR 0.8; 95%CI 0.5-1.2; model 4).

Similar results were found when analyses were performed separately in men below 50 years of age and in men of 50 years and older. Although the age-adjusted risk in young men, was higher (OR 2.6; 95%CI 1.3-5.0; Q4 vs Q1) than in the older (OR 1.3; 95%CI 0.9-1.9; Q4 vs Q1), the increased risks disappeared after further adjustment for smoking, alcohol use and
levels of triglycerides, total cholesterol, and α2-antiplasmin (model 4) (OR 0.8; 95%CI 0.3-1.8 in men <50 years of age and 0.7; 95%CI 0.4-1.1 in men >50 years).

**PAI-1**

Mean PAI-1 level in patients was 107.4 ng/ml (median 69.7 ng/ml; 5th-95th percentile 14.7-316.9 ng/ml) and 88.8 ng/ml in controls (median 54.9 ng/ml; 5th-95th percentile 13.0-302.7 ng/ml). Those with high PAI-1 levels had an increased risk of myocardial infarction (OR 1.7; 95%CI 1.2-2.3; Q4 vs Q1) but no dose response relation was found after adjusting for age (model 1)(Table 5). As PAI-1 is a marker of the insulin resistance syndrome, we first adjusted for triglycerides, HDL and total cholesterol, BMI and diabetes (model 2) resulting in an OR of 1.1 (95%CI 0.8-1.6), with the largest effect after adjusting for triglycerides and HDL cholesterol (data not shown). Adjusting only for age and CRP to determine the role of inflammation in the association between PAI-1 and myocardial infarction only slightly decreased the OR to 1.5 (95% 1.1-2.1; Q4 vs Q1). Further adjustments for other potential confounders (CRP, VWF, systolic and diastolic blood pressure; model 3) did not change the results, neither did additional adjustment for t-PA (model 4). Analysing men above and below 50 years of age separately gave similar results (data not shown).

**t-PA**

Mean t-PA level in patients was 9.2 ng/ml (median 8.9 ng/ml; 5th-95th percentile 5.0-14.6 ng/ml) and 8.8 ng/ml in controls (median 8.1 ng/ml; 5th-95th percentile 4.5-15.3 ng/ml).

The risk of myocardial infarction was increased in men with t-PA levels above the median (age-adjusted OR 2.0; 95%CI 1.4-2.7 (Q3) and OR 1.7; 95%CI 1.2-2.4 (Q4 vs Q1)) (Table 6). Additional adjusting for lipid levels, diabetes and BMI (model 2) attenuated the risk (OR 1.5; 95%CI 1.1-2.2 (Q3) and OR 1.1; 95%CI 0.8-1.6 (Q4)), again with the largest reduction in risk after adjustment for triglycerides or HDL cholesterol (data not shown). In contrast, adjusting
for age and CRP had only little effect on risk, reducing the OR for Q3 to 1.8 (95% 1.3-2.6) and Q4 to 1.6 (95%CI 1.1-2.2) and similar results were found after adjusting for age and VWF as marker of endothelial activation (OR 1.9; 95%CI 1.3-2.6 (Q3) and OR 1.6; 95%CI 1.2-2.3 (Q4)). Including all potential confounders (lipid levels, diabetes, BMI, VWF, CRP, systolic and diastolic blood pressure) reduced the risk slightly more (OR 1.4; 95%CI 1.0-2.0 (Q3); OR 1.0; 95%CI 0.7-1.5 (Q4) model 3). Adding PAI-1 to the statistical model did not influence the risk (model 4). Results were similar in men of 50 years and older (data not shown). In younger men the age-adjusted risk was slightly decreased in Q2 (OR 0.8; 95%CI 0.4-1.4) and over two-fold increased in the upper two quartiles (OR 2.5; 95%CI 1.3-1.6 (Q3) and OR 2.8; 95%CI 1.4-5.4 (Q4)). These ORs decreased to 0.5 (95%CI 0.2-1.0) for Q2, 1.7 (95%CI 0.8-3.4) for Q3 and 1.6 (95%CI 0.7-3.6) when using multivariate model 4.

Discussion

In this study we have shown that increased levels of α2-antiplasmin are associated with a two-fold increased risk of a first myocardial infarction in men. After adjusting for several potential confounders the risk was still 40% increased as compared to individuals with low levels of α2-antiplasmin. Risk of myocardial infarction was also increased in men with elevated levels of plasminogen, PAI-1 and t-PA in age-adjusted models, but this increased risk was not independent of other risk factors.

The present study is the first to show increased α2-antiplasmin levels to be associated with an increased risk of myocardial infarction. This is consistent with established findings on the association between hypofibrinolysis as measured with overall clot lysis assays and an increased risk of arterial thrombosis and with the bleeding tendency observed in patients with α2-antiplasmin deficiency. The ECAT study is the only other large study that examined the relation between α2-antiplasmin levels and risk of arterial disease. In this cohort study, which consisted of approximately 2600 patients with angina pectoris at baseline and 97 events
after two years of follow-up, no association between α2-antiplasmin levels and myocardial infarction and cardiovascular death was found. This difference in study population may explain the divergent results in the ECAT study and the study presented in this paper.

Epidemiological literature on α2-antiplasmin in relation to risk factors for arterial thrombosis is limited. Here we show that plasma levels of α2-antiplasmin are only marginally influenced by established cardiovascular risk factors. Although levels of the majority of haemostatic factors increase with age, α2-antiplasmin levels decreased with age, which is in agreement with results of the ECAT study. There was also a weak negative association between α2-antiplasmin and HDL cholesterol and a positive association with BMI and total cholesterol. Although the association between lipid levels and α2-antiplasmin has been found before, to our knowledge studies on the mechanism linking lipid levels to levels of α2-antiplasmin are at present lacking. There is no obvious explanation for the strong correlation between plasminogen and α2-antiplasmin plasma levels. It is, however, a common finding that several coagulation and fibrinolytic factors cluster together and correlate possibly via a common regulatory mechanism.

We found elevated plasminogen levels to be associated with risk of myocardial infarction, although the risk disappeared after adjustment for potential confounders. The ECAT and ARIC study also found a positive association between plasminogen levels and risk of arterial disease, which is contradictory considering the role of plasminogen in fibrinolysis. In the present study the association disappeared after adjusting for lipid levels, CRP, smoking, and alcohol use. In the ECAT study no adjustments were made apart from study center, age and sex, and the risk of myocardial infarction or sudden death remained only slightly increased after these adjustments. In the ARIC study adjustments were made for several cardiovascular risk factors, although not for triglycerides and CRP, and substantially increased risks were found even after these adjustments. The differences in results between
studies may thus be explained by differences in confounding factors considered in the analyses.

We found that regular drinkers of alcoholic beverages had increased plasminogen levels. In a study on hepatocyte cell lines it has indeed been shown that alcohol increases plasminogen gene expression and a moderate dose of alcohol also increased plasmin levels in mice. Similar to α2-antiplasmin, the association between plasminogen and lipid levels was shown previously but the mechanism behind the association has not been described. In contrast, various pathways relating plasminogen to inflammation have been described. Elevated plasma levels of plasminogen may reflect an increased inflammatory state as plasminogen transcription is increased by interleukin-6. This may also explain the association between plasminogen and smoking, as smoking leads to an increased inflammatory state. Conversely, there is evidence that plasminogen induces an inflammatory response. When bound to several plasminogen receptors, shown for example for the receptors enolase-1 and histone H2B, plasminogen facilitates transmigration and basement membrane degradation and aids in the degradation of the extracellular matrix and recruitment of macrophages. Thus, increased inflammation can cause increased plasminogen levels and, vice versa, increased plasminogen levels can increase the inflammatory state. Consequently, as inflammation increases the risk of myocardial infarction, the positive association between plasminogen and myocardial infarction can be either indirect and just reflecting the inflammatory state or it can indeed be causal. However, after adjusting for smoking alone, which, in contrast to high CRP levels, can only cause and not result from an increased inflammatory state or high plasminogen levels, we find that the association between plasminogen and myocardial infarction disappears. This provides evidence against a causal role for plasminogen in myocardial infarction.

Both t-PA and PAI-1 were not independently associated with myocardial infarction in our study. Previous studies on t-PA and PAI-1 levels gave conflicting results. While some
studies have found increased t-PA and PAI-1 levels to be associated with an increased risk of arterial disease,\textsuperscript{27-29} several studies have found no association,\textsuperscript{5,30,31} and in two studies even a trend towards a decreased risk was found in subjects with elevated PAI-1.\textsuperscript{30,32} These inconsistent results have been ascribed to the adjustments made for confounders in the analyses as these vary across studies.\textsuperscript{4,6} In the ECAT study, the prognostic value of t-PA and PAI-1 was studied after adjustment for clusters of confounding variables.\textsuperscript{4} Our results on t-PA and PAI-1 are to a large extent in agreement with this study. In the ECAT study the risk of arterial disease associated with increased PAI-1 levels disappeared after adjustment for parameters associated with insulin resistance (BMI, triglycerides, HDL cholesterol, systolic blood pressure, diabetes). In our study these factors, and particularly lipid levels, could also explain the association between PAI-1 and myocardial infarction. In accordance with results of the ECAT study, the effect of t-PA on arterial disease risk was explained by the combination of the markers of insulin resistance, and although to a lesser extent, to CRP as marker of inflammation and VWF as indicator of endothelial activation. Our results are also, at least in part, in agreement with two meta-analyses conducted on the association between t-PA and PAI-1 and coronary heart disease.\textsuperscript{33} PAI-1 was not associated with coronary heart disease in this meta-analysis including 6 prospective cohort studies. In the meta-analysis including 12 prospective cohort studies on t-PA and coronary heart disease an OR of 1.48 was found for the third tertile of t-PA compared with the first. There was, however, evidence of publication bias, showing a tendency to more extreme ORs in the smaller studies and no adjustments were made for inflammation or VWF suggesting that this risk estimate was overestimated.

Besides differences in statistical models used, the roles of the fibrinolytic proteins other than in clot dissolution may also have attributed to the inconsistency of results of different studies (reviewed in Meltzer \textit{et al.} \textsuperscript{6}). Besides its role in clot lysis, plasminogen possibly contributes to destabilisation of atherosclerotic plaques independent of fibrin
proteolysis. Plasmin activates several matrix metalloproteinases (MMPs)\textsuperscript{34} and it has been consistently shown that MMPs increase atherosclerosis progression and plaque instability (reviewed by Newby \textsuperscript{35}).

Furthermore, PAI-1 can both promote and suppress vascular remodeling by mechanisms not directly related to clot lysis. Local PAI-1 levels have been shown to be associated with severity of atherosclerosis\textsuperscript{36} and PAI-1 deficient mice were found to be protected against atherosclerosis progression in the carotid artery.\textsuperscript{37} In contrast PAI-1 may also protect against abnormal matrix remodeling in advanced atherosclerotic plaques as well as plaque rupture and destabilisation of a plaque caused by urokinase plasminogen activator.\textsuperscript{38,39} Consequently, heterogeneity in atherogenic state within and between study populations may hamper direct comparison of study results. Future research may gain more insight in the role of fibrinolytic factors in arterial disease by taking this into account.

While decreased overall fibrinolytic potential as measured with an overall plasma clot lysis assay has been shown to be associated with an increased risk of myocardial infarction in the SMILE,\textsuperscript{1} only elevated $\alpha_2$-antiplasmin levels were associated with risk, whereas increased TAFI levels even strongly protected against myocardial infarction.\textsuperscript{11,40} An explanation for our findings is that CLT measures the combination of the individual factors, also taking the interplay between them into account. The complex interplay between proteins in the fibrinolytic process is not taken into account when plasma levels of the individual factors are measured. Indeed, we recently showed that the clot lysis time is sensitive for fibrinolytic factors, but that the outcome of the assay appears to be determined by factors beyond plasma levels of fibrinolytic proteins.\textsuperscript{40}

A limitation of the case-control study design is that levels of the fibrinolytic proteins are measured after the event, and that post-event levels may not reflect levels prior to the myocardial infarction. Blood samples were drawn several months or even years after the event by which we attempted to minimize the likelihood that an acute phase reaction was
responsible for the plasma levels of the fibrinolytic proteins. Furthermore, we made adjustments for CRP levels, which is an acute phase protein. Caution is, however, needed in interpreting the results and replication of our results is required using studies with other study designs, preferably adequately powered prospective studies. From our study we conclude that increased levels of α2-antiplasmin are associated with an increased risk of myocardial infarction in men. PAI-1, t-PA and plasminogen are no independent risk factors for myocardial infarction. Plasminogen is primarily a marker of inflammation, while high PAI-1 and t-PA levels predominantly reflect increased lipid levels and to a lesser extent inflammation. In addition, endothelial activation may in part explain the association between elevated levels of t-PA and myocardial infarction.

Acknowledgements

This work was supported by grants from the Netherlands Heart Foundation (Grant no. 2005B060 and 92.345).

The authors thank the cardiologists at Leiden University Medical Center and Diaconessenhuis Leiden, F. J. M. van der Meer, T. Visser, J. J. Schreijer, I. de Jonge, J. H. M. Souverijn, and J. Adelmeijer for their assistance.

Contributions: M.E.M. designed the present study, analyzed and interpreted the data, drafted the manuscript; C.J.M.D. designed the overall study, performed the data collection, interpreted the data, and critically reviewed the analyses and the manuscript; P.G.d.G. interpreted the data and critically reviewed the analyses and the manuscript; F.R.R. designed the overall study, interpreted the data, and critically reviewed the analyses and the manuscript; T.L. designed the present study, interpreted the data, critically reviewed the analyses and participated in writing the manuscript.

Conflict-of-interest disclosure: The authors declare no competing financial interests.
Correspondence: Frits R. Rosendaal, Department of Clinical Epidemiology, C7-P, Leiden University Medical Center, P.O. Box 9600, 2300 RC Leiden, The Netherlands; e-mail: f.r.rosendaal@lumc.nl
Reference List


Table 1 Characteristics of men with first myocardial infarction and control subjects

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Patients</th>
<th>Control subjects</th>
</tr>
</thead>
<tbody>
<tr>
<td>N</td>
<td>555</td>
<td>635</td>
</tr>
<tr>
<td>Mean age, y (5th-95th percentile)</td>
<td>56.3 (40.0-68.8)</td>
<td>57.4 (34.7-72.1)</td>
</tr>
<tr>
<td>Current smoking, no. (%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>208 (37.5)</td>
<td>426 (67.1)</td>
</tr>
<tr>
<td>Yes</td>
<td>347 (62.5)</td>
<td>209 (32.9)</td>
</tr>
<tr>
<td>Alcohol use, no. (%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Never</td>
<td>86 (15.5)</td>
<td>64 (10.1)</td>
</tr>
<tr>
<td>Occasionally</td>
<td>24 (4.3)</td>
<td>20 (3.1)</td>
</tr>
<tr>
<td>Regularly</td>
<td>445 (80.2)</td>
<td>551 (86.8)</td>
</tr>
<tr>
<td>BMI, * no. (%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Less than 20 kg/m²</td>
<td>4 (0.7)</td>
<td>10 (1.6)</td>
</tr>
<tr>
<td>20-24 kg/m²</td>
<td>152 (27.4)</td>
<td>182 (28.7)</td>
</tr>
<tr>
<td>25-30 kg/m²</td>
<td>305 (55.1)</td>
<td>341 (53.8)</td>
</tr>
<tr>
<td>At least 30 kg/m²</td>
<td>93 (16.8)</td>
<td>101 (15.9)</td>
</tr>
<tr>
<td>Diabetes, no. (%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Absent</td>
<td>529 (95.3)</td>
<td>614 (96.7)</td>
</tr>
<tr>
<td>Present</td>
<td>26 (4.7)</td>
<td>21 (3.3)</td>
</tr>
</tbody>
</table>
### Hypertension, no. (%)  

<table>
<thead>
<tr>
<th>Absent</th>
<th>449 (80.9)</th>
<th>529 (83.3)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Present</td>
<td>106 (19.1)</td>
<td>106 (16.7)</td>
</tr>
</tbody>
</table>

### Hypercholesterolemia, no. (%)  

<table>
<thead>
<tr>
<th>Absent</th>
<th>544 (98.0)</th>
<th>623 (98.1)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Present</td>
<td>11 (2.0)</td>
<td>12 (1.9)</td>
</tr>
</tbody>
</table>

Data for patients refer to the period prior to myocardial infarction, apart from BMI.

BMI indicates body mass index.

* For 1 patient and 1 control subject BMI was missing.
<table>
<thead>
<tr>
<th>Age (year)</th>
<th>n</th>
<th>Mean α2-antiplasmin level</th>
<th>5th-95th percentile</th>
<th>Mean plasminogen level</th>
<th>5th-95th percentile</th>
</tr>
</thead>
<tbody>
<tr>
<td>27-39</td>
<td>54</td>
<td>104</td>
<td>84-122</td>
<td>93</td>
<td>74-107</td>
</tr>
<tr>
<td>40-49</td>
<td>103</td>
<td>98</td>
<td>80-121</td>
<td>94</td>
<td>77-113</td>
</tr>
<tr>
<td>50-59</td>
<td>178*</td>
<td>97</td>
<td>77-115</td>
<td>97</td>
<td>78-114</td>
</tr>
<tr>
<td>60-69</td>
<td>243</td>
<td>93</td>
<td>74-114</td>
<td>94</td>
<td>77-114</td>
</tr>
<tr>
<td>70-75</td>
<td>57</td>
<td>90</td>
<td>78-108</td>
<td>91</td>
<td>72-115</td>
</tr>
<tr>
<td>BMI (kg/m²)†</td>
<td>Less than 20</td>
<td>10</td>
<td>97</td>
<td>74-115</td>
<td>99</td>
</tr>
<tr>
<td></td>
<td>20-24</td>
<td>182</td>
<td>94</td>
<td>74-112</td>
<td>92</td>
</tr>
<tr>
<td></td>
<td>25-29</td>
<td>341</td>
<td>96</td>
<td>78-116</td>
<td>94</td>
</tr>
<tr>
<td>diastolic blood pressure (mmHg)‡</td>
<td>At least 95</td>
<td>174</td>
<td>97</td>
<td>78-116</td>
<td>96</td>
</tr>
<tr>
<td>----------------------------------</td>
<td>-------------</td>
<td>-----</td>
<td>----</td>
<td>--------</td>
<td>----</td>
</tr>
<tr>
<td>no</td>
<td>21</td>
<td>95</td>
<td>78-123</td>
<td>92</td>
<td>70-113</td>
</tr>
<tr>
<td>yes</td>
<td>614*</td>
<td>96</td>
<td>77-116</td>
<td>94</td>
<td>77-113</td>
</tr>
<tr>
<td>le&lt; than 80</td>
<td>199</td>
<td>96</td>
<td>76-119</td>
<td>93</td>
<td>77-115</td>
</tr>
<tr>
<td>85</td>
<td>90*</td>
<td>96</td>
<td>81-116</td>
<td>96</td>
<td>76-114</td>
</tr>
<tr>
<td>90</td>
<td>167</td>
<td>94</td>
<td>74-114</td>
<td>93</td>
<td>74-114</td>
</tr>
<tr>
<td>at least 95</td>
<td>174</td>
<td>97</td>
<td>78-116</td>
<td>96</td>
<td>78-112</td>
</tr>
<tr>
<td>systolic blood pressure (mmHg)‡</td>
<td>less than 125</td>
<td>157</td>
<td>98</td>
<td>75-121</td>
<td>94</td>
</tr>
<tr>
<td>130-135</td>
<td>114</td>
<td>95</td>
<td>77-116</td>
<td>94</td>
<td>80-109</td>
</tr>
<tr>
<td>140-150</td>
<td>200*</td>
<td>96</td>
<td>78-115</td>
<td>94</td>
<td>75-115</td>
</tr>
<tr>
<td>at least 155</td>
<td>159</td>
<td>93</td>
<td>78-112</td>
<td>95</td>
<td>76-115</td>
</tr>
<tr>
<td>triglycerides (mmol/L)‡</td>
<td>less than 0.90</td>
<td>157</td>
<td>94</td>
<td>74-115</td>
<td>90</td>
</tr>
<tr>
<td>Range</td>
<td>Value1</td>
<td>Value2</td>
<td>Value3</td>
<td>Value4</td>
<td>Value5</td>
</tr>
<tr>
<td>------------</td>
<td>--------</td>
<td>--------</td>
<td>--------</td>
<td>--------</td>
<td>--------</td>
</tr>
<tr>
<td>total cholesterol (mmol/L)†</td>
<td>0.90-1.24</td>
<td>159</td>
<td>95</td>
<td>76-113</td>
<td>94</td>
</tr>
<tr>
<td></td>
<td>1.25-1.82</td>
<td>161</td>
<td>96</td>
<td>78-117</td>
<td>96</td>
</tr>
<tr>
<td></td>
<td>at least 1.83</td>
<td>157</td>
<td>98</td>
<td>81-118</td>
<td>98</td>
</tr>
<tr>
<td></td>
<td>less than 5.17</td>
<td>161</td>
<td>94</td>
<td>75-113</td>
<td>91</td>
</tr>
<tr>
<td></td>
<td>5.17-5.81</td>
<td>156</td>
<td>95</td>
<td>76-117</td>
<td>93</td>
</tr>
<tr>
<td></td>
<td>5.82-6.59</td>
<td>157</td>
<td>97</td>
<td>80-119</td>
<td>94</td>
</tr>
<tr>
<td></td>
<td>at least 6.60</td>
<td>160</td>
<td>98</td>
<td>80-118</td>
<td>99</td>
</tr>
<tr>
<td>HDL-cholesterol (mmol/L)§</td>
<td>less than 1.1</td>
<td>159</td>
<td>98</td>
<td>78-116</td>
<td>95</td>
</tr>
<tr>
<td></td>
<td>1.1-1.28</td>
<td>156</td>
<td>97</td>
<td>80-119</td>
<td>94</td>
</tr>
<tr>
<td></td>
<td>1.29-1.53</td>
<td>162</td>
<td>94</td>
<td>73-114</td>
<td>94</td>
</tr>
<tr>
<td></td>
<td>at least 1.54</td>
<td>155</td>
<td>95</td>
<td>78-117</td>
<td>94</td>
</tr>
<tr>
<td>C-reactive protein (mg/L)</td>
<td>less than 0.78</td>
<td>159</td>
<td>95</td>
<td>74-113</td>
<td>89</td>
</tr>
<tr>
<td>Smoking</td>
<td>No</td>
<td>Yes</td>
<td>Alcohol Use</td>
<td>No</td>
<td>Occasionally</td>
</tr>
<tr>
<td>------------------</td>
<td>-----</td>
<td>-----</td>
<td>----------------------</td>
<td>------</td>
<td>--------------</td>
</tr>
<tr>
<td></td>
<td>161</td>
<td>96</td>
<td>78-116</td>
<td>159*</td>
<td>96</td>
</tr>
<tr>
<td></td>
<td>156</td>
<td>96</td>
<td>78-116</td>
<td>159*</td>
<td>96</td>
</tr>
<tr>
<td></td>
<td>156</td>
<td>96</td>
<td>78-116</td>
<td>159*</td>
<td>96</td>
</tr>
<tr>
<td></td>
<td>156</td>
<td>96</td>
<td>78-116</td>
<td>159*</td>
<td>96</td>
</tr>
</tbody>
</table>

- Smoking
  - No: 426* 96 78-117 93 76-112
  - Yes: 209 95 75-115 98 80-116

- Alcohol Use
  - No: 64 96 79-117 92 74-111
  - Occasionally: 20 95 55-123 89 70-106
  - Yes: 551* 96 77-116 95 77-114

- VWF (%)
  - Less than 97: 159 97 77-117 92 74-108
  - 98-124: 151 97 81-118 94 78-114
  - 125-158: 145 96 74-114 96 77-115
  - At least 159: 152 93 75-114 96 77-117
<table>
<thead>
<tr>
<th></th>
<th>less than 87</th>
<th>171</th>
<th>91</th>
<th>74-109</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>87-93</td>
<td>152</td>
<td>95</td>
<td>76-114</td>
</tr>
<tr>
<td></td>
<td>94-100</td>
<td>155</td>
<td>97</td>
<td>81-116</td>
</tr>
<tr>
<td>at least 101</td>
<td>156</td>
<td>100</td>
<td></td>
<td>81-120</td>
</tr>
<tr>
<td>α2-antiplasmin (%)</td>
<td>less than 87</td>
<td>163</td>
<td></td>
<td>91</td>
</tr>
<tr>
<td></td>
<td>87-94</td>
<td>163</td>
<td>93</td>
<td>76-112</td>
</tr>
<tr>
<td></td>
<td>95-104</td>
<td>153</td>
<td>95</td>
<td>76-113</td>
</tr>
<tr>
<td>at least 105</td>
<td>155</td>
<td></td>
<td>99</td>
<td>84-116</td>
</tr>
</tbody>
</table>

BMI indicates body mass index; HDL-cholesterol, High-density lipoprotein cholesterol; and VWF von Willebrand factor.

* For 1 control subject plasminogen levels were missing

† For 1 control subject BMI was missing

‡ For 5 control subjects systolic and diastolic blood pressure was missing
§ For 3 control subjects HDL cholesterol was missing

|| For 28 control subjects WVF was missing
Table 3 Risk of myocardial infarction with increasing quartiles of α2-antiplasmin

<table>
<thead>
<tr>
<th>Quartile</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
</tr>
</thead>
<tbody>
<tr>
<td>cut-off point (%)</td>
<td>86</td>
<td>93</td>
<td>100</td>
<td></td>
</tr>
<tr>
<td>patients</td>
<td>96</td>
<td>128</td>
<td>152</td>
<td>178</td>
</tr>
<tr>
<td>control subjects</td>
<td>163</td>
<td>163</td>
<td>154</td>
<td>155</td>
</tr>
<tr>
<td>model 1: age</td>
<td>1</td>
<td>1.3 (0.9-1.9)</td>
<td>1.6 (1.2-2.3)</td>
<td>1.9 (1.3-2.6)</td>
</tr>
<tr>
<td>model 2: Model 1 + HDL and total cholesterol</td>
<td>1</td>
<td>1.2 (0.8-1.7)</td>
<td>1.5 (1.0-2.1)</td>
<td>1.5 (1.0-2.2)</td>
</tr>
<tr>
<td>model 3: Model 2 + BMI</td>
<td>1</td>
<td>1.2 (0.8-1.7)</td>
<td>1.5 (1.0-2.1)</td>
<td>1.5 (1.0-2.2)</td>
</tr>
<tr>
<td>model 4: Model 3 + plasminogen</td>
<td>1</td>
<td>1.2 (0.8-1.7)</td>
<td>1.4 (1.0-2.0)</td>
<td>1.4 (1.0-2.0)</td>
</tr>
</tbody>
</table>

BMI indicates body mass index; and HDL-cholesterol, High-density lipoprotein cholesterol.

* For 1 patient α2-antiplasmin levels were missing
Table 4 Risk of myocardial infarction with increasing quartiles of plasminogen

<table>
<thead>
<tr>
<th>Quartile</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
</tr>
</thead>
<tbody>
<tr>
<td>cut-off point (%)</td>
<td>86</td>
<td>94</td>
<td>104</td>
<td></td>
</tr>
<tr>
<td>patients</td>
<td>112</td>
<td>125</td>
<td>144</td>
<td>174</td>
</tr>
<tr>
<td>control subjects</td>
<td>171</td>
<td>152</td>
<td>155</td>
<td>156</td>
</tr>
<tr>
<td>model 1: age</td>
<td>1</td>
<td>1.2 (0.9-1.7)</td>
<td>1.4 (1.0-1.9)</td>
<td>1.7 (1.2-2.3)</td>
</tr>
<tr>
<td>model 2: Model 1 + CRP and smoking</td>
<td>1</td>
<td>1.0 (0.7-1.5)</td>
<td>1.1 (0.8-1.5)</td>
<td>1.1 (0.7-1.5)</td>
</tr>
<tr>
<td>Model 3: Model 2 + triglycerides, total cholesterol, and alcohol use</td>
<td>1</td>
<td>1.0 (0.7-1.4)</td>
<td>1.0 (0.7-1.5)</td>
<td>0.9 (0.6-1.3)</td>
</tr>
<tr>
<td>Model 4: Model 3 + α2-antiplasmin</td>
<td>1</td>
<td>0.9 (0.6-1.3)</td>
<td>0.9 (0.6-1.3)</td>
<td>0.8 (0.5-1.2)</td>
</tr>
</tbody>
</table>

CRP indicates C-reactive protein.

* For 1 control subject plasminogen levels were missing;
Table 5 Risk of myocardial infarction with increasing quartiles of PAI-1

<table>
<thead>
<tr>
<th>Quartile</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
</tr>
</thead>
<tbody>
<tr>
<td>cut-off point (ng/ml)</td>
<td>32.9</td>
<td>54.9</td>
<td>99.0</td>
<td></td>
</tr>
<tr>
<td>patients</td>
<td>118</td>
<td>109</td>
<td>121</td>
<td>202</td>
</tr>
<tr>
<td>control subjects</td>
<td>158</td>
<td>159</td>
<td>159</td>
<td>158</td>
</tr>
</tbody>
</table>

Model 1: age

|          | 1      | 0.9 (0.7-1.3) | 1.0 (0.7-1.4) | 1.7 (1.2-2.3) |

Model 2: Model 1 + HDL and total cholesterol, triglycerides, BMI and diabetes

|          | 1      | 0.8 (0.6-1.2) | 0.8 (0.6-1.2) | 1.1 (0.8-1.6) |

Model 3: Model 2 + CRP, VWF, systolic and diastolic blood pressure

|          | 1      | 0.8 (0.6-1.2) | 0.8 (0.6-1.2) | 1.1 (0.8-1.7) |

Model 4: Model 3 + t-PA

|          | 1      | 0.8 (0.6-1.2) | 0.8 (0.6-1.2) | 1.1 (0.8-1.7) |

PAI-1 indicates plasminogen activator inhibitor 1; BMI, body mass index; HDL-cholesterol, High-density lipoprotein cholesterol; CRP, C-reactive protein; VWF, von Willebrand factor; and t-PA, tissue plasminogen activator.

* For 1 control subject and 5 patients PAI-1 levels were missing
Table 6 Risk of myocardial infarction with increasing quartiles of t-PA

<table>
<thead>
<tr>
<th>Quartile</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
</tr>
</thead>
<tbody>
<tr>
<td>cut-off point (ng/ml)</td>
<td>6.3</td>
<td>8.1</td>
<td>10.6</td>
<td></td>
</tr>
<tr>
<td>patients</td>
<td>104</td>
<td>104</td>
<td>185</td>
<td>160</td>
</tr>
<tr>
<td>control subjects</td>
<td>158</td>
<td>159</td>
<td>159</td>
<td>158</td>
</tr>
<tr>
<td>Model 1: age</td>
<td>1</td>
<td>1.1 (0.8-1.5)</td>
<td>2.0 (1.4-2.7)</td>
<td>1.7 (1.2-2.4)</td>
</tr>
<tr>
<td>Model 2: Model 1 + HDL and total cholesterol, triglycerides, BMI and diabetes</td>
<td>1</td>
<td>0.9 (0.6-1.3)</td>
<td>1.5 (1.1-2.2)</td>
<td>1.1 (0.8-1.6)</td>
</tr>
<tr>
<td>Model 3: Model 2 + CRP, VWF, systolic and diastolic blood pressure</td>
<td>1</td>
<td>0.9 (0.6-1.4)</td>
<td>1.4 (1.0-2.0)</td>
<td>1.0 (0.7-1.5)</td>
</tr>
<tr>
<td>Model 4: Model 3 + PAI-1</td>
<td>1</td>
<td>0.9 (0.6-1.3)</td>
<td>1.4 (1.0-2.0)</td>
<td>1.0 (0.6-1.5)</td>
</tr>
</tbody>
</table>

t-PA indicates tissue plasminogen activator; BMI, body mass index; HDL-cholesterol, High-density lipoprotein cholesterol; CRP, C-reactive protein; VWF, von Willebrand factor; and PAI-1, plasminogen activator inhibitor 1.

* For 1 control subject and 2 patients t-PA levels were missing