SCIENTIFIC LETTER

Novel mutations in the lamin A/C gene in heart transplant recipients with end stage dilated cardiomyopathy

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Dilated cardiomyopathy (DCM) is a primary myocardial disease characterised by impaired systolic function and dilatation of the left or both ventricles. The aetiology and clinical presentation of DCM are heterogeneous. At least one third of idiopathic DCM cases are familial. Knowledge of the genetics of DCM has progressed considerably in recent years.[1]

Mutations in the lamin A/C gene seem to be important aetiological factors in familial DCM. So far, several research groups have described about 40 DCM associated mutations in this gene.[1–4] Heart disease caused by lamin A/C gene mutations is characterised by conduction system disorders with the need for permanent pacemaker implantation, atrial fibrillation, severe heart failure, and increased risk for sudden cardiac death.[4] Patients with mutations in the lamin A/C gene often develop a progressive form of disease leading to heart transplantation or sudden cardiac death.[4] Therefore, we decided to investigate a homogeneous group of consecutive Finnish heart transplant recipients with end stage DCM and to search for mutations in the lamin A/C gene.

PATIENTS AND METHODS

All surviving Finnish patients who received a heart transplant between 1984 and 1998 were enrolled in the study. Of all 158 surviving patients, 81 had an initial diagnosis of primary DCM. A DNA sample was obtained from 81% (n = 66) of these 81 patients, who fulfilled the commonly approved diagnostic criteria for DCM (left ventricular ejection fraction $< 45\%$ and left ventricular end diastolic diameter > 27 mm/m^2) at the time of diagnosis. The diagnosis of idiopathic DCM was confirmed by excluding all specific causes of left ventricular dysfunction. The index subjects did not have any symptoms or signs of skeletal muscle disease and their creatine kinase concentrations were normal. All patients had undergone thorough clinical, echocardiographic, and cardiac catheterisation examinations before their operation and all relevant data were collected from the medical records. Relatives of all mutation carriers also underwent clinical and echocardiographic examinations whenever possible. The control group consisted of 150 clinically healthy subjects without a family history of DCM; 82 of them had echocardiography. The ethics committees of the universities of Kuopio and Helsinki approved the study protocol, which was in accordance with the Declaration of Helsinki.

Genomic DNA was prepared from peripheral blood leucocytes. Primers for the lamin A/C gene were designed from GenBank sequences L12399, L12400, and L12401. Genomic DNA was reamplified from the same primers (NuSieve GTG, FMC Bioproducts, Rockland, Maine, USA) and sequenced with the ABI Prism BigDye Terminator v1.1 Cycle Sequencing Kit with the ABI Prism 3100 Genetic Analyzer/Hitachi (Applied Biosystems, Foster City, California, USA).

Table 1 Clinical characteristics of lamin A/C mutation carriers

Family and member	Sex and age (years)	LVEDD (mm)	EF (%)	AVB or slow AF	PM	VT	Comments
A II: 2	M 47	67	25	AF	Yes	No	HT
A II: 3	M 45	53	42	AF	Yes	No	NYHA II
A II: 5	F 49	46	62	1° AVB	No	No	Bradycardia, palpitations
A III: 1	M 21	53	63	No	No	No	LVH
B II: 2	F 62	39	66	No	No	No	Syncope
B II: 3	M 58	64	11	No	No	No	HT
B II: 5	M 56	59	41	No	No	No	None
B III: 1	M 40	61	20	No	No	No	Recurrent AF, ablation ×3
B III: 5	F 30	44	65	No	No	No	None
B III: 6	M 26	57	56	No	No	No	Syncope, palpitations
C III: 4	M 45	57	28	2° AVB	Yes	No	HT
D II: 2	F 57	55	33	AF	Yes	Yes	ICD
D II: 4	M 40	53	68	1° AVB	No	No	None
D III: 2	M 37	51	55	1° AVB	No	No	None
E II: 3	M 46	69	29	AF	Yes	Yes	HT
F II: 2	M 32	83	22	No	No	Yes	HT
G II: 2	M 56	61	21	2° AVB	Yes	Yes	SVT, HT
G II: 4	F 55	56	36	1° AVB, LBBB	No	No	None
G II: 5	F 51	53	56	1° AVB, LAHB	No	No	None
G III: 2	M 29	54	59	No	No	No	LVH, LA size ↑
G III: 3	F 27	49	70	1° AVB	No	No	LA size ↑
G III: 5	F 20	46	67	No	No	No	None

AF, atrial fibrillation; AVB, atrioventricular block; EF, ejection fraction; F, female; HT, heart transplantation; ICD, implantable cardioverter-defibrillator; LA, left atrium; LAHB, left anterior hemiblock; LBBB, left bundle branch block; LVEDD, left ventricular end diastolic diameter; LVH, left ventricular hypertrophy; M, male; NYHA, New York Heart Association functional class; PM, pacemaker; SVT, supraventricular tachycardia; VT, ventricular tachycardia.

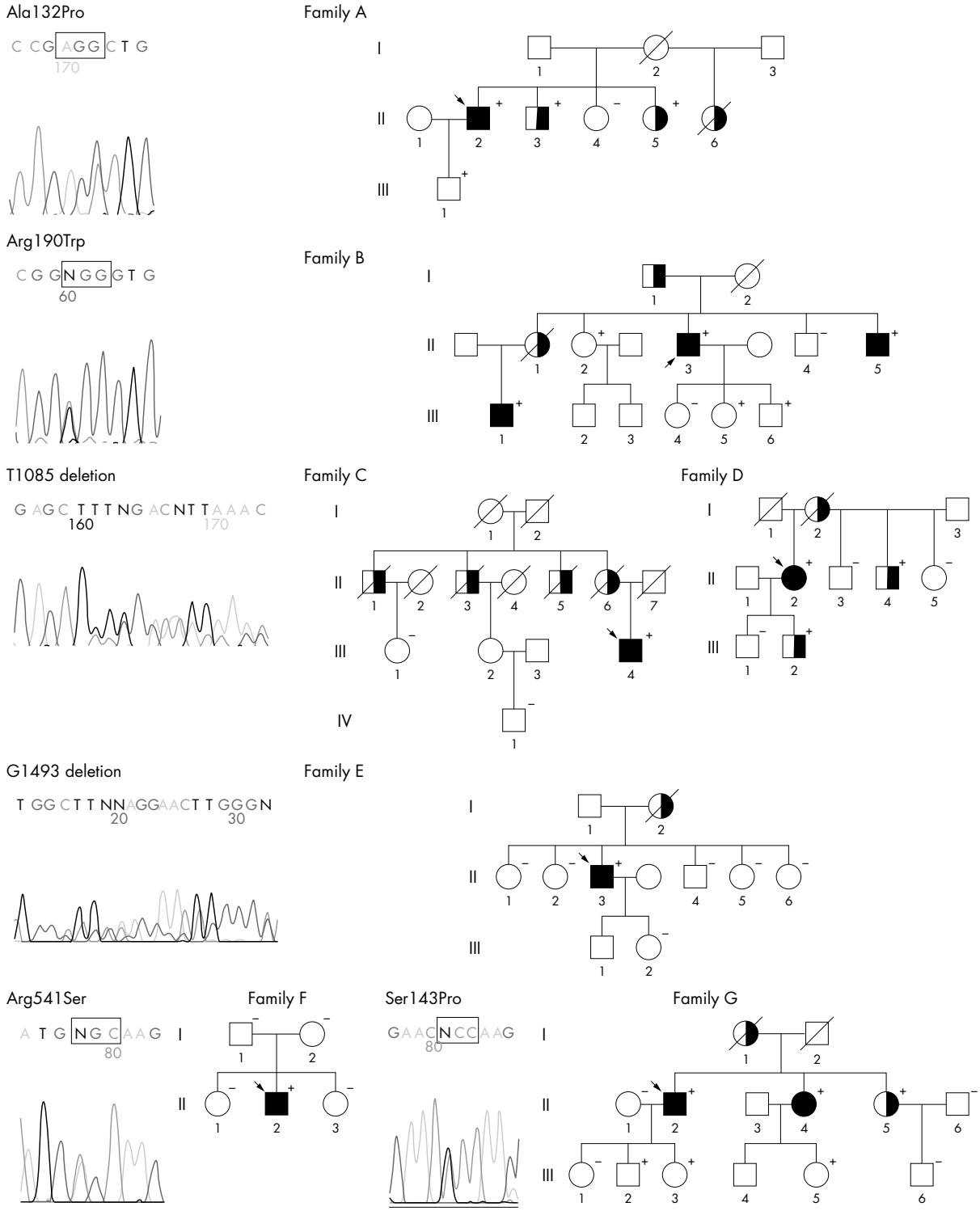

Figure 1 Pedigrees of the families carrying mutations in the lamin A/C gene. Arrow indicates the proband of the family. Squares indicate male family members, circles female family members, and symbols with a slash deceased members. Solid symbols indicate family members who fulfilled diagnostic criteria for dilated cardiomyopathy (DCM), open symbols indicate unaffected members, and semicircles and squares indicate members who do not yet fulfil all diagnostic criteria for DCM but are clinically affected (for example, they have conduction system disorder). +, mutation carrier; −, a family member who is not a mutation carrier.

RESULTS

All 12 coding exons of the lamin A/C gene from 66 heart transplant recipients with DCM were screened by polymerase chain reaction and direct sequencing. We detected six mutations (Ala132Pro, Ser143Pro, Arg190Trp, T1085 deletion, G1493 deletion, and Arg541Ser) in six unrelated index patients. Four of these mutations appeared to be novel, whereas two mutations (Arg190Trp and Ser143Pro) have been detected previously.[3–5] One of the mutations, Arg541Ser, seems to be a de novo mutation because the index patient's parents did not carry this mutation. We detected the G1493 deletion only in the index patient, and none of the family

members carried the mutation. However, according to medical records, this patient's mother had arrhythmias and died of heart disease.

We screened an additional group of 20 patients with DCM for the mutations and found the T1085 deletion in exon 6 in another family with DCM. The two families carrying this mutation are unrelated and do not have a shared ancestry at least in the closest generations. Figure 1 presents the mutations and the pedigrees of the families. Table 1 shows the clinical data of the mutation carriers.

DISCUSSION

The primary goal of this study was to investigate patients with end stage DCM necessitating heart transplantation and to search for mutations in the lamin A/C gene. This is warranted, since lamin A/C mutations are known to cause relatively malignant disease with an increased risk for sudden cardiac death and need for heart transplantation.[4] In this homogeneous group of patients with DCM we found five disease associated mutations in the lamin A/C gene. These mutations are likely to be disease causing because none of these mutations were found in 150 healthy control patients, all mutations affect conserved residues of the gene, and the phenotype of the patients is quite similar to the phenotype of previously described mutations in the lamin A/C gene. Most often DCM associated mutations have been identified in the rod domain of the protein. The rod domain forms the basic structural unit of lamin assembly and seems to be critical for the development of the DCM phenotype. The morphological changes documented to be associated with lamin A/C mutations include focal disruptions, bleb formation, nuclear pore clustering, herniations, changes in assembly properties, and altered emerin localisation.[5]

This study strengthens the view of a similar phenotype in lamin A/C mutation carriers. Similarly to patients with previously reported phenotypes caused by lamin A/C gene mutations our patients also had conduction system disorders (55%), need for a permanent pacemaker (27%), atrial fibrillation (27%), ventricular tachycardia (14%), and end stage heart failure necessitating heart transplantation (27%). Typically, sinoatrial or atrioventricular block seems to be the first manifestation of progressive disease and usually permanent pacemaker insertion becomes necessary at some stage. In fact, of all the DCM patients who underwent heart transplantation in Finland during the data collection period, only the carriers of lamin A/C mutations needed a permanent pacemaker before their operation. Atrial fibrillation and paroxysmal tachyarrhythmias were present in four of the heart transplant recipients and they were also common in two of the families (A, B). Although dilatation of the heart and left ventricular dysfunction are late clinical manifestations, the disease seems to progress fairly rapidly. The diagnosis was established when our patients were a mean age of 38 years (range 26–47) and they underwent transplantation at a mean age of 42 (32–48). However, the relatives of the index patients seemed to have a milder form of the disease than index patients. One obvious reason for this is that almost all index patients were older than the relatives. Typically, the first signs of the disease (typically conduction system disorder) developed during their 30s or 40s and congestive heart failure somewhat later.

In this study, the six mutations found in the lamin A/C gene explained DCM in about 9% of the patients. This prevalence is the same as that observed in our previous study of patients carrying the founder mutation Ser143Pro in the lamin A/C gene.[6] In that study, the Ser143Pro mutation explained 7% of all cases in an unselected DCM population. The prevalence of the lamin A/C mutations in this series can also be underestimated, since these mutations tend to cause rapidly progressive disease and a considerable proportion of the patients may have actually died suddenly before being considered for cardiac transplantation. All Finnish transplant recipients are evaluated at the same clinic and these patients come from all regions of the country. Owing to genetic isolation the Finnish population is ideally suited to investigation of genetic diseases.

We conclude that the mutations in the lamin A/C gene are relatively common in Finnish patients with DCM and that these mutations should probably be screened at least among patients with conduction system disorders.

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