



Lipoprotein(a) concentration is not associated with venous thromboembolism in a case control study

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ABSTRACT

Background and Objective. Lipoprotein(a) is an LDL-like particle displaying strong athero-thrombotic properties. Although Lp(a) plays a pivotal role in the genesis and progression of thrombosis in the arterial district, its role in promoting thrombosis in the venous district is still unclear.

Design and Methods. To give further insight into the thrombotic potential of Lp(a), 100 potentially eligible consecutive outpatients who had suffered from previous episodes of venous thrombosis (deep vein thrombosis with or without pulmonary embolism) were enrolled into the study. Thirty-six of these patients who did not fulfil the entry criteria were then excluded from the study. The concentration of Lp(a) was thus measured in 64 patients, and compared to that of 64 control subjects, matched for sex ($p=0.46$), age ($p=0.25$) and pharmacological treatment; no subject belonging to the control group had a familial or personal history of venous thromboembolism. Exclusion criteria for both groups included: diabetes mellitus, liver or kidney diseases and malignancy, as established by both laboratory analysis and physical examination. To rule out false elevations of Lp(a) due to the presence of a concurrent acute phase response, C reactive protein (CRP) was measured in both groups using a commercial immunonephelometric assay.

Results. No statistically significant differences were observed in the median Lp(a) concentration between patients and controls (median: 69 vs 83 mg/L, respectively; $p=0.34$). Neither were any significant differences found between patients who had suffered from deep venous thrombosis with ($n=18$) or without ($n=46$) pulmonary embolism (median: 73 vs 69 mg/L, respectively; $p=0.83$). The concentration of CRP did not differ significantly between cases and controls (median: 1.8 vs 2.3 g/L, respectively; $p=0.37$).

Interpretation and Conclusions. Although there are several plausible biological mechanisms to explain the strong thrombogenicity of Lp(a) *in vitro*, we failed

to demonstrate a convincing association between Lp(a) and thrombosis in the venous district. Besides the proven prothrombotic role of Lp(a) in some selected clinical settings, it is thus conceivable that the contribution of Lp(a) to genesis and progression of the venous thrombosis might be marginal or efficiently counterbalanced *in vivo*. The clinical usefulness of including the measurement of Lp(a) among the screening tests for thrombophilic patients, therefore, remains questionable.

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Key words: lipoprotein(a); venous thromboembolism, thrombosis, atherosclerosis, risk factor

Venous thromboembolism (VTE) is a major cause of morbidity and mortality in Western Countries. The main clinical manifestations are represented by deep venous thrombosis (DVT) and pulmonary embolism (PE).¹ Venous thrombosis is a multifactorial disease and usually occurs as a result of an imbalance between prothrombotic and anti-thrombotic potentials, a defective function of the fibrinolytic system or both.^{2,3} Several biochemical and environmental risk factors have been associated with the occurrence of thromboembolic episodes over the past decades; among these, some biochemical alterations leading to a prothrombotic status, such as immunologic or functional deficiencies of the physiological anticoagulant systems, factor II and VIII excesses, hyperhomocysteinemia, anti-phospholipid antibodies and others have been well established as risk factors for VTE.²⁻⁶ Conversely, the contribution of acquired or inherited conditions leading to impaired fibrinolysis is still debated. Plasminogen abnormalities, reduced levels of tissue-type plasminogen activator (t-PA) and increased levels of the inhibitor of plasminogen activator-1 (PAI-1) have occasionally been associated with VTE, but so far these conditions have not satisfactorily fulfilled the criteria required to establish a definitive casual relationship.^{4,7}

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Among the leading causes of impaired fibrinolysis, lipoprotein(a) [Lp(a)] plays a pivotal role.⁸ Lp(a) is a genetic variant of a low density lipoprotein (LDL) composed by the association of a unique and highly glycosylated apolipoprotein(a) [apo(a)] with apolipoprotein B100 through a single disulphide bond.⁹ Due to the structural mimicry with plasminogen, apo(a) inhibits plasminogen binding and activation at the surface of stabilized fibrin, endothelial cells and platelets in a dose-dependent fashion.⁸ Additional studies demonstrated that Lp(a) increases the endothelial synthesis and secretion of PAI-1 *in vitro* by more than 2-fold, inhibits the secretion of t-PA from human endothelial cells and binds reversibly to t-PA inhibiting the tPA-mediated activation of Glu-plasminogen.⁸ Although the contribution of Lp(a) to genesis, progression and complications of atherosclerotic lesions is fairly certain, no definitive data are available on the role of Lp(a) in the genesis of thrombosis in the venous district. In this perspective, the present study was designed to verify whether raised concentrations of Lp(a), the most frequent lipoprotein abnormality observed in patients with premature myocardial infarction, might also be a significant risk factor in patients with VTE.

Design and Methods

Population study

The study originally consisted of 100 potentially eligible consecutive patients. Thirty-six patients did not fulfil the entry criteria and so were excluded from the study. The final study group therefore consisted of 64 patients (27 women and 37 men, mean age 59 ± 15 years) with prior episodes of venous thromboembolism who were not taking any medication known to influence the metabolism of Lp(a). Deep vein thrombosis was diagnosed by ascending venography or Doppler ultrasonography; pulmonary embolism was always confirmed by pulmonary angiography. The control group consisted of 64 subjects matched for sex ($p=0.46$), age ($p=0.25$) and pharmacological treatment; none had a familial or personal history of VTE. Exclusion criteria for both groups included: diabetes mellitus, liver or kidney diseases and malignancy as established by both laboratory analysis and physical examination. Besides malignancy, the presence of either congenital or acquired thrombophilic states was not evaluated. All patients gave informed consent to participation in the study.

Laboratory measurements

Blood was collected from patients on admission after an overnight fasting between September 1st and December 5th, 1998. After centrifugation, serum was stored at -80°C until measurement. Lp(a) was measured employing an N Lp(a) latex-enhanced immunonephelometric assay (Dade Behring, Marburg, Germany), on a Behring Nephelometric Analyser (BNA)

(Behring). Final results of the assay were reported in terms of total lipoprotein mass. Intra- and inter-assays CVs were respectively $<4\%$. C reactive protein was measured by an immunonephelometric assay (Dade Behring) on a BNA.

Statistics

The statistical analysis was performed using Astute (DDU Software, The University of Leeds, UK). As Lp(a) values do not follow Gaussian distribution, results were expressed in terms of medians and ranges. The Wilcoxon Mann-Whitney U test, a non-parametric alternative to the Student's t-test, was employed for the comparison of Lp(a) values between groups. Probability values of <0.05 were considered statistically significant. The concentration of CRP between groups was compared by Student's t-test.

Results

No statistically significant differences were observed in the median of Lp(a) concentrations between patients with previous episodes of VTE (median: 69 mg/L, range: 2-769 mg/L) and matched controls (median: 83 mg/L, range: 2-936 mg/L; $p=0.34$), nor was there any evidence of a relevant trend towards higher levels of Lp(a) among groups (Figure 1). Nor were any significant difference found between patients who had suffered from deep venous thrombosis with ($n=18$) or without ($n=46$) pulmonary embolism (73 mg/L vs 69 mg/L, respectively; $p=0.83$). Possible interference from the acute phase response could be excluded as the concentration of CRP did not differ significantly between cases and controls (1.8 vs 2.3 g/L, respectively; $p=0.37$).

Discussion

Lp(a) is an atherogenic lipoprotein particle which displays adjunctive thrombotic properties.⁹ It is now well established that Lp(a) exerts its thrombotic potential through inhibition of the fibrinolytic pathway; no prothrombotic activity has been yet ascribed to Lp(a).¹⁰ Several clinical observations confirmed *in vivo* the antifibrinolytic potential of Lp(a) *in vitro*. Raised Lp(a) concentrations were demonstrated in patients with central retinal artery occlusion¹¹ and interference in placental circulation causing fetal growth retardation.¹² Lp(a) levels are strong predictors of occlusive events following both vascular and endovascular surgical procedures¹³ and, finally, the confluence of the atherogenic and thrombotic potential of Lp(a) plays a pivotal role in the occurrence of thrombotic complications resulting from rupture or ulceration of atherosclerotic plaques.⁸ Although there is considerable consolidated evidence that confirms the central role of Lp(a) in the genesis of arterial thrombosis, its role in promoting thrombosis in the venous district is still unclear and results of previous investigations are rather contradictory. Increased lev-

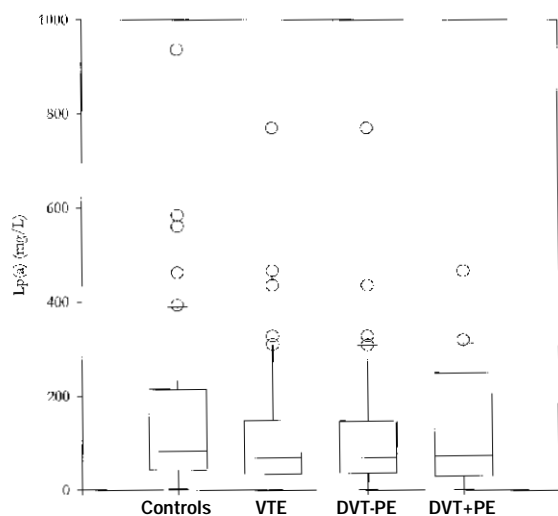


Figure 1. Lp(a) concentration (medians, 10th-90th and 5th-95th percentile distributions) of all patients with venous thromboembolism (VTE), patients with deep vein thrombosis with (DVT+PE) or without (DVT-PE) pulmonary embolism and matched controls.

els of Lp(a) were observed in patients with pulmonary embolism,¹⁴ chronic thromboembolic pulmonary hypertension,¹⁵ and were associated with a pattern of changes in the levels of D-dimer, plasminogen and PAI-1 suggestive of a relative impairment of the fibrinolytic process.¹⁶ Moreover, Lp(a) levels were good predictors of thromboembolic complications in patients with rheumatologic and malignant disorders.¹⁷⁻¹⁹ Conversely, Murata *et al.* did not find statistical differences in Lp(a) levels between patients with central retinal vein occlusion and controls²⁰ and, in a subsequent investigation, no relevant association was observed between apo(a) concentrations and the risk of venous thrombosis in young subjects.²¹ Finally, a recent study did not show convincing evidence of an association between raised concentrations of Lp(a) and childhood thromboembolism.²² These latter observations are consistent with results of our investigation. As shown in Figure 1, we observed no statistically significant differences in the distribution of Lp(a) concentration between controls and patients with earlier episodes of VTE. Additionally, no major differences were found between DVT patients with or without PE. Thus, although Lp(a) can be considered a consolidated risk factor for arterial thrombosis, we suggest that its role in the genesis of venous thrombosis might be marginal or efficiently counterbalanced *in vivo*. This observation is not really surprising as the pathogenesis of venous and arterial thromboses is different and some proven risk factors might be more relevant for venous than for arterial thrombosis and *vice versa*.^{3, 23}

The apparent dissociation between the antifibri-

olytic potential of Lp(a) in the genesis of venous and arterial thromboses has some possible explanations. Differently than for arterial thrombosis, earlier studies failed to show convincing evidence that a link exists between impaired fibrinolysis and increased risk of VTE. Hence, although impaired fibrinolysis contributes to the complications of arterial occlusive diseases,^{8, 24} venous thrombosis remains a disorder mainly associated with an imbalance between prothrombotic and antithrombotic potential.^{6,7} Indeed, the composition of mural thrombi in complicated atherosclerotic lesions is different from that of thrombi in the venous system, confirming that, although the hypercoagulable or thrombophilic state might be considered as a single clinical entity, the relative contribution of some underlying pathogenic mechanisms might vary between the venous and arterial districts.² Finally, given the extensive evidence of preferential accumulation and marked concentration of Lp(a) in atherosclerotic lesions as compared to in uninjured vessel wall,²⁵ the inhibitory effect of apo(a) on fibrinolysis might be greatly enhanced in the arterial district.

In conclusion, our results do not support a hypothesis of a causal relationship between Lp(a) and VTE; the clinical usefulness of including the measurement of Lp(a) among the screening tests for thrombophilic patients, therefore, remains uncertain and further in-depth investigations exploring the association between raised concentration of Lp(a) and venous thrombosis are needed.

Contributions and Acknowledgments

GL: conception and design of the study; collection, classification and storage of the samples, medical interview and physical examination of the patients, supervision of whole laboratory testing, interpretation of the data, draft of the article and final approval of the version. AB: design of part of the practical organisation of study, collection of the samples, Lp(a) and CRP analysis of part of the samples, interpretation of the data, revision of the article for important intellectual content and final approval of the version. GB and FM: design of part of the practical organization of the study, collection of the samples, medical interview and physical examination of the patients, participation in laboratory testing, interpretation of the data, revision of the article for important intellectual content and final approval of the version. MM: Lp(a) and CRP analysis of part of the samples, revision of the article for important intellectual content and final approval of the version. GG: conception and design of the study, interpretation of the data, draft and final revision of the article and final approval of the version.

The criteria followed to assign the order of the names of the Authors are basically those already expressed in the qualification for authorship.

Disclosures

Conflict of interest: none.

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Manuscript processing

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