SCIENTIFIC CORRESPONDENCE

Lupus anticoagulant positivity in insulin dependent diabetic patients: an additional risk factor in the pathogenesis of diabetic retinopathy?

Cristiano Giusti, Riccardo Schiaffini, Daniela Bosco, Paolo Ciampalini, Antonio Pantaleo, Enzo M Vingolo, Patrizia Gargiulo

Abstract

Aims—To investigate whether lupus anticoagulant (LA) positivity, a marker of endothelial dysfunction, might be relevant to the pathogenesis of diabetic retinopathy (DR).

Methods—32 IDDM patients were examined for LA, fibrinogen, prothrombin (PT), PTT, prothrombin degradation products (F1+2), and activated protein C (APC).

Results—APC decreased and F1+2 increased significantly in LA positive but not in LA negative patients; 60% of LA positive and 18% of LA negative subjects had DR. PT, PTT, and fibrinogen levels were insignificant.

Conclusion—These preliminary findings suggest that LA positivity could represent an additional risk factor for DR, acting as a link between the immunological and haemostatic systems.

(Br J Ophthalmol 2000;84:531–533)

The natural history and aetiopathogenesis of diabetic retinopathy (DR) are still not completely understood. Vascular damage and disturbed endothelial function occur early in the course of diabetic microangiopathy converting the endothelium from a thromboresistant to a thrombogenic surface.^{1,2} Moreover, it has been suggested that immunological mechanisms have a role in the pathogenesis of diabetic microangiopathy via immune complex deposition.³ Autoantibodies directed against endothelial antigens could be responsible for initiating vascular injury,⁴ but could also be a marker of endothelial dysfunction.

Lupus anticoagulant (LA), an antiphospholipid antibody, is thought to be involved in this endothelial alteration and is frequently associated with thromboembolic events, miscarriage, systemic lupus erythematosus, diabetic vascular complications (overt nephropathy or macroangiopathy), and retinal occlusive vasculopathies.^{5–9}

Because of the evidence of immune and haemostatic abnormalities in diabetes and the strong correlation observed between LA and similar alterations in other diseases, our study

was designed to determine whether LA positivity was associated with thrombotic tendency and increased prevalence of DR.

Materials and methods

A total of 32 IDDM patients (14 males, 18 females; age range 25–68 years), classified according to the National Diabetes Data Group criteria,¹⁰ with a duration of the disease longer than 5 years, were enrolled. Poor glycaemic control ($HbA_{1c} > 7\%$), borderline hypertension ($> 140/90$), hypertriglyceridaemia (> 1.9 mmol/l), or hypercholesterolaemia (> 5.6 mmol/l) were exclusion criteria. Fifty healthy, non-diabetic subjects aged between 20 and 50 years, served as a control group.

All diabetics were treated with subcutaneous human insulin (regular and long acting) only. Both diabetics and controls were non-smokers and had not taken drugs which could affect haemostasis for at least 4 weeks before the study.

Plasma samples were examined for LA, fibrinogen, prothrombin (PT), PTT, prothrombin degradation products (F1+2), and activated protein C (APC). DR was classified according to the ETDRS-Airlie House Classification.¹¹

Informed consent was obtained from all subjects after full explanation of the nature of the study. This investigation was approved by the bioethics committee of the University of Rome “La Sapienza”.

Data were expressed as mean (SD) and statistical analyses were performed using the Mann–Whitney U test. Bonferroni’s correction was applied to control for the increase of type 1 error probability due to multiple comparison. A p value of less than 0.05 was considered significant.

Results

Table 1 summarises both the clinical features and the laboratory findings of diabetic patients and controls. While the 50 control subjects were all negative for LA, the presence of LA (> 1.31), in contrast, was detected in 10 (four males, six females) of the 32 enrolled diabetics (31.2%). The mean titre in the positive patients was 1.59 (0.03) (v 0.98 (0.001) in the

University “La Sapienza”, Rome, Italy
C Giusti
E M Vingolo

Institute of Internal Medicine II
R Schiaffini
D Bosco
P Ciampalini
A Pantaleo
P Gargiulo

Correspondence to:
Cristiano Giusti, MD, Via
Cassia 1280 (Pal B1, int 10),
I-00189 Rome, Italy
crigiust@tin.it

Accepted for publication
13 January 2000

Table 1 Clinical and biochemical data of diabetic patients and healthy controls (n=mean value (SD); ns= p value not significant)

	LA positive (n = 10)	LA negative (n = 22)	Control group (n = 50)	p Value		
				LA positive v LA negative	LA positive v controls	LA negative v controls
Age (years)	37.9 (13.6)	39.8 (10.3)	37.4 (6.6)	0.4161	0.3023	0.7229
Sex (M, F)	4, 6	10, 12	18, 32	—	—	—
Duration of diabetes (years)	16.7 (11.4)	15.8 (7.9)	—	ns	—	—
Body mass index (kg/m ²)	21.6 (2.08)	22.53 (2.48)	22.9 (2.05)	0.2550	0.0694	0.5613
Systolic blood pressure (mm Hg)	119.0 (11.01)	122.73 (9.73)	127.8 (11.26)	0.2609	0.0198	0.0456
Diastolic blood pressure (mm Hg)	72.5 (6.77)	74.09 (9.08)	72.3 (8.28)	0.6242	0.8007	0.6105
Glycosylated haemoglobin (%)	6.57 (1.43)	6.03 (1.67)	4.54 (0.64)	0.5827	0.0004	0.0022
Total cholesterol (mmol/l)	4.49 (0.71)	4.72 (1.02)	4.99 (0.56)	0.5419	0.0354	0.2100
HDL cholesterol (mmol/l)	1.17 (0.23)	1.18 (0.25)	1.14 (0.17)	0.9838	0.8659	0.5901
Total triglyceride (mmol/l)	1.04 (0.10)	1.12 (0.11)	1.37 (0.23)	0.0294	<0.0001	<0.0001
Creatinine (mmol/l)	79.56 (18.56)	78.67 (20.33)	72.48 (13.26)	0.9350	0.3714	0.4053
No diabetic retinopathy	4 (40%)	18 (82%)	—	<0.001	—	—
Diabetic retinopathy (DR):	6 (60%)	4 (18%)	—	<0.001	—	—
Background DR	2 (33.3%)	2 (50%)	—	ns	—	—
Non-proliferative DR	0	0	—	—	—	—
Preproliferative DR	0	0	—	—	—	—
Proliferative DR	4 (66.6%)	2 (50%)	—	ns	—	—
Nephropathy:	6 (60%)	4 (18%)	—	<0.001	—	—
Microalbuminuria	6 (60%)	4 (18%)	—	<0.001	—	—
Macroalbuminuria	0	0	—	—	—	—

LA negative subjects, $p < 0.001$) (Table 2). LA positive and LA negative patients were similar for clinical, biochemical, and haemodynamic variables (Table 1) but anticardiolipin antibodies were found in 80% of LA positive and in only 9% of LA negative patients ($\chi^2 = 12.959$, $p < 0.01$).

Clinically manifest microangiopathy (retinopathy and/or nephropathy) was found in six of 10 LA positive (60%) and in four of 22 LA negative (18%) diabetics with similar duration of the disease and metabolic control (Table 1). Background and proliferative retinopathy were observed only. In particular, a higher prevalence of proliferative DR was observed in LA positive patients (66.6% *v* 50%), although this difference did not achieve statistical significance, probably because of the small number of subjects. Other diabetic complications (for example, neuropathy, macroangiopathy) were not observed.

There were no statistically significant differences for PT, PTT, and fibrinogen levels between both groups of patients ($p > 0.05$). In contrast, APC levels were reduced ($p < 0.05$) and F1+2 concentrations were increased ($p < 0.05$) in LA positive compared with LA negative cases (Table 2).

Discussion

The clinical association between LA positivity and thrombotic events in other diseases is thought to be due to an LA induced endothelial cell dysfunction.⁵⁻⁶ In IDDM, a similar dysfunction of the coagulant and anticoagulant pathways, as a result of endothelial cell damage, has

been described.¹²⁻¹⁹ In fact, altered coagulation is manifested by enhanced prothrombin conversion to thrombin—as demonstrated by increased prothrombin degradation fragments (F1+2) plasma concentrations²⁰—and down-regulation of the anticoagulant pathway, caused by reduced antithrombin III activity²⁰ and thrombomodulin's endothelial receptors.²¹

In our study, decreased APC concentrations and increased F1+2 plasma levels were observed in LA positive compared with LA negative patients (Table 2), as an evident expression of prothrombotic condition and endothelial dysfunction. APC and F1+2 values in LA negative cases and in healthy controls were similar. The lack of macroangiopathy in LA positive subjects may be due to the chronic nature of the endothelial dysfunction: in fact, comparable PT, PTT, and fibrinogen values were observed in both groups.

In conclusion, this investigation pointed out a significant correlation between LA positivity and: increased prothrombin conversion to thrombin, downregulation of the endothelial anticoagulant pathway, and higher prevalence of DR ($\chi^2 = 7.35$, $p < 0.05$). No correlations were found between LA titre and other specific factors (for example, age, duration of diabetes, HbA_{1c} level, type of DR) ($\chi^2 = 15.2$, $p = 0.83$).

Although not conclusive, these preliminary results suggest that LA positivity might be considered an additional risk factor in the pathogenesis of microangiopathy (retinopathy and/or nephropathy) in IDDM patients, representing an intersection point between immune and haemostatic alterations. Nevertheless, fur-

Table 2 Haemocoagulative variables in diabetic patients and in healthy controls (n=mean value (SD))

	LA positive (n = 10)	LA negative (n = 22)	Control group (n = 50)	p value		
				LA positive v LA negative	LA positive v controls	LA negative v controls
Lupus anticoagulant (titre)	1.59 (0.03)	0.98 (0.001)	—	<0.001	—	—
Activated protein C (mmol/l)	82.0 (17.3)	102.82 (19.9)	95.22 (13.7)	0.0082	0.0168	0.1064
F1+2 (mmol/l)	1.14 (0.37)	0.68 (0.42)	0.80 (0.23)	0.0146	0.0190	0.2002
Fibrinogen (g/l)	2.99 (0.47)	2.89 (0.51)	2.94 (0.46)	0.6694	0.5320	0.6248
PT (ratio)	1.10 (0.40)	1.20 (0.30)	1.10 (0.20)	0.3600	0.6125	0.2399
PTT (ratio)	1.20 (0.30)	1.30 (0.20)	1.20 (0.20)	0.0914	0.5254	0.0622

ther investigations on larger diabetic populations are required in order to clarify and support this issue.

To F Chiarotti, for performing all statistical analyses, and Janet Hill, for her generous help in reviewing the manuscript.

- 1 Haefliger IO, Meyer P, Flammer J, *et al.* The vascular endothelium as a regulator of the ocular circulation: a new concept in ophthalmology? *Surv Ophthalmol* 1994;**39**:123–32.
- 2 Kohner EM, Patel V, Rassman SMB. Role of blood flow and impaired autoregulation in the pathogenesis of diabetic retinopathy. *Diabetes* 1995;**44**:603–7.
- 3 Andreani D. Malignant microangiopathy. *Diabetologia* 1980;**18**:255.
- 4 Jones DB, Wallace R, Frier BM. Vascular endothelial cell antibodies in diabetic patients. Association with diabetic retinopathy. *Diabetes Care* 1992;**15**:552–5.
- 5 Lechner K, Pabinger Fasching I. Lupus anticoagulants and thrombosis. A study of 25 cases and review of the literature. *Haemostasis* 1985;**15**:254–62.
- 6 Triplett DA. Obstetrical complications associated with antiphospholipid antibodies. In: Coulama BC, Faulk WP, McIntyre JA, eds. *Immunological obstetrics*. New York: WW Norton, 1992:377–403.
- 7 Montehermoso A, Cervera R, Font J, *et al.* Association of antiphospholipid antibodies with retinal vascular disease in systemic lupus erythematosus. *Sem Arthritis Rheum* 1999;**28**:326–32.
- 8 Galtier-Dereure F, Biron C, Vies M, *et al.* Vascular complications of diabetes mellitus: what role for phospholipid-binding antibodies? *Lupus* 1998;**7**:469–74.
- 9 Primary antiphospholipid antibody syndrome and retinal occlusive vasculopathy. *Am J Ophthalmol* 1997;**123**:848–50.
- 10 National Diabetes Data Group. Classification of diabetes mellitus and other categories of glucose intolerance. *Diabetes* 1979;**28**:1039–57.
- 11 Diabetic Retinopathy Study Research Group. Report 7. A modification of the Airlie House classification of diabetic retinopathy. *Invest Ophthalmol Vis Sci* 1981;**21**:210–26.
- 12 Sharma SC. Platelet adhesiveness, plasma fibrinogen and fibrinolytic activity in juvenile-onset and maturity-onset diabetes mellitus. *J Clin Pathol* 1981;**34**:501–3.
- 13 Colmann RW, Hirsh J, Marder VJ, *et al.* *Retinal microangiopathy, hemostasis and thrombosis: basic principles and clinical practice*. Philadelphia: Lippincott, 1987.
- 14 Walmsley D, Hampton KK, Grant PF. Contrasting fibrinolytic responses in type 1 (insulin-dependent) and type 2 (non-insulin-dependent) diabetes. *Diabetic Med* 1991;**8**:954–9.
- 15 Pandolfi M, Almer LO, Holmberg L. Increased von Willebrand-antithrombophilic factor A in diabetic retinopathy. *Acta Ophthalmol Scand* 1974;**52**:823–8.
- 16 Brenner BM, Troy JL, Ballermann BJ. Endothelium-dependent vascular responses: mediator and mechanism. *J Clin Invest* 1989;**84**:1373–8.
- 17 Fuller JH, Keen H, Jarret RJ. Haemostatic variables associated with diabetes and its complications. *Br Med J* 1979;**2**:964–6.
- 18 Garcia Frade LJ, De la Calle H, Torado MC, *et al.* Hypofibrinolysis associated with vasculopathy in non-insulin-dependent diabetes mellitus. *Thromb Res* 1990;**59**:51–9.
- 19 Mansfield MW, Grant PJ. Fibrinolysis and diabetic retinopathy in NIDDM. *Diabetes Care* 1995;**18**:1577–81.
- 20 Ceriello A, Giacomello R, Colatutto A, *et al.* Increased prothrombin fragment 1+2 in Type 1 diabetic patients. *Haemostasis* 1992;**22**:50–1.
- 21 Iwashima Y, Sato T, Watanabe K, *et al.* Elevation of plasma thrombomodulin level in diabetic patients with early nephropathy. *Diabetes* 1990;**39**:983–8.