Increased Lipoprotein(a) Is an Important Risk Factor for Venous Thromboembolism in Childhood

Ulrike Nowak-Göttl, MD; Ralf Junker, MD; Marion Hartmeier, MD; Hans-Georg Koch, MD; Nicole Münchow, MD; Gerd Assmann, MD; Arnold von Eckardstein, MD

***Background*—**Serum levels of lipoprotein(a) [Lp(a)] are determined largely by genetic variation in the gene encoding for apolipoprotein(a) [apo(a)], the specific protein component of Lp(a) that is very homologous to plasminogen. High plasma levels of Lp(a) increase the risk for premature atherosclerotic vessel diseases. We investigated the little-characterized role of Lp(a) as a risk factor for venous thromboembolic diseases, alone and in conjunction with established thrombophilic risk factors of proteins regulating blood coagulation and fibrinolysis.

***Methods and Results*—**Serum levels of Lp(a) and lipids, protein C, protein S, and antithrombin, as well as the size of apo(a) isoforms and the presence of the factor V:Q^{506} mutation, were determined in 186 consecutively admitted children from neonates to 18 years old with a history of venous thrombosis and in 186 age- and disease-matched control subjects. Children with a history of venous thrombosis had a significantly higher median Lp(a) level (19 versus 4.4 mg/dL) than control subjects. The risk for thromboembolic events in children with Lp(a) levels in the upper quartile, ie, >30 mg/dL, was 7.2 (95% CI, 3.7 to 14.5). The size of apo(a) isoforms was inversely related to Lp(a) levels and to the risk for thromboembolic events. Compared with the highest quartile of the apo(a) size distribution, the lowest quartile was associated with a risk of 8.2. In addition, multivariate statistical analysis gives evidence that the factor V:Q^{506} mutation (OR/CI, 2.8/1.6 to 4.9), protein C (OR/CI, 6.5/2.1 to 19), and antithrombin deficiency (OR/CI, 10.4/1.2 to 90) were independent risk factors of childhood venous thrombosis. Coincidence of elevated Lp(a) with factor V:Q^{506} mutation or deficiencies of protein C or antithrombin further increased the risk for thromboembolic events to 8.4.

***Conclusions*—**Lp(a) >30 mg/dL is a risk factor for venous thromboembolism in childhood. Lp(a) measurements should be included in the screening of causal factors in children with venous thromboembolic events. **(*Circulation*. 1999;100:743-748.)**

Key Words: lipoproteins ■ thrombus ■ embolism ■ pediatrics ■ genetics

Various genetic defects of proteins regulating blood coagulation have been established as risk factors for thromboembolic diseases. Most of them affect the protein C pathway, namely, deficiencies of protein C and protein S as well as the factor V mutation FV:Q^{506}, which underlies the resistance of activated protein C. Prothrombotic states have also been associated with defects in the genes of antithrombin, plasminogen, and fibrinogen.[1–3] Finally, the previously described 20210GA variant of the prothrombin gene also seems to be a common but probably mild risk factor for arterial and venous thromboembolism.[4]

Many prospective and case-control studies identified elevated levels of lipoprotein(a) [Lp(a)] as a risk factor for premature myocardial infarction and stroke.[5–13] Lp(a) resembles LDL in its high cholesterol content and in the presence of 1 molecule of apolipoprotein (apo) B. The additional protein in Lp(a) is called apo(a) and contains a protease domain, a kringle V–like domain, and a variable number of kringle IV repeats, all of which have strong structural homologies to plasminogen.[14,15] The genetically determined variation in the number of kringle IV repeats leads to the synthesis of apo(a) isoforms whose molecular weight varies between 200 and 800 kDa and is inversely correlated to Lp(a) plasma levels.[14,16] The size polymorphism and additional genetic variation of the apo(a) gene largely control Lp(a) levels so that Lp(a) concentrations have intraindividual and interindividual variability, with serum levels ranging between 0 and 300 mg/dL.[14,16]

Both in vitro and in vivo, Lp(a) has antifibrinolytic properties[15,17–19] that have in part been made responsible for its association with cardiovascular disease. In this respect, it is surprising that only few data are available on the role of Lp(a) as a risk factor for venous thrombosis and thromboembolic stroke of venous origin. Some small-scale studies and

Received October 19, 1998; revision received April 6, 1999; accepted May 26, 1999.

From the Department of Pediatrics (U.N.-G., M.H., H.-G.K., N.M.), the Department of Clinical Chemistry and Laboratory Medicine (R.J., G.A., A.v.E.), and the Department of Arteriosclerosis Research (R.J., G.A., A.v.E.), Westfälische Wilhelms-Universität, Münster, and the University of Hamburg (N.M.), Germany.

Correspondence to Prof Dr Ulrike Nowak-Göttl, Westfälische Wilhelms-Universität, Department of Pediatrics, Pediatric Hematology, and Oncology, Albert-Schweitzer-Straße 33, D-48149 Münster, Germany.

© 1999 American Heart Association, Inc.

Circulation **is available at http://www.circulationaha.org**

single case reports have indicated that elevated Lp(a) levels may further increase the risk of thrombotic events in patients suffering from an acquired prothrombotic state due to rheumatic diseases, polycythemia vera, or nephrotic syndrome.[20–25] In 35 children who were consecutively referred to us because of venous thrombosis, we previously observed an increased prevalence of Lp(a) >50 mg/dL.[26] To investigate whether elevated Lp(a) levels are associated with the occurrence of thromboembolic events in childhood, we have now conducted a controlled multicenter case-control study. The frequency distribution of apo(a) isoforms was also analyzed to rule out the possibility that differences in Lp(a) levels among case and control subjects are secondary to the thromboembolic events or their treatment. Finally, we compared the relative risks associated with elevated Lp(a) or established thrombophilic risk factors and determined their cooperative effects on the occurrence of thromboembolic events.

Methods

Study Population

The present multicenter case-control study was performed in accordance with the ethical standards laid down in the relevant version of the 1964 Declaration of Helsinki and approved by the medical ethics committee at the Westfälische Wilhelms-University, Münster, Germany.

Inclusion in this study was subject to the following criteria: (1) thrombotic events had to occur before the age of 18 years; (2) thromboembolism had to be confirmed objectively by standard imaging methods, ie, duplex sonography, venography, CT and MR imaging for the diagnosis of venous thromboembolism, and cerebral CT scanning, MR imaging, MR angiography, or transcranial Doppler ultrasonography for the diagnosis of thromboembolic ischemic stroke; and (3) to prevent confounding effects of acute reactive processes or oral anticoagulation, ≥3 months had to pass between the last thrombotic episode and blood sample collection for the quantitative assays described below. Patients with abnormal quantitative laboratory test results were followed up for a minimum of 6 weeks after the first examination to obtain ≥1 further blood sample for reanalysis.

Since 1996, the above criteria were fulfilled by 186 newly admitted patients 0 to 18 years old (median age at first thrombotic event 5.5 years) from different geographic areas of Northwest Germany. The thrombotic manifestations included isolated deep vein thrombosis (n=42), central nervous thrombosis (n=32), renal venous thrombosis (n=10), axillar vein thrombosis (n=8), subclavian vein thrombosis (n=9), superior caval vein thrombosis (n=11), inferior caval vein thrombosis (n=12), portal vein thrombosis (n=12), hepatic vein thrombosis (n=1), veno-occlusive disease (n=2), multiple venous thrombosis (n=5), deep vein thrombosis with pulmonary embolism (n=2), pulmonary embolism (n=10), retinal venous thrombosis (n=2), and thromboembolic stroke of venous origin (patent foramen ovale: n=28).

With informed parental consent, samples from 186 age- and disease-matched children from the same geographic areas were investigated as controls. Underlying diseases of patients and control subjects are shown in Table 1.

TABLE 1. Underlying Diseases in Patients (n=186) and Control Subjects (n=186)

	Patients	Control Subjects
Age, y	5 (0–18)	5.5 (0–18)
Male:female ratio	1:1.1	1:1.1
No. of underlying diseases	74	77
Central lines†	28	25
Severe bacterial/viral infection†	20	18
Streptococci	3	3
Staphylococci	4	4
Haemophilus influenzae	2	2
Pneumococci	2	2
Escherichia coli	3	2
Pseudomonas aeruginosa	2	1
Varicella-zoster virus	2	2
Adenovirus	2	2
Malignancy†	22	23
Acute leukemia	14	15
Non-Hodgkins lymphoma	2	2
Hodgkins lymphoma	1	1
Neuroblastoma	2	2
Nephroblastoma	3	3
Rheumatic diseases†	5	4
Cardiac diseases†‡	28	30
TGA	4	4
TrA	5	5
HLHS	3	3
TAC	3	3
TOF	1	1
VSD	7	8
ASD	2	4
DCM	2	1
Ebstein anomaly	1	1
Peripartal asphyxia†	5	5
Acute dehydration†	4	4

TGA indicates transposition of the great arteries; TrA, tricuspid atresia; HLHS, hypoplastic left heart syndrome; TAC, truncus arteriosus; TOF, tetralogy of Fallot; VSD, ventricular septal defect; ASD, atrial septal defect; and DCM, dilated cardiomyopathy.

*Median and range values.

†Absolute numbers of patients or control subjects.

‡Hemi-Fontan correction was performed in 2 patients and 3 control subjects; operation according to Fontan in a further 2 patients and 2 children of the control group.

Laboratory Methods

Blood Sampling

With informed parental consent, blood samples were collected by peripheral venipuncture into 3.8% trisodium citrate (1 part anticoagulant, 9 parts blood; Sarstedt tubes) and placed immediately on melting ice. Platelet-poor plasma was prepared by centrifugation at 3000*g* for 20 minutes at 4°C, divided into aliquots in polystyrene tubes, stored at −70°C, and thawed immediately before the assay. For gene analysis, we obtained venous blood in EDTA-treated S-Monovettes (Sarstedt), from which cells were separated by centrifugation at 3000*g* for 15 minutes. The buffy-coat layer was then removed and stored at −70°C until DNA extraction was performed by standard techniques.

Measurement of Lipids, Lipoproteins, and Lp(a)

Total cholesterol, triglycerides, and HDL cholesterol were measured with enzymatic assays and (for HDL cholesterol) a precipitation method from Boehringer Mannheim on a Hitachi autoanalyzer. LDL cholesterol was calculated with the Friedewald formula.[27] Lp(a) plasma concentrations were measured with an ELISA technique

using mouse monoclonal anti-apo(a) capture antibodies and sheep polyclonal anti-apoB detection antibodies [COALIZA Lp(a), Chromogenix]. Standards and controls were purchased from Immuno. Intra-assay/interassay coefficients of variation are 4.5%/3.6% at 6 mg/dL and 7.0%/4.2% at 40 mg/dL. The lower detection limit of this assay was 0.001 mg/dL.

Determination of Apo(a) Size Polymorphism

In a randomly selected subgroup of 51 patients and 46 control subjects with detectable Lp(a) levels on immunoassay, we determined the apo(a) size polymorphism as a phenotype by agarose gel electrophoresis of plasma and subsequent anti-apo(a) immunoblotting, principally as described by Marcovina et al.[28] After electrophoresis and electrotransfer onto nitrocellulose membranes, apo(a) isoforms were immunodetected by successive incubation of the nitrocellulose membranes with a polyclonal rabbit anti-apo(a) antiserum (Behringwerke), biotinylated donkey antibodies against rabbit IgG (Amersham), streptavidin-conjugated horseradish peroxidase (Amersham), and a chemiluminescence blotting substrate (Boehringer Mannheim). All incubations were performed as recommended by the supplier of the chemiluminescence blotting substrate (Boehringer Mannheim). The chemiluminescent immunoreaction was visualized by photoimaging with the BAS1500 (Fuji, Photo Film). The number of kringle IV repeats of the apo(a) isoforms was estimated by their relative electrophoretic mobility in comparison with standard apo(a) isoforms, which were purchased from Immuno, and by means of the TINA program (Raytest), which supports the BAS 1500 photoimager. In most cases, only 1 of 2 possible isoforms was immunodetectable, either because of true homozygosity or because of the presence of undetectable null alleles, which are the most frequent ones in white populations.[14,16] In a few cases in which 2 isoforms were immunodetectable, we considered only the smaller one for statistical evaluation.

Assays of Hemostatic Factors

The FV:Q[506] mutation, amidolytic protein C activity, and antithrombin activity (in both cases, chromogenic substrates from Chromogenix), free protein S antigen, total protein S antigen, protein C antigen (Asserachrom, Stago), and anticardiolipin IgM and IgG antibodies (ELISA, Chromogenix) were measured as described previously.[29]

For all quantitative plasma-based assays, a prothrombotic defect was diagnosed only when the plasma level of a protein was outside the limits of its normal range in at least 2 independent samples. A heterozygous type I deficiency state was diagnosed when functional plasma activity and immunological antigen concentration of a protein were <50% of normal of the lower age-related limit. A homozygous state was defined by activity levels and antigen concentrations <10% of normal.

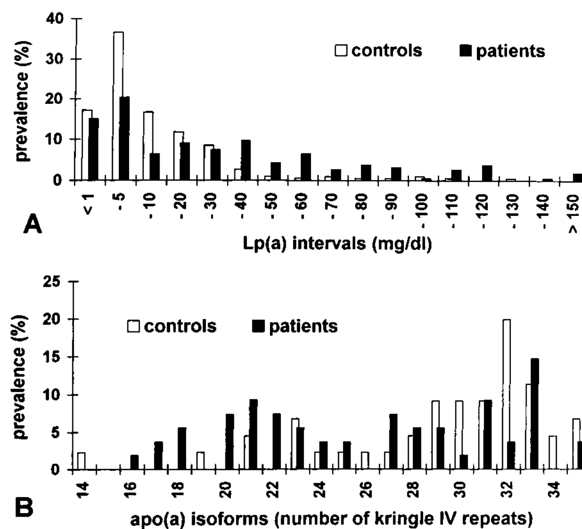
Statistical Analysis

An exploratory analysis was performed using the Statistical Package for the Social Sciences (SPSS-X).[30] Mean values and SDs were compared by Student's t test. Because of their non-Gaussian frequency distributions, data on Lp(a) and triglycerides are presented as medians and were evaluated statistically after logarithmic transformation. Correlations between apo(a) size and Lp(a) levels were calculated by the Spearman rank test. P values <0.05 were considered significant. Multivariate logistic procedure (logistic regression: maximum likelihood ratio) was applied to identify independent risk factors for venous thrombosis. Variables tested for inclusion in the model were Lp(a), FV:Q[506], protein C, protein S, and antithrombin. The level of significance for differences in prevalences was calculated by the Wald χ^2 test.

Results

Lipids, Lipoproteins, and Lp(a)

Children (n=186) who had suffered venous thrombosis differed significantly from 186 age- and disease-matched control subjects by a higher median/range of Lp(a) (19/0 to 170 mg/dL versus 4.4/0 to 125 mg/dL) but not by plasma levels of cholesterol, triglycerides, HDL cholesterol, and LDL cholesterol. As known from many studies in white populations,[6–12] the frequency distribution of Lp(a) levels was skewed to lower concentrations in both patients and control subjects (Figure, panel A). However, very high levels of Lp(a) were much more frequent in patients than in control subjects: After stratification for quartiles of Lp(a), the risk for thromboembolic events did not differ within the 3 lowest quartiles, in which Lp(a) levels were ≤30 mg/dL, but was increased by a factor of 7.7 in the highest quartile compared with the lowest quartile as reference group (Table 2). Lp(a) levels >30 mg/dL, which is also the commonly accepted cardiovascular risk,[6] occurred with a prevalence of 42.0% among patients compared with a prevalence of only 10.3% among control subjects (odds ratio 7.2, with a 95% CI of 3.7 to 14.5) (Table 3).



Frequency distribution of Lp(a) levels among 186 children with venous thromboembolic diseases (ie, patients) and 186 control subjects (controls) (A) as well as frequency distribution of apo(a) isoforms among a randomly selected subgroup of 51 patients and 46 controls (B). Medians (ranges) of Lp(a) levels were 19.0 (1 to 170) and 4.4 (0 to 125) among patients and controls, respectively ($P<0.001$). Mean values and SDs for estimated number of kringle IV repeats were 29±4 and 25±6, respectively ($P<0.01$).

Subgroup Analysis of Lp(a)

Results of a subgroup analysis of Lp(a) in 74 patients without underlying diseases and 74 healthy matched control subjects did not differ significantly from results obtained from the entire study population: Children with spontaneous thrombosis differed significantly from 74 age-and sex-matched healthy control subjects by a higher median/range of Lp(a) (17/0 to 145 mg/dL versus 4/0 to 80 mg/dL; $P=0.001$). The odds ratio/95% CI of Lp(a) >30 mg/dL was 7.1/2.7 to 18.6.

Apo(a) Size Polymorphism

Lp(a) levels are determined largely by variation of the apo(a) gene and are little influenced by environmental factors.[14,16] Nevertheless, to rule out the possibility of high Lp(a) levels in patients being secondary to the thromboembolic events or treatment modalities, we determined the apo(a) size poly-

TABLE 2. Odds Ratio of Thromboembolic Diseases in Children According to Quartiles of Lp(a) Levels and Quartiles of Estimated Numbers of Apo(a) Kringle IV Repeats

	Quartile			
	1	2	3	4
Lp(a), mg/dL	0–2	3–7	7–30	>30
Odds ratios according to quartiles of Lp(a) (95% CIs)	1 (Reference)	0.78 (0.43–1.43)	1.07 (0.60–1.88)	7.68* (3.90–15.11)
No. of kringle IV repeats	14–22	23–28	29–31	32–35
Odds ratios according to quartiles of apo(a) isoforms (95% CIs)	8.21* (2.22–30.38)	2.69 (0.88–8.23)	1.30 (0.42–4.05)	1

Quartiles of Lp(a) levels were defined in 186 children with thromboembolic events (patients) and 186 age- and disease-matched control subjects; quartiles of kringle IV repeats were defined in a randomly selected subgroup of 51 patients and 46 control subjects. Odds ratios presented here indicate the risk multiplicator associated with Lp(a) levels of the lowest quartile as reference group and the number of kringle IV repeats of the highest quartile, respectively.
*$P<0.001$ (χ^2 test).

morphism in the randomly selected subgroup of 51 patients and 46 control subjects with immunodetectable Lp(a) levels. As expected, the estimated number of kringle IV repeats was negatively correlated with Lp(a) levels in both patients ($r=-0.775$; $P<0.001$) and control subjects ($r=-0.360$; $P=0.0169$). Small isoforms were more frequently present in the plasma of patients with thromboembolic events than in that of control subjects (Figure, panel B). Compared with the highest quartile as reference group of the number of kringle IV repeats, the lowest quartile was associated with an 8.2-fold increased risk for thromboembolic events (Table 2).

Established Prothrombotic Risk Factors

Lp(a) >30 mg/dL, the FV:Q^{506} mutation, protein C, protein S, and antithrombin were analyzed by a multivariate logistic procedure to determine their independent contributions to the risk of venous thrombosis. In addition to Lp(a), the FV:Q^{506} mutation, protein C, and antithrombin were found to be independent risk factors of childhood venous vascular occlusion (Table 3). In contrast, protein S deficiency was not different between patients and control subjects. None of the patients or control subjects had elevated titers of IgM or IgG anticardiolipin antibodies.

Increased Lp(a) in Combination With Established Prothrombotic Risk Factors

Based on results obtained from multivariate analysis that Lp(a) >30 mg/dL, the heterozygous FV:Q^{506} mutation, protein C, and antithrombin deficiency were independent risk factors of venous thrombosis in childhood, an intensification of risks in the presence of >1 factor was observed: In 1.6% of the control subjects but in 12.2% of the patients, Lp(a) levels >30 mg/dL were coincident with ≥1 of the above-mentioned genetic hemostatic disorders (OR/CI, 8.4/2.5 to 28.5). Increased Lp(a) was most frequently combined with the heterozygous FV:Q^{506} mutation, which was found in 1.6% of control subjects compared with 9.6% of patients.

TABLE 3. Prevalence of Risk Factors in 186 Children With Thromboembolic Events (Patients) and 186 Age- and Disease-Matched Control Subjects (Logistic Regression Model)

	Prevalence, Control Subjects, %	Prevalence, Patients, %	Odds Ratio (95% CI)
Lp(a) >30 mg/dL	10.3	42.0	7.2 (3.7–14.5)
Presence of FV:Q^{506}	10.9	24.6	2.8 (1.6–4.9)
Protein C deficiency	2.2	8.5	6.5 (2.1–19)
Protein S deficiency	1.1	1.6	2.2 (0.3–14.7)
Antithrombin deficiency	0.5	3.7	10.4 (1.2–90)

Discussion

In the multicenter study presented here, elevated Lp(a) serum concentrations, clearly correlated with the presence of small apo(a) isoforms, were identified as risk factors for the occurrence of venous thromboembolic events in childhood. Lp(a) levels located in the upper quartile of this white population exceeded 30 mg/dL and increased the risk of a thromboembolic event in childhood by factor of 7.2. This odds ratio is higher than that found in children heterozygous for the FV:Q^{506} mutation, which in the white population has been considered the most important single risk factor for thrombotic events and which was shown to have a prevalence similar to that of elevated Lp(a).[2]

In vitro, Lp(a) inhibits the activation of plasminogen by streptokinase and tissue plasminogen activator (tPA) and competes with plasminogen for binding to fibrin as well as for binding to annexin II, the plasminogen/tPA receptor on endothelial cells and platelets.[15,17–19,31–34] Because of these properties and the great structural homology between Lp(a) and plasminogen, it has been hypothesized that Lp(a) inhibits fibrinolysis. In fact, apo(a) transgenic animals were resistant to tPA-dependent lysis of artificially induced fibrin thrombi.[18] These antifibrinolytic properties of Lp(a) have been made responsible in part for the association of elevated Lp(a) and risk for atherosclerotic vessel diseases.[6–15,17–19] Surprisingly, much less attention has been paid to the role of elevated Lp(a) as a risk factor for venous thromboembolic diseases. Some small-scale studies and single case reports have indicated that elevated Lp(a) levels may further increase the risk of thrombotic events in patients suffering from an acquired prothrombotic state due to rheumatic diseases, polycythemia vera, or nephrotic syndrome.[20–25] Studies in unselected adult patients

with venous thrombosis or embolism as well as in women practicing hormonal contraception revealed no association between Lp(a) and thromboembolic diseases.[35,36]

With an annual incidence of 0.07/10 000, thromboembolic disease in childhood is rare and probably develops through concomitant hemodynamic disturbances in the cardiovascular system and hereditary disturbances of the hemostatic system.[37,38] In this regard, children with thromboembolic events represent a high-risk population similar to patients with rheumatic diseases, nephrotic syndrome, or polycythemia vera. Interestingly, elevated Lp(a) has previously evolved as a prothrombotic risk factor in these selected high-risk cohorts[20–25] but not in unselected cohorts with sporadic thromboembolic events.[35] We therefore hypothesize that Lp(a) acts as an additional important risk factor for the precipitation of venous thromboembolic events in the white population studied here. Furthermore, on the basis of results obtained from multivariate analysis that Lp(a) >30 mg/dL, the heterozygous FV:Q^{506} mutation, protein C, and antithrombin deficiency were independent risk factors of venous thrombosis in childhood, an intensification of risks in the presence of >1 factor was observed. Although the findings presented here in a white childhood thrombosis population are not generalizable to other racial and ethnic groups, in particular Americans of African origin, measurement of Lp(a) should be included in screening programs performed in young patients suffering from venous thromboembolism, in addition to FV:Q^{506}, protein C, protein S, and antithrombin. Currently, no specific Lp(a)-lowering therapy is available. However, elevated Lp(a) levels may be an indication for the initiation of anticoagulant therapy, just as other prothrombotic factors currently serve as an indication for this treatment. Further studies are necessary to determine whether anticoagulant treatment reduces the risk of future events in these high-risk patients.

Appendix

Coinvestigators were as follows (alphabetical order within different institutions): A. Heinecke (Institute of Medical Statistics, University Hospital Münster, Germany); H. Pollmann, H. Vielhaber (University Children's Hospital Münster, Germany); K. Auberger (University Children's Hospital Munich, Germany); A. Claviez, R. Schneppenheim (University Children's Hospital Kiel, Germany); U. Göbel, C. Mauz-Körholz (University Children's Hospital Düsseldorf, Germany); C. Wermes (University Children's Hospital Hannover, Germany); and B. Zieger (Scripps Research Institute, La Jolla, Calif).

Acknowledgments

Dr Nowak-Göttl was supported by a grant from Deutsche Krebshilfe, and Dr von Eckardstein by a grant from the Interdisziplinäres Zentrum für Klinische Forschung at the Medical Faculty of the Westphalian Wilhelms-University (Project A3). The authors thank the technicians from the participating laboratories, in particular Christiane Schettler, Bertram Tambyrajah, and Doris Weber, for excellent technical assistance. We also gratefully acknowledge Susan Griesbach for editing the manuscript.

References

1. Stefano de V, Finazzi G, Mannucci PM. Inherited thrombophilia: pathogenesis, clinical syndromes and management. *Blood*. 1996;87:3531–3544.
2. Lane DA, Mannucci PM, Bauer KA, Bertina RM, Bochkov NP, Boulyjenkov V, Chandy M, Dahlbäck B, Ginter EK, Miletich JP, Rosendaal FR, Seligsohn U. Inherited thrombophilia, I. *Thromb Haemost*. 1996;76:651–662.
3. Lane DA, Mannucci PM, Bauer KA, Bertina RM, Bochkov NP, Boulyjenkov V, Boulyjenkov V, Chandy M, Dahlbäck B, Ginter EK, Miletich JP, Rosendaal FR, Seligsohn U. Inherited thrombophilia, II. *Thromb Haemost*. 1996;76:824–834.
4. Poort SR, Rosendaal FR, Reitsma PH, Bertina RM. A common genetic variation in the 3′-untranslated region of the prothrombin gene is associated with elevated plasma prothrombin levels and an increase in venous thrombosis. *Blood*. 1996;88:3698–3703.
5. Berg K, Dahlen G, Frick MH. Lp (a) lipoprotein and pre-beta 1 lipoprotein in patients with coronary heart disease. *Clin Genet*. 1974;6:230–235.
6. Rader DJ, Hoeg JM, Brewer HB Jr. Quantification of plasma apolipoproteins in the primary and secondary prevention of coronary artery disease. *Ann Intern Med*. 1994;120:1012–1015.
7. Maher VMG, Brown BG. Lipoprotein(a) and coronary heart disease. *Curr Opin Lipidol*. 1995;6:229–235.
8. Rosengren A, Wilhelmsen L, Eriksson E, Risberg B, Wedel H. Lipoprotein(a) and coronary heart disease: a prospective case control study in a general population of middle-aged men. *Br Med J*. 1990;301:1248–1251.
9. Schaefer EJ, Lamon-Fava S, Jenner JL, McNamara JR, Ordovas JM, Davis CE, Abolafia JM, Lippel K, Levy RI. Lipoprotein(a) levels and risk of coronary heart disease in men: the Lipid Research Clinics Primary Prevention Trial. *JAMA*. 1994;271:999–1003.
10. Wald NJ, Law M, Watt HC, Wu T, Bailey A, Johnson AM, Craig WY, Ledue TB, Haddow JE. Apolipoproteins and ischaemic heart disease: implications for screening. *Lancet*. 1994;343:75–79.
11. Assmann G, Schulte H, von Eckardstein A. Hypertriglyceridemia and elevated lipoprotein(a) are risk factors for major coronary events in middle-aged men. *Am J Cardiol*. 1996;77:1179–1184.
12. Cremer P, Nagel D, Labrot B, Mann H, Muche R, Elster H, Seidel D. Lipoprotein Lp(a) as a predictor of myocardial infarction in comparison to fibrinogen, LDL cholesterol and other risk factors: results from the Göttingen Risk Incidence and Prevalence Study (GRIPS). *Eur J Clin Invest*. 1994;24:444–453.
13. Nagayama M, Shinohara Y, Nagayama T. Lipoprotein (a) and ischaemic cerebrovascular disease in young adults. *Stroke*. 1994;25:74–78.
14. Utermann G. The mysteries of lipoprotein(a). *Science*. 1989;246:904–910.
15. Haijjar KA, Nachman RL. The role of lipoprotein(a) in atherogenesis and thrombosis. *Annu Rev Med*. 1996;47:423–442.
16. Boerwinkle E, Leffert CC, Lin J, Lackner C, Chiesa G, Hobbs HH. Apolipoprotein(a) gene accounts for greater than 90% of the variation in plasma lipoprotein(a) concentrations. *J Clin Invest*. 1992;90:52–60.
17. Hajjar KA, Gavish D, Breslow JL, Nachman RL. Lipoprotein(a) modulation of endothelial cell surface fibrinolysis and its potential role in atherosclerosis. *Nature*. 1989;339:303–305.
18. Palabrica TM, Liu AC, Aronovitz MJ, Furie B, Lawn RM, Furie BC. Antifibrinolytic activity of apolipoprotein(a) in vivo: human apolipoprotein(a) transgenic mice are resistant to tissue plasminogen activator-mediated thrombolysis. *Nat Med*. 1995;1:256–259.
19. Loscalzo J, Weinfeld M, Fless GM, Scanu AM. Lipoprotein(a), fibrin binding and plasminogen activation. *Arteriosclerosis*. 1990;10:240–245.
20. Seriolo B, Accardo S, Fasciolo D, Bertolini S, Cutolo M. Lipoproteins, anticardiolipin antibodies and thrombotic events in rheumatoid arthritis. *Clin Exp Rheumatol*. 1996;14:593–599.
21. Seriolo B, Accardo S, Cutolo M. Lipoprotein(a) and anticardiolipin antibodies as risk factors for thrombotic events in polymyalgia rheumatica and giant cell arteritis. *J Rheumatol*. 1996;23:1478–1479.
22. Örem A, Deger O, Memis Ö, Bahadir S, Ovali E, Cimsit G. Lp(a) lipoprotein levels as a predictor of risk for thrombogenic events in patients with Behcet's disease. *Ann Rheum Dis*. 1995;54:726–729.
23. Levy PJ, Cooper CF, Gonzalez MF. Massive lower extremity arterial thrombosis and acute hepatic insufficiency in a young adult with premature atherosclerosis associated with hyperlipoprotein(a)emia and antiphospholipid syndrome. *Angiology*. 1995;46:853–857.
24. LeBlanc K, Samuelsson J. Elevated plasma lipoprotein(a): a potential risk factor for thromboembolic complications in polycythaemia vera. *Eur J Haematol*. 1996;57:257–258.

25. Fujita T, Saito E, Ohi H, Yasugi T, Hatano M. Lipoprotein(a) predicts the risk of thrombogenic complications in nephrotic syndrome. *Nephron*. 1992;61:122. Letter.
26. Nowak-Göttl U, Debus O, Findeisen M, Kassenböhmer R, Koch HG, Pollmann H, Postler CH, Weber P, Vielhaber H. Lipoprotein (a): its role in childhood thromboembolism. *Pediatrics*. 1997;99:e6–e11.
27. Friedewald WT, Levy RI, Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clin Chem*. 1972;18:499–502.
28. Marcovina SM, Hobbs HH, Albers JJ. Relation between number of apolipoprotein (a) kringle 4 repeats and mobility of isoforms in agarose gel: basis for a standardized isoform nomenclature. *Clin Chem*. 1996;42:436–439.
29. Nowak-Göttl U, Koch HG, Aschka I, Kohlhase B, Vielhaber H, Kurlemann G, Olescuk-Raschke K, Kehl HG, Jürgens H. Resistance to activated protein C (APCR) in children with venous or arterial thromboembolism. *Br J Haematol*. 1996;92:992–996.
30. Nie NH. *SPSSX User's Guide*. New York, NY: McGraw-Hill; 1983.
31. Harpel PC, Gordon BR, Parker TS. Plasmin catalyses binding of lipoprotein(a) to immobilized fibrinogen and fibrin. *Proc Natl Acad Sci U S A*. 1989;86:3847–3851.
32. Edelberg JM, Gonzalez-Gronow M, Pizzo SV. Lipoprotein(a) inhibits streptokinase- mediated activation of human plasminogen. *Biochemistry*. 1989;28:2370–2374.
33. Simon DI, Fless GM, Scanu AM, Loscalzo J. Tissue-type plasminogen activator binds to and is inhibited by surface-bound lipoprotein (a) and low density lipoprotein. *Biochemistry*. 1991;30:6671–6677.
34. Ezratty A, Simon DI, Loscalzo J. Lipoprotein(a) binds to human platelets and attenuates plasminogen binding and activation. *Biochemistry*. 1993; 32:4628–4633.
35. März W, Trommlitz M, Scharrer I, Gross W. Apolipoprotein(a) concentrations are not related to the risk of venous thrombosis. *Blood Coagul Fibrinolysis*. 1991;2:595–599.
36. Delplanque B, Beaumont V, Lemort N, Beaumont JL. Lp(a) levels and antiestrogen antibodies in women with and without thrombosis in the course of oral contraception. *Atherosclerosis*. 1993;100:183–188.
37. Andrew M. Developmental hemostasis: relevance to thromboembolic complications in pediatric patients. *Thromb Haemost*. 1995;74:415–425.
38. Grotta J. Cerebrovascular disease in young patients. *Thromb Haemost*. 1997;78:13–23.