Anti-vimentin antibodies are an independent predictor of transplant-associated coronary artery disease following cardiac transplantation

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Introduction

Transplant-associated coronary artery disease (TxCAD), a rapidly progressing obliterative vascular disease developing in transplanted heart, is the major complication after the first year of cardiac transplantation[1]. A similar vasculopathy occurs following kidney transplantation where it is designated chronic rejection[2]. We have recently developed an enzyme-linked immunoassay for detection of anti-vimentin antibodies[3]. The aim of this study was to investigate whether anti-vimentin antibodies are also associated with development of TxCAD[4].

Methods

Patient study group. One hundred and nine patients who received orthotopic cardiac allografts at Harefield Hospital between 1987 and 1993 were studied. Their clinical details are described in table I[4]. Serum samples were collected pre-transplant and at 3, 6, 9, 12, 18, 24, 36, 48 and 60 months post-transplant. In total 880 samples were assayed, 109 pre-transplant and 771 post-transplant. Serum samples from 20 healthy subjects were used as negative control.

Angiography. Coronary angiograms were reviewed for TxCAD (> 25% stenosis of one or more coronary arteries in two successive annual angiograms). All patients underwent angiography annually, 1-5 years following transplantation.

Acute rejection. During the first post-transplant year all patients were monitored by surveillance endomyocardial biopsy. Biopsy fragments were graded according to the histological criteria of the International Society of Heart and Lung Transplantation[5]. Indications of rejection in two or more subsequent biopsies spanning more than 3 weeks were considered as persistent rejection.

ELISA for anti-vimentin antibodies. Recombinant human vimentin (Cymbus Biotechnology Ltd, Chandlers Ford, Hants, UK) was used and the assay was performed as previously described. Results are given as mean titre unit ± SE for IgM anti-vimentin antibodies. Normal sera (from 20 samples) gave a mean titre unit of 53 ± 32.1.

Statistical analysis. Difference in means were assessed using various tests as detailed in table I. A two-sided p value of < 0.05 was considered statistically significant. Kaplan-Meier was used to assess relationship between vimentin titre and time to develop TxCAD. Multivariate analysis was carried out using a logistic regression model and the Cox proportional hazards model for survival (time till occurrence of TxCAD).

Results

Correlation between anti-vimentin antibodies and transplant-associated coronary artery disease. Thirty-eight patients out of 109 developed TxCAD at 5 years giving an incidence of 34.9%. The mean titre of anti-vimentin antibodies in patients prior to transplantation (59.0 ± 11.4) was not different to normal sera (53 ± 32.1). There was no significant difference between the pre-transplant titre units of anti-

Table I. Patient characteristics, mean values ± SE and univariate analysis of association of risk factors with transplant-associated coronary artery disease (TxCAD).

	TxCAD (n=38)	Non-CAD (n=71)	p	Test used
Recipient age (n=106)	47.8 ± 1.9	44.2 (1.6)	0.14	2-sample t-test
Donor age (n=102)	30.4 ± 1.8	26.6 (1.2)	0.084	2-sample t-test
Recipient sex (n=109)	M 35/F 3	M 58/F 13	0.17	Fisher exact test
Donor sex (n=107)	M 21/F 17	M 41/F 28	0.69	Fisher exact test
Diagnosis (n=109)	IHD 28/CM 10	IHD 37/CM 34	0.04	Fisher exact test
No. rejections in year 1 (n=107)	1.89 ± 0.20	1.41 ± 0.16	0.020	Mann-Whitney
Persistent rejection	17	9	0.0002	Fisher exact test
Not persistent rejection	19	62		
HLA-A mismatch (n=99)			0.033	χ^2 test
0	7	3		
1	11	31		
2	17	30		
HLA-B mismatch (n=99)			0.89	χ^2 test
0	2	3		
1	11	23		
2	22	38		
HLA-DR mismatch (n=80)				χ^2 test
0	4	4		
1	13	26	0.57	
2	10	23		
Lipoprotein(a) (n=39) (mg/dl)	52.4 ± 11	47.8 ± 8	0.82	Mann-Whitney
Median	48.8	37.3		
Vimentin titre over 5 years (n=109)	217 ± 25	112 ± 13	0.0001	Mann-Whitney
Median	188	90		

CM = cardiomyopathy; HLA = human leukocyte antigen; IHD = ischaemic heart disease. From Jurcevic et al.[4], with permission.

vimentin antibodies in patients who later developed TxCAD (73.0 ± 2.7) and those who remained disease free at 5 years (51.4 ± 9.7, p = 0.88). The majority (107 out of 109) of patients increased their anti-vimentin antibody titres after transplantation. The average titre unit obtained over 5 years was significantly higher in the TxCAD positive group (217 ± 25) than in the non-CAD group (112 ± 13, p < 0.0001). Assuming that chronic rejection is an ongoing process and in order to devise a predictive test that can be performed in the first one or 2 years after transplantation, we analysed the data from the first 2 years only. The average titre for year 1 (188 ± 30 for TxCAD vs 101 ± 16 for non-CAD) and the average (combined) titre for years 1 and 2 (202 ± 23 for TxCAD vs 105 ± 14 non-CAD) were significantly higher in the TxCAD group than those who remained disease free at 5 years (p = 0.0038 and p < 0.0001, respectively).

Multivariate analysis for occurrence of transplant-associated coronary artery disease. In this series of patients, number of rejection episodes, persistent rejec-

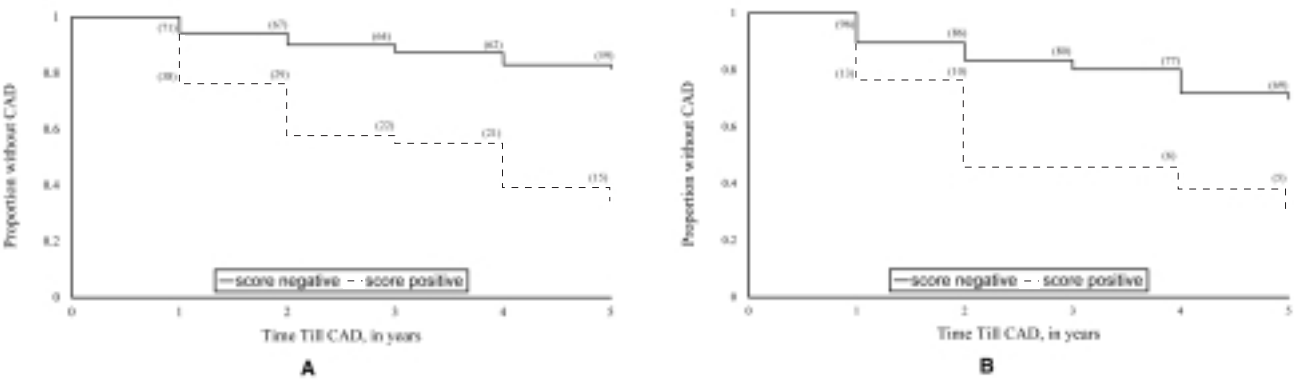

Figure 1. A: *Kaplan-Meier actuarial survival to demonstrate time to development of transplant-associated coronary artery disease (CAD) in patients with 1-year mean anti-vimentin titre ≥ 270 or persistent rejection* (dotted line) *compared to patients who are negative for this test* (solid line). B: *Kaplan-Meier actuarial survival to demonstrate time to development of transplant-associated CAD in patients with 1-year mean anti-vimentin titre ≥ 270* (dotted line) *compared to patients who are negative for this test* (solid line). *Numbers of patients at risk at each time point are given in parenthesis. From Jurcevic et al.[4], with permission.*

tion, diagnosis (ischaemic heart disease) and number of matches at the human leukocyte antigen-A locus correlated significantly with TxCAD (Table I). Multivariate logistic regression demonstrated that persistent rejection, and both the 1-year mean titre and 2-year mean titres were independent predictors of TxCAD.

Time till occurrence of transplant-associated coronary artery disease: Kaplan-Meier survival curve. Figure 1A shows the estimated probability of surviving without TxCAD for up to 5 years after transplantation. It is clear that patients with titre ≥ 270 or persistent rejection tend to have shorter times till occurrence of TxCAD. Patients with 1-year high antibody titre without persistent rejection (Fig. 1B) also show a shorter time to disease development.

References

1. Hosenpud JD, Shipley GD, Wagner CR. Cardiac allograft vasculopathy: current concepts, recent developments, and future directions. Heart Lung Transplant 1992; 11: 9-23.
2. Tullius SG, Tilney NL. Both alloantigen-dependent and -independent factors influence chronic allograft rejection. Transplantation 1995; 59: 313-8.
3. Jurcevic S, Dunn MJ, Crisp SJ, et al. A new enzyme-linked immunosorbent assay to measure anti-endothelial antibodies after cardiac transplantation demonstrates greater inhibition of antibody formation by tacrolimus compared with cyclosporine. Transplantation 1998; 15: 1197-202.
4. Jurcevic S, Ainsworth M, Pomerance A, et al. Antivimentin antibodies are an independent predictor of transplant-associated coronary artery disease after cardiac transplantation. Transplantation 2001; 71: 886-92.
5. Billingham ME, Cary NRB, Hammond ME, et al. A working formulation of the standardization of nomenclature in the diagnosis of heart and lung rejection. J Heart Lung Transplant 1990; 9: 587-93.