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Original Article

Clinical profile in arrhythmogenic cardiomyopathy and a recessive plakophilin-2 gene mutation

Muzaffar Ali[a,*], Imtiyaz A. Bhat[b], Imran Hafeez[a], Mohd Iqbal Dar[a], Jahangir Rashid Beig[a], Zafar Amin Shah[b], Khurshid Iqbal[a]

[a] Department of Cardiology, Sher-I-Kashmir Institute of Medical Sciences, Srinagar, J&K, India
[b] Department of Immunology and Molecular Medicine Sheri Kashmir Institute of Medical Sciences, Srinagar, J&K, India

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ABSTRACT

Objective: Arrhythmogenic Cardiomyopathy (ACM) is not an uncommon cause of cardiac morbidity in Kashmir valley. This study was designed to document various clinical features and to sequence exons 11 and 12 of plakophilin 2 (*PKP2*) gene in these patients.
Methods: ACM patients who attended Cardiology outpatient department of our institute from January 2014 to April 2015 were included in the study. Their records were reviewed. Controls were randomly selected from cardiology OPD who had no history or family history of cardiac illness and had a normal cardiac examination. A blood sample was also taken from both the groups for sequencing of exon 11 and 12 of plakophilin 2 gene. ACM patients were followed-up until July 2016.
Results: Eleven ACM patients and seven controls were included in the study. Most common mode of presentation was ventricular tachycardia (VT). Two patients had LV systolic dysfunction. One patient had a splice site mutation in exon 12 of plakophilin 2 gene and one patient died during follow-up. One of the controls had an intronic variation that has no pathogenic significance vis-à-vis ACM.
Conclusion: Our study describes various clinical parameters in ACM patients and a recessive plakophilin 2 mutation after a limited PKP2 gene sequencing.

1. Introduction

Arrhythmogenic Cardiomyopathy (ACM) is a genetic and structural heart disease characterized by gradual replacement of cardiac myocytes by adipocytes and fibrosis, predominantly of the right ventricle.[1] The left ventricle is involved in up to 50% of cases.[2] The most common gene involved in ACM is Plakophilin-2 gene (*PKP2*) that accounts for approximately 20% of the cases.[3] This study was designed to document various clinical features of ACM and to sequence exons 11 and 12 of *PKP2* gene in these patients.

2. Methods

Patients of ACM (diagnosed on the basis of Revised Task Force Criteria of ACM),[4] who were under Cardiology outpatient department follow-up of our Institute, and attended the hospital from January 2014 to April 2015 were included in the study. Their previous records were reviewed, and a blood sample was taken for sequencing, after written informed consent.

Controls were randomly selected from cardiology outpatient department who had no history or family history of cardiac illness and had a normal cardiac examination. A blood sample was also taken from them for sequencing after written informed consent.

The study was cleared by our institute's ethical committee.

Peripheral blood samples were used as the source of DNA. DNA was extracted by DNA extraction kit (Qiagen, US). The quality of the DNA obtained from the blood samples was analysed on 1% agarose gel. PCR amplification was done using the primers (Table 1) which were designed by using PrimerQuest® program, IDT, Coralville, USA. PCR amplified products were checked on 2% agarose gel and sent for sequencing to Macrogen Ltd, Seoul for sequencing of exons 11 and 12.The results were compared with *PKP2* gene reference sequence (ENST00000070846.10).[5]

Exons 11 and 12 were selected based on the number of reports that have reported pathogenic variations of these two exons and because of financial constraints. Exons 11 and 12, when taken as combined, have the maximum number of reported pathogenic

* Corresponding author.
E-mail address: dralimuzaffar@gmail.com (M. Ali).

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Table 1
PKP2 primers.

Gene	Primers
PKP-Exon 11-F	5′- GCCTGAATGACAGAGAGAGAC-3′
PKP-Exon 11-R	5- CCGGTTTATCACCTACTCCTTAC-3′
PKP Exon 12-F	5′-ACTCTCCCTGATTTGGTTTCC-3′
PKP Exon 12-R	5′-GACTCCTGCTTTGCCTACAT-3′

variations than any other two contiguous exons of Plakophilin 2 gene.[6]

3. Results

Eleven patients presented to our outpatient department from January 2014 to April 2015. All of them were unrelated. Their records were reviewed.

Table 2
Baseline characteristics of ACM patients.

No.	Year of diagnosis	Age at presentation	Sex	Mode of presentation	Reduced LVEF
1	2008	43	M	Syncope(VT)	No
2	2012	18	F	Syncope(VT)	No
3	2013	14	M	Syncope(VT)	Yes
4	2013	18	M	Syncope(VT)	Yes
5	2010	45	M	Palpitations(VT)	No
6	2012	38	M	Syncope(VT)	No
7	2012	22	M	Palpitations(VT)	No
8	2010	50	M	Palpitations(VT)	No
9	2010	40	M	Palpitations(VT)	No
10	2012	22	M	Palpitations(VT)	No
11	2007	37	M	Palpitations(VPCs)	No

Table 3
Revised Task Force criteria and other findings in each case.

No.	Major criteria present	Minor criteria present	Other Findings
1	1. ECG: T wave inversion in leads V1-V4 2. Echo: Thinned out RV apex and free wall and PSAX RVOT diameter of 37 mm 3. LBBB morphology VT with superior axis		
2.	1. ECG: T wave inversion in leads V1–V6 2. Echo: Grossly dilated RA & RV, PSAX RVOT diameter of 38 mm 3. Cardiac MRI: RV free wall hypokinesia and RV ejection fraction of 26.3% 4. LBBB morphology VT with superior axis	ECG: More than 1000 VPCs on 24-h Holter monitoring	Echo: Severe Low-Pressure TR
3.	1. LBBB morphology VT with superior axis 2. Cardiac MRI: Grossly dilated and hypokinetic RV; RVEF of 17%	1. Echo: Dilated and hypokinetic RV and PSAX RVOT diameter of 35 mm	Echo: LV systolic dysfunction
4.	1. ECG: T wave inversion in leads V1–V6 2. Echo: Grossly dilated RA & RV, RV free wall hypokinetic and PSAX RVOT diameter of 39 mm 3. LBBB morphology VT with superior axis 4. Cardiac MRI: Thinned out RV free wall and markedly decreased RV systolic function		Echo: Severe low-pressure TR, LV systolic dysfunction
5.	1. ECG: T wave inversion in leads V1–V4 2. Echo: Grossly dilated RA & RV, PSAX RVOT 40 mm, RV systolic dysfunction 3. Cardiac MRI: Thinning of RV free wall and RVEF OF 21.9%	2. LBBB morphology VT with inferior axis	
6.	1. ECG: T wave inversion in leads V1–V4 2. LBBB morphology VT with superior axis		Echo: Mildly dilated RA & RV, PSAX RVOT = 31 mm, mild TR Cardiac MR: Fatty infiltration of RV free wall, mild hypokinesia of RV free wall, RVEF 41%[††]
7.	1. ECG: T wave inversion in leads V1–V5 2. ECG: Epsilon waves 3. Echo: Thinning of RV apex and PSAX RVOT of 40 mm	1. LBBB morphology VT with inferior axis	
8.	1. ECG: T wave inversion in leads V1-V5 2. ECG: Epsilon waves 3. LBBB morphology VT with superior axis	1. RV Apical thinning and PSAX RVOT dia of 35 mm	
9.	1. Echo: Thinned out RV lateral wall and PSAX RVOT of 40 mm 2. CMRI: Severe RV lateral wall thinning and RVEF of 19%	1. History of ACM in brother[†] 2. LBBB morphology VT with inferior axis	
10.	1. ECG: T wave inversion in leads V1–V4 2. Echo: RV free wall hypokinesia and PSAX RVOT diameter 0f 41.5 mm 3. LBBB morphology VT with superior axis		
11.	1. Echo: RV lateral wall thinning and PSAX RVOT diameter of 49 mm 2. Cardiac MRI: Thinned out anterior RV wall and RVEF OF 27%		

[†] In whom it was not possible to determine whether he met current Task Force criteria.
[††] These finding do not fulfil any major or minor criterion.

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