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The genetics underlying idiopathic ventricular fibrillation: A special role for catecholaminergic polymorphic ventricular tachycardia?☆

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ABSTRACT

Background: Ventricular fibrillation (VF) is a major cause of sudden cardiac death. In some cases clinical investigations fail to identify the underlying cause and the event is classified as idiopathic (IVF). Since mutations in arrhythmia-associated genes frequently determine arrhythmia susceptibility, screening for disease-predisposing variants could improve IVF diagnostics.

Methods and results: The study included 76 Finnish and Italian patients with a mean age of 31.2 years at the time of the VF event, collected between the years 1996–2016 and diagnosed with idiopathic, out-of-hospital VF. Using whole-exome sequencing (WES) and next-generation sequencing (NGS) approaches, we aimed to identify genetic variants potentially contributing to the life-threatening arrhythmias of these patients. Combining the results from the two study populations, we identified pathogenic or likely pathogenic variants residing in the *RYR2*, *CACNA1C* and *DSP* genes in 7 patients (9%). Most of them (5, 71%) were found in the *RYR2* gene, associated with catecholaminergic polymorphic ventricular tachycardia (CPVT). These genetic findings prompted clinical investigations leading to disease reclassification. Additionally, in 9 patients (11.8%) we detected 10 novel or extremely rare (MAF < 0.005%) variants that were classified as of unknown significance (VUS).

Conclusion: The results of our study suggest that a subset of patients originally diagnosed with IVF may carry clinically-relevant variants in genes associated with cardiac channelopathies and cardiomyopathies. Although misclassification of other cardiac channelopathies as IVF appears rare, our findings indicate that the possibility of CPVT as the underlying disease entity should be carefully evaluated in IVF patients.

1. Introduction

Ventricular fibrillation (VF) is a severe form of cardiac arrhythmia, often resulting in sudden cardiac death (SCD). Typically, VF results from an underlying ischemic, electrical, infectious or structural disease of the heart. Rarely, clinical examinations fail to identify an underlying cause and VF is classified as idiopathic [1–3]. Idiopathic ventricular fibrillation (IVF) is a diagnosis by exclusion, with a likely complex etiology, although in some cases it may have a strong genetic basis. During the past decades, several genetic arrhythmia disorders, such as Brugada syndrome and long QT syndrome (LQTS), used to reside within the category of IVF [3,4]. Despite the improvements in the diagnosis of these syndromes, concealed forms of these known genetic disorders may still explain a proportion of IVF or SCD incidents [3,5]. For instance, catecholaminergic polymorphic ventricular tachycardia (CPVT), typically caused by mutations in the *RYR2* gene, may still get misclassified as IVF [6]. Moreover, mutations in other arrhythmia-associated genes such as *SCN5A* and *KCNH2* may initially manifest as VF, although in most of these cases an underlying electrical disease is later identified [7–9].

☆ All authors take the responsibility for all aspects of the reliability and freedom from bias of the data presented and their discussed interpretation.

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Compared to other arrhythmogenic diseases, there have been relatively few studies focusing on characterizing the genetic landscape of IVF [3,10]. Genes more recently associated to IVF are *DPP6* and *SEMA3A*, identified in a Dutch family and in a Japanese cohort, respectively, and the newly-identified *CALM1* [11–13]. The reported yield of genetic testing in all IVF cohorts is however relatively low [3,10]. Overall, these observations suggest that the genetic background of IVF is likely heterogeneous and that it could also be of non-monogenic origin [3,10].

By combining the results from two independent European IVF cohorts, we aimed to characterize the spectrum of genetic variation underlying IVF. Our study suggests that a subset of patients initially diagnosed with IVF carry clinically-relevant variants in genes associated to inherited arrhythmogenic diseases.

2. Methods

2.1. Subjects

The Finnish cohort includes 36 unrelated patients (15 males and 21 females) drawn from the Finnish Inherited Arrhythmia Disorder Registry. These patients were resuscitated following out of-hospital VF of unknown cause between the years 1996 and 2011, and had a mean age of 31 ± 11 years when experiencing the arrhythmic episode (range 12–48 years). The Italian cohort includes 40 patients (20 males and 20 females) referred to the investigators of the Center for Cardiac Arrhythmias of Genetic Origin from 2006 to 2016. The patients' mean age at VF was 32 ± 16 years (ranges 0–63 years). The main clinical characteristics of the patients are summarized in Tables 1 and 2.

All patients underwent routine clinical work-up in order to identify the cause of VF, before receiving a diagnosis of IVF and an implantable cardiac defibrillator (ICD) in the referring hospital. The details of the clinical tests performed for each patient are provided in Supplemental Tables A.1 and A.2. No patient had a previous history of a cardiac condition, nor clinical findings in resting electrocardiogram (ECG), coronary artery angiography, or cardiac imaging explaining the VF. For the purpose of the current study, patients' medical history was re-evaluated by reviewing available hospital records. The patients were contacted if abnormal findings, related to potentially significant genetic variants, emerged. In the analysis of the relatives we relied on anamnestic information and refrained from a broader clinical investigation if there was no reason to suspect a particular disease entity. Informed consent was obtained from all patients. All investigations were performed in accordance with the Helsinki Declaration and approved by the local ethical review boards.

2.2. DNA sequencing

The Finnish cohort was genetically evaluated with whole exome sequencing (WES) focusing on 100 genes associated with channelopathies and cardiomyopathies selected based on the review by Wilde and Behr (Supplemental Table A.3) [14]. DNA sequencing was performed at the Technology Center at the Institute for Molecular Medicine Finland, FIMM. The exome targets were captured with the NimbleGenSeqCapEz Human Exome Library v2.0 (www.nimblegen.com/products/seqcap/ez/index.html), followed by sequencing with the Illumina Genome Analyzer-IIx platform. The alignment to the human reference genome hg19 and variant calling of

Table 1
Clinical, electrophysiological and molecular genetic findings – The Finnish cohort.

			Electrocardiogram				24 h ECG	EP study		Echocardiography			
DNA#	Age at VF	Sex	hr (min^{-1})	PQ (ms)	QRS (ms)	QTc (ms)	VES/24 h	HV (ms)	VT/VF	LVEDD (mm)	LVEDD (mm)*	EF %	Variant(s)
F1	25	M	80	144	98	416	4	39	No	45	34	55	
F2	37	M	80	130	100	393	0	NA	No	46	46	66	
F3	38	F	68	150	85	436	1646	NA	No	54	53	63	
F4	39	F	73	166	92	474	4110	56	No	51	52	65	
F5	20	M	47	198	112	372	8	NA	No	57	52	68	
F6	36	M	50	170	130	475	NA	62	No	53	49	50	
F7	18	F	50	184	86	374	24,987	50	No	47	53	64	*MYBPC3*-p.Arg1138His
F8	30	M	76	180	70	439	NA	49	No	49	56	65	
F9	12	F	94	120	76	432	0	NA	No	42	51	74	
F10	**12**	**M**	**75**	**150**	**100**	**447**	**NA**	**42**	**No**	**48**	**57**	**62**	***RYR2*-p.Leu575Phe**
F11	44	F	93	148	80	423	2981	NA	NA	55	58	68	
F12	23	F	68	144	76	383	NA	45	No	50	56	60	
F13	22	M	64	152	84	449	NA	41	No	57	57	60	
F14	44	F	60	180	150	470	NA	69	No	56	48	51	*DMD*-p.Leu1264Ser; *MYBPC3*-p.Arg1138His
F15	30	M	71	136	104	392	NA	44	No	53	55	69	
F16	15	M	73	178	96	474	10	45	Yes	51	50	66	*HCN4*-p.Arg713His; *RYR2*-p.Asn3308Ser
F17	39	M	60	150	70	400	11,321	44	Yes	53	47	60	*SGCD*-p.Ala204Ser
F18	42	F	70	164	86	464	1696	NA	NA	54	59	61	
F19	43	M	53	144	106	395	84	38	No	52	51	60	*MYH7B*-p.Arg231Trp, p.Lys108Asn; *SCN5A*-p.Ala572Asp
F20	23	F	70	140	80	432	NA	59	No	46	52	57	
F21	43	F	87	166	74	458	9797	48	No	51	50	70	*MYH7B*-p.Asp1096His; *FKRP*-p.Trp418*
F22	18	M	72	166	100	394	NA	50	No	41	43	65	
F23	22	M	90	148	94	429	NA	37	No	53	53	68	
F24	34	M	63	173	100	410	36	50	No	60	48	66	*TTN*-p.Arg24987*, p.Lys29627Thr; *SCN5A*-p.Pro2006Ala
F25	35	F	66	142	95	420	NA	36	No	56	50	54	
F26	17	F	96	120	100	430	NA	NA	NA	49	51	60	
F27	**18**	**F**	**79**	**154**	**100**	**459**	**NA**	**NA**	**NA**	**57**	**48**	**67**	***RYR2*-p.Gln3774Arg**
F28	48	M	67	176	94	391	NA	64	No	61	61	50	
F29	43	F	53	152	92	432	41	46	No	45	44	70	
F30	**13**	**F**	**65**	**150**	**80**	**448**	**0**	**32**	**No**	**35**	**43**	**60**	***CACNA1C*-p.Gly402Ser;***CRYAB-p.Pro46Leu; *DMD*-p.Leu2792Phe
F31	19	F	52	156	92	410	NA	NA	NA	48	49	65	*RYR2*-p.Gly2367Arg; *SCN5A*-p.Ala572Asp
F32	47	F	90	150	108	416	NA	NA	NA	58	58	63	
F33	38	F	53	156	90	385	NA	NA	No	53	48	71	
F34	31	M	61	170	80	403	3676	39	No	56	57	51	
F35	44	F	50	152	90	374	13	50	No	46	44	58	
F36	39	F	84	154	100	473	40	49	No	46	41	57	*SCN5A*-p.Ala572Asp
Mean	30.6		69.5	155.9	93.6	424.2		47.4		50.9	50.7	62.2	

Clinical, electrophysiological and molecular genetic findings of IVF patients. The carriers of pathogenic and likely pathogenic variants are indicated in bold. 24 h ECG = 24 h ambulatory electrocardiogram, EP study = electrophysiological study, VF = ventricular fibrillation, HR = heart rate, VES = ventricular extrasystole, VT = ventricular tachycardia, LVEDD = left ventricular end diastolic diameter, EF = ejection fraction, NA = not available. * = LVEDD and EF at the end of the follow-up.

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