Filamin-C variant-associated cardiomyopathy: A pooled analysis of individual patient data to evaluate the clinical profile and risk of sudden cardiac death

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BACKGROUND Mutations in filamin-C *(FLNC)* are involved in the pathogenesis of arrhythmogenic cardiomyopathy (ACM) and dilated cardiomyopathy (DCM), and have been associated with a left ventricular (LV) phenotype, characterized by nonischemic LV fibrosis, ventricular arrhythmias, and sudden cardiac death (SCD).

OBJECTIVE The purpose of this study was to investigate the prevalence of *FLNC* variants in a gene-negative ACM population and to evaluate the clinical phenotype and SCD risk factors in *FLNC*-associated cardiomyopathies.

METHODS ACM probands who tested negative for mutations in ACM-related genes underwent *FLNC* genetic screening. Clinical and genetic data were collected and pooled together with those of previously published *FLNC*-ACM and *FLNC*-DCM patients.

RESULTS In a cohort of 270 gene-elusive ACM probands, 12 (4.4%) had *FLNC* variants, and 13 additional family members carried the same mutation. Eighteen *FLNC* variant carriers (72%) had a diagnosis of ACM (72% male; mean age 45 years). On pooled analysis, 145 patients with *FLNC*-associated cardiomyopathies were included. Electrocardiographic (ECG) low QRS voltages were detected in 37%, and T-wave inversion (TWI) in inferolateral/lateral leads in 24%. Among 67 patients who had cardiac magnetic resonance (CMR), LV nonischemic late gadolinium enhancement (LGE) was found in 75%. SCD occurred in 28 patients (19%), 15 of whom showed LV nonischemic LGE/fibrosis. Compared with patients with no SCD, those who experienced SCD more frequently had inferolateral/lateral TWI ($P = .013$) and LV LGE/fibrosis ($P = .033$).

CONCLUSION Clinical phenotype of FLNC cardiomyopathies is characterized by late-onset presentation and typical ECG and CMR features. SCD is associated with the presence of LV LGE/fibrosis but not with severe LV systolic dysfunction.

KEYWORDS Arrhythmogenic cardiomyopathy; Cardiac magnetic resonance; Dilated cardiomyopathy; Filamin-C; Sudden cardiac death

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Introduction

Arrhythmogenic cardiomyopathy (ACM) is a rare inherited heart muscle disease characterized by myocardial scar, systolic right ventricular (RV) and/or left ventricular (LV) dysfunction, and malignant ventricular arrhythmias (VAs).[1–3] The hallmark of ACM is the replacement of ventricular myocardium by fibrofatty tissue, which progresses over time.[1–3] Approximately one-half of ACM patients harbor genetic variants in genes encoding major components of the cardiac desmosomes, although mutations in nondesmosomal genes have been also described in association with ACM.[3,4] Among them, mutations in gene

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encoding filamin-C (*FLNC*), traditionally linked to myofibrillar myopathy (MIM#609524) and hypertrophic cardiomyopathy (MIM#617047), are raising particular interest given their possible involvement in the pathogenesis of a peculiar LV phenotype, characterized by extensive nonischemic LV fibrosis, life-threatening VAs, and sudden cardiac death (SCD).[5–10] This *FLNC*-associated phenotype combined with the cellular function of the protein, which serves as a structural actin cross-linker between sarcomeric Z-disc and sarcolemma, is increasingly recognized as a blend of dilated cardiomyopathy (DCM) and ACM.[9,10] Notwithstanding, the presence of *FLNC* variants in the context of a largely fibrotic LV seems to have significant implications for treatment of these patients, because their arrhythmic and SCD risk seem unrelated to the degree of systolic dysfunction.[5,9,10]

Here we present a genotype-phenotype study focused on *FLNC* variants, with the aim of reporting their prevalence in our ACM population from a tertiary referral hospital. Moreover, by pooling individual patient data from the published series of *FLNC*-related ACM/DCM, we aimed to characterize their clinical phenotype, arrhythmic outcome, and SCD risk factors.

Methods

Study population and cardiomyopathy evaluation

This is an observational, single-center, retrospective study from the Clinical Genetic Center of Arrhythmic Cardiomyopathies, University Hospital of Padua, Italy. All patients provided written informed consent before inclusion in the study, in accordance with the protocol approved by the regional ethics committee. All clinical investigations were conducted according to the principles expressed in the Declaration of Helsinki. Of all consecutive index cases who tested negative for likely pathogenic/pathogenic mutations in ACM-related genes, we reanalyzed the DNA searching for genetic variants in *FLNC*. All of these patients were referred to our tertiary center between January 2011 and January 2021 and had a definite, borderline, or possible diagnosis of ACM according to the 2010 Task Force diagnostic criteria.[11] A reclassification according to the recently published ACM 2020 Padua Criteria also was performed, particularly in the presence of biventricular or left-dominant phenotypic variants.[12]

Detailed clinical evaluation included medical and family history, 12-lead electrocardiogram (ECG), 24-hour ambulatory ECG, 2-dimensional transthoracic echocardiogram, and cardiac magnetic resonance (CMR) imaging with late gadolinium enhancement (LGE). Technical equipment and protocols of each routine investigation have been reported in detail elsewhere.[13,14] When appropriate, genetic testing and clinical phenotyping were extended to consenting family members.

In the setting of SCD and/or heart transplantation, the heart was studied according to the Guidelines of the Association for European Cardiovascular Pathology.[15]

Search strategies and selection criteria

Electronic search engines included PubMed/Medline and Scopus for the following search keyword strings: (filamin-c) OR FLNC AND arrhythmogenic cardiomyopathy and (filamin-c) OR FLNC AND dilated cardiomyopathy. Other keyword strings also were tested: (filamin-c) OR FLNC AND ARVC/D, (filamin-c) OR FLNC AND ARVD/C, (filamin-c) OR FLNC AND ACM, (filamin-c) OR FLNC AND AC, (filamin-c) OR FLNC AND DCM. We carefully reviewed reference lists of original publications and review articles for missing studies. Duplicates were eliminated. All studies were filtered independently by 2 reviewers (RC, KP), and occasional disagreements were settled by an additional author (BB).

Only original peer-reviewed articles published since the first description of *FLNC* in 2000[16] providing accurate phenotypic data of patients with *FLNC* were considered. If the same cohort was reported by multiple studies, only the first study published or the one containing more clinical details was included. If multiple and discordant diagnostic criteria for ACM were applied to the same cohort, then the study was excluded from the analysis. Healthy *FLNC* variant carriers (genotype+/phenotype–) were excluded. Patients with *FLNC*-associated hypertrophic and restrictive cardiomyopathy were not object of this analysis.

Pooled clinical data analysis

Published individual patient data together with data of our cohort were used for the pooled analyses. Data extracted included demographics, ECG and cardiac imaging (echocardiogram and CMR) characteristics, phenotype diagnosis, arrhythmic history, and outcome. Among ECG characteristics, the presence/absence of low QRS voltages, V_1–V_3, or inferolateral/lateral T-wave inversion (TWI) were considered only when adequately reported. Among imaging features, RV involvement was considered in the presence of RV criteria for ACM (or when adequately reported) or LV dilation in the presence of LV end-diastolic diameter $\geq$55 mm by echocardiogram (or when adequately specified). Left ventricular ejection fraction (LVEF) from CMR imaging (or echocardiogram, when CMR was not available) was considered. The phenotype diagnosis (ACM or DCM) was that reported in each study for each patient. Presence of >500 premature ventricular beats (PVBs) in 24 hours, sustained ventricular tachycardia, SCD, cardiac death, or cardiac transplantation were considered when adequately reported. SCD composite endpoint included SCD, aborted cardiac arrest, and implantable cardioverter-defibrillator intervention on ventricular fibrillation.

Allele threshold for variants inclusion

The minor allele frequency threshold to consider a variant clinically relevant was <0.01%, as the estimated prevalence of the disease ranges between 1:2000 and 1:5000. Moreover, we also considered the frequency of the most recurrent ACM variant (PKP2: c.2146-1G>C) and the algorithm proposed

by Whiffin et al,[17] which estimates the expected frequency of ACM variants at 6.7×10^{-5} ($4.1–9.2 \times 10^{-5}$).

Statistical analysis

Statistical analysis is reported in the Supplemental Methods.

Results

FLNC variants in the Padua cohort

Two hundred seventy gene-elusive ACM index cases with genetic testing detected 12 *FLNC* rare variants (4.4%) (Table 1), including 7 "radical" variants (4 deletions/ insertions, 2 nonsense and 1 splice site variants) classified as pathogenic/likely pathogenic (P/LP), and 5 missense variants (minor allele frequency <.01%) classified as variants of unknown significance according to American College of Medical Genetics and Genomics recommendations. In order to avoid overestimating genetic variants causality in *FLNC*, radical variants prevalence was compared in our ACM cohort vs gnomAD control v2.1.1 database, showing 76.55-fold (32.65–178.7, $P < .0001$) enrichment.

Cascade genetic screening was feasible in 5 of the 12 families (Figure 1); 13 of the 24 family members (54%) tested carried a *FLNC* genetic variant (9 radical variants, *FLNC-R*; 4 missense *FLNC-M*). Overall clinical reassessment was performed in 25 *FLNC* variant carriers (*FLNC*+ 25/36 [69%]), both index cases (n = 12) and relatives (n =13).

FLNC variant carriers' clinical phenotype

FLNC+ index cases had a mean age of 51 ± 16 years (range 17–66 years), and 4 of 12 reported a family history for undefined nonischemic cardiomyopathy (Families A, C, D, L). In Family C, the father's proband had a nonischemic cardiomyopathy and received an implantable cardioverter-defibrillator for secondary prevention. In family L, the paternal cousin of the proband (Figure 2) underwent cardiac transplantation at age of 34 years due to arrhythmic storm in mildly dilated LV (Figure 3).

Detailed clinical and histopathological findings of the *FLNC*+ probands are given in Table 2 and Supplemental Table 1.

Based on the 2010 Task Force diagnostic criteria, a diagnosis of definite in 4, borderline in 5, and possible in 9 ACMs was achieved, whereas classification according to the 2020 Padua Criteria was right dominant in 4 (22%), left dominant in 10 (56%), and biventricular in 4 (22%).

Because 18 patients fulfilled criteria for ACM diagnosis, the disease penetrance in all *FLNC* carriers was therefore incomplete and estimated 72% (18/25) and 79% (15/19) in patients older than ≥35 years. As such, we recalculated the allele frequency threshold of *FLNC* genetic variants using the algorithm of Whiffin et al[17] and penetrance of about 70%. The estimated threshold for a putative causative *FLNC* variant in ACM is 4.65×10^{-5} ($2.84–6.38 \times 10^{-5}$), which is slightly lower compared to the frequency of desmosomal variants with 30%–50% penetrance.

Pooled analysis of patients with *FLNC* variant ACM- and DCM-associated cardiomyopathy

The initial search identified 51 studies that fully examined patients carrying *FLNC* variants and affected by ACM and/or DCM. Eight records satisfied the inclusion criteria for analysis (Supplemental Figure 1). Among the 160 *FLNC*+ carriers identified, 42 patients with a diagnosis of ACM (22 index cases and 20 family members) and 85 with DCM (48 index cases and 37 family members) were included for analysis. Among ACM patients, the diagnosis provided was definite in 11, borderline in 6, and possible in 25. A left-dominant phenotype was described in 28 (67%).

The results of the pooled genotype-phenotype analysis of all *FLNC* cardiomyopathy patients (n = 18 + 127) are given in Table 3 and Supplemental Table 2.

In terms of outcome, the SCD composite endpoint occurred in 28 patients (20%), of whom 6/6 CMR and 9/9 postmortem analyses showed LV nonischemic fibrosis. Compared with patient who did not experience the SCD

Table 1 *FLNC* variants identified in patients of the Padua cohort

No.	g.DNA	c.DNA	AA change	dbSNP ID	MAF (gnomAD)	ACMG
1	g.128478819C>G	c.1373C>G	p.Pro458Arg	rs7734005500	//	VUS
2	g.128493639	c.6325 A>G	p.Ile2109Val	rs755736125	ALL:0.002%	VUS
3	g.128488758	c.4724C>G	p.Thr1575Ser	rs773294974	ALL:0.0016%	VUS
4	g.128482300C>G	c.2137C>G	p.Pro713Ala	rs771843379	ALL:0.000402%	VUS
5	g.128475622G>C	c.595G>C	p.Ala199Pro	//	//	VUS
6	g.128492728C>T	c.5926C>T	p.Gln1976*	//	//	P
7	g.128485300	c.3781G>T	p.Glu1261*	//	//	P
8	g.128480675_128480676insT	c.1623_1624insT	p.Pro542Serfs*21	//	//	LP
9	g.128481258_128481262del	c.1848_1852delCGAAT	p.Ile616Metfs*2	//	//	LP
10	g.128490538	c.5398+1 G>T	p.?	//	//	LP
11	g.128494869dupG	c.7037dup	p.Leu2347Profs*9	//	//	P
12	g.128475403delAACCT GAAGCTGATCCT	c.376_392delAACCTGA AGCTGATCCT	p.Asn126GlyfsTer20	//	//	P

AA = amino acid change; ACMG = American College of Medical Genetics and Genomics classification; c.DNA = coding exon localization; dbSNP = Single Nucleotide Polymorphism Database; FLNC = filamin-C; g.DNA = genomic DNA localization; LP = likely pathogenic; MAF = minor allele frequency; P = pathogenic; VUS = variant of unknown significance.

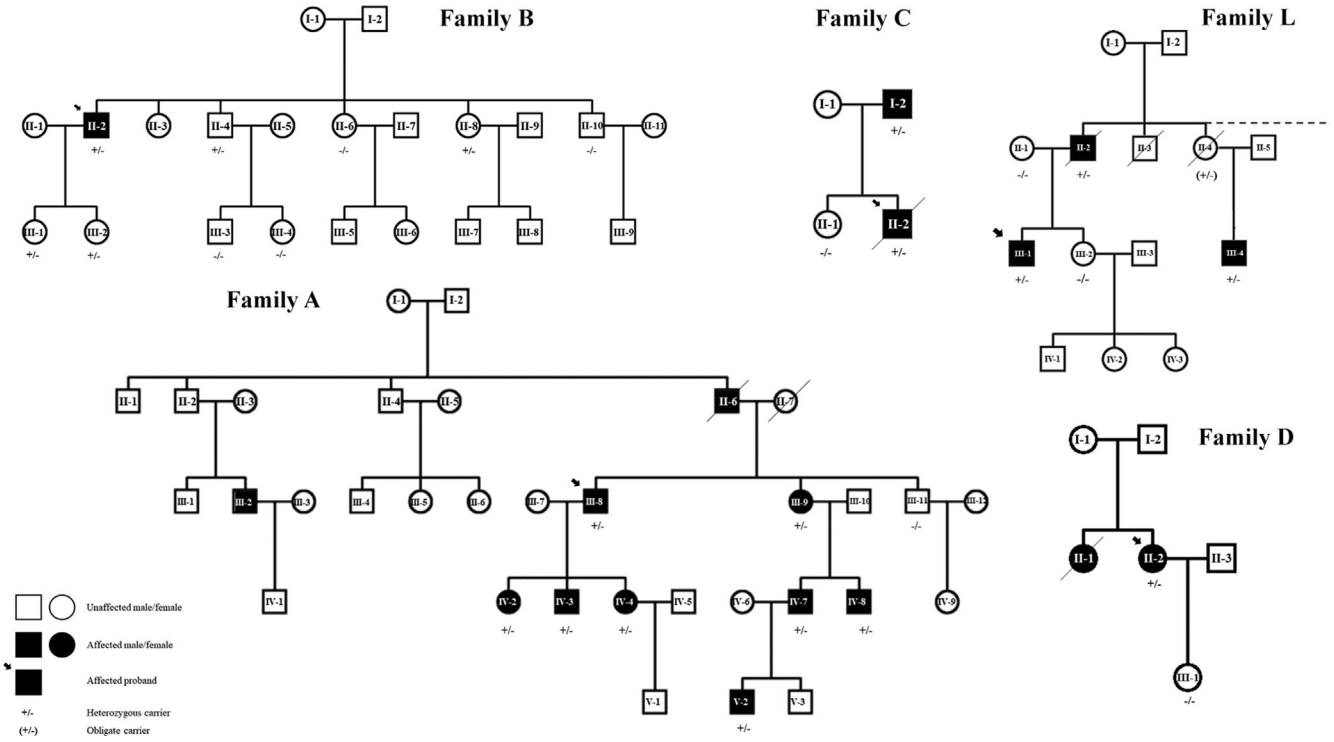

Figure 1 Pedigrees of families with filamin-C *(FLNC)* variants. *Squares* indicate males; *circles* indicate females; *slashes* indicate deceased individuals; *black symbols* indicate *in vivo* or postmortem diagnosis of *FLNC* cardiomyopathy; (+/–) indicates heterozygous carrier; *arrows* indicate the index case in each family.

composite endpoint, patients who did more frequently had low QRS voltages ($P = .013$), inferolateral/lateral TWI ($P = .010$), and LV LGE/fibrosis ($P = .033$). Frequent PVBs, LV dilation, and LVEF <35% were not associated with the SCD composite endpoint ($P = .116$, $P = .804$, and $P = .835$, respectively) (Figure 4). Results of univariate and multivariable logistic regression analyses focused on clinical predictors of the SCD composite endpoint are given in Supplemental Table 2.

FLNC variants protein domains analysis

In total, 64 unique different variants, 54 radical and 10 missense (Supplemental Table 3), were described in 160 *FLNC*+ carriers (144 *FLNC-R* and 16 *FLNC-M*) for which gene region localization was evaluated (Figure 5 and Supplemental Figure 2). About one-half of these variants (32/64 [50%]) were clustered on the ROD1 domain; the remaining 32 were distributed all along FLNC. Specifically, 11 were localized between N-terminal actin-binding

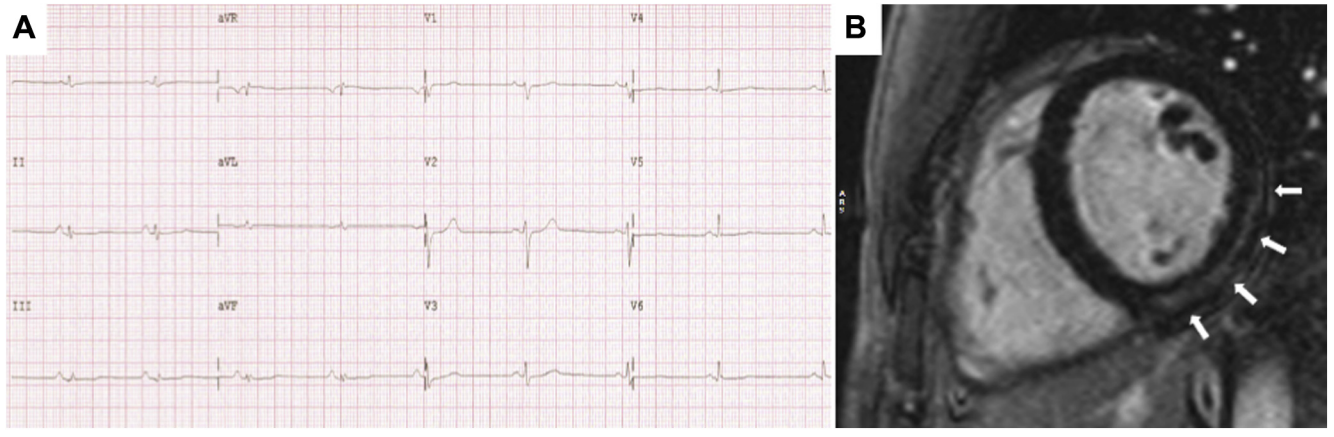

Figure 2 Electrocardiographic and cardiac magnetic resonance (CMR) features of Family L proband (III:1). **A:** Basal electrocardiogram showing low voltages in limb leads and flattened T waves in the inferolateral leads. **B:** Postcontrast CMR images in short-axis view showing normal left ventricular (LV) cavity size and subepicardial late gadolinium enhancement *(white arrows)* involving the LV inferolateral wall.

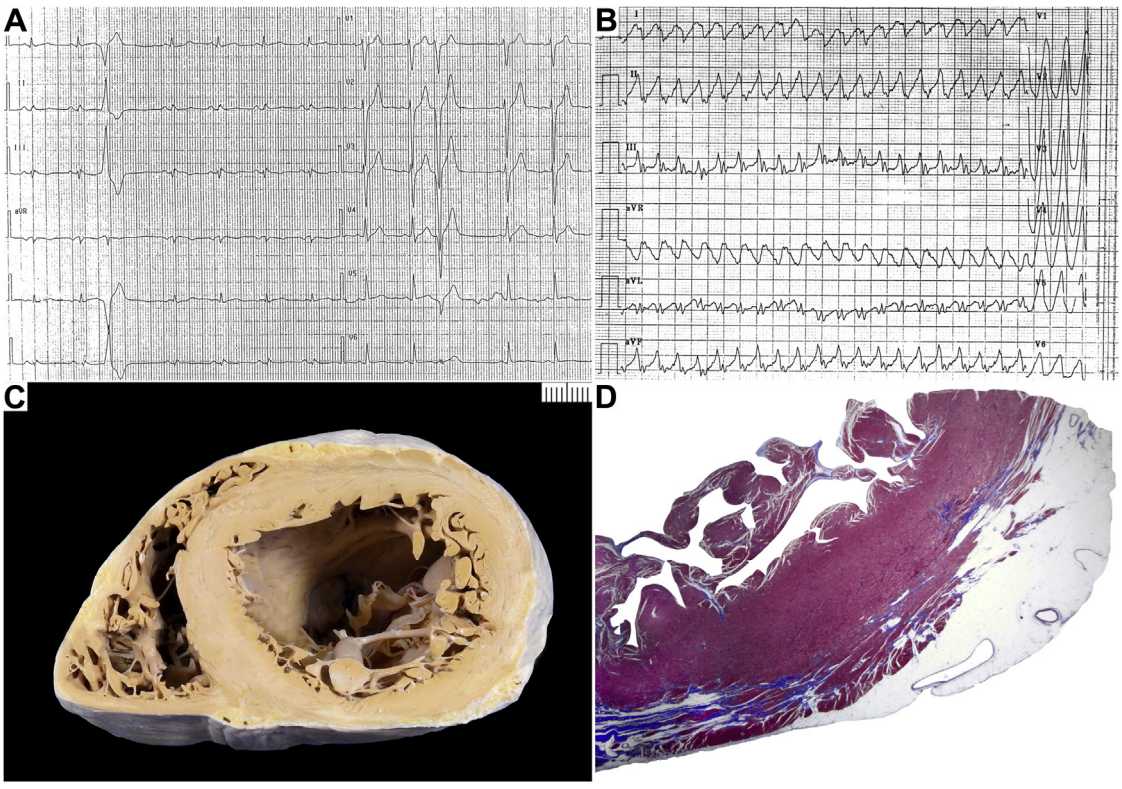

Figure 3 Electrocardiographic (ECG), arrhythmic, and histopathological features of Family L patient (III:4) undergoing heart transplantation. **A:** Basal ECG showing low voltages in limb leads and flattened T waves in the inferior leads. **B:** ECG strip recorded during sustained ventricular tachycardia. **C:** Short-axis section of the heart showing focal thinning of the posterior and lateral left ventricular free walls with whitish appearance. **D:** Panoramic histologic section of the posterolateral wall showing subepicardial and midmural fibrofatty replacement (trichrome stain).

domains, 20 on the ROD2 domain, and 1 variant in the C-terminal dimerization domain. Of note, none of the variants was located in the hinge domains, and when considering the length of each domain no significant distribution was observed. Further description of this analysis is given in the Supplemental Results.

Discussion

Only a handful of studies have investigated cardiomyopathy-related *FLNC* variants in single cases/families and smaller/larger cohorts.[5–10] However, large-scale studies are totally missing and as such fail to determine specific diagnostic clues associated with a specific disease phenotype (either ACM- or DCM-related). This is the first independent replication study of *FLNC* rare variants that aimed to pool data from all available studies matching our stringent quality criteria.

The main results of our study are as follows. (1) Novel *FLNC* variants were detected in about 4% of gene-elusive ACM index cases with late disease onset (after 40 years). (2) The most common ACM disease phenotype was the left-dominant one, identified in more than one-half of *FLNC*-associated ACM, but right-dominant forms also were also described. (3) The clinical phenotype of patients with *FLNC*-associated cardiomyopathy was characterized by ECG abnormalities such as low QRS voltages and inferolateral/lateral TWI, frequent and complex VAs, and extensive nonischemic LV LGE/myocardial fibrosis on CMR or postmortem analysis. (4) Risk factors associated with SCD were the presence of ECG inferolateral/lateral TWI and LV LGE/fibrosis, but not LV dilation or severe LV systolic dysfunction.

FLNC-related cardiomyopathy: prevalence and disease penetrance

In this study, among 270 genotype-negative ACM index cases, 12 *FLNC* novel variants (4.4%) were identified (7 radical, 5 variants of unknown significance). This finding, combined with that of other case series reporting *FLNC* mutations in 3% to 7.5% of gene-elusive ACM patients,[5,6] confirmed the link between *FLNC* and ACM, and the possible role of *FLNC* variants in the pathogenesis of the disease. Filamin C is an actin cross-linking protein localized in the intercalated discs of both cardiac and skeletal muscle cells, which is encoded by a 48-exon gene situated in chromosome 7q32-35.[18] Its main function is the binding of actin rods in the adherens junction, which are the structures that, together with desmosomes, contribute to maintenance of the cellular integrity and force transduction of tissues exposed to mechanical stress.[18,19] Mutations in *FLNC* may affect filamin C protein function and produce adherens junction abnormalities, which, as occurs with abnormal desmosomes,[1,3] may confer a predisposition over time to

Table 2 Clinical characteristics of *FLNC* variant carriers of the Padua cohort

Family	Index case	Sex	Radical variant	Age (y)	ECG low QRS voltages	ECG TWI V_1–V_3	ECG TWI inferolateral/lateral leads	Major arrhythmias/ 24-h PVB count	Abnormal echo results	CMR LVEF (%)	CMR LV dilation	CMR RVEF (%)	CMR RV dilation	CMR LV LGE	ACM phenotype
A, III-8	+	M	+	63	+	–	+	sVT	+	45	–	N*	–	N/A	LD ACM
A, III-9	–	F	+	68	–	–	–	–	–	69	–	60	–	+	LD ACM
A, IV-2	–	F	+	32	–	–	–	–	–	55	–	56	–	–	–
A, IV-3	–	M	+	31	–	–	–	673	–	58	–	55	–	+	LD ACM
A, IV-4	–	F	+	36	–	–	–	–	–	55	–	52	–	–	–
A, IV-7	–	M	+	43	–	–	–	1133	–	59	–	59	–	+	LD ACM
A, IV-8	–	M	+	35	+	–	+	sVT	+	49	–	69	–	+	LD ACM
A, V-2	–	M	+	12	–	–	–	–	–	60*	–	N*	–	N/A	–
B, II-2	+	M	–	65	+	–	–	sVT	+	70	–	43	–	–	RD ACM
B, II-4	–	M	–	67	–	–	–	N/A	–	65*	–	N*	–	N/A	–
B, II-8	–	F	–	52	–	–	–	N/A	–	71	–	65	–	–	–
B, III-1	–	F	–	39	–	–	–	N/A	–	64*	–	N*	–	N/A	–
B, III-2	–	F	–	33	+	–	–	N/A	–	63	–	65	–	–	–
C, II-2	+	M	+	17	N/A	N/A	N/A	SCD	N/A	N/A	N/A	N/A	N/A	N/A	LD ACM
C, I-2	–	M	+	63	–	–	–	sVT	–	52	–	56	–	+	LD ACM
D	+	F	+	66	–	+	+	Rare	+	66	–	45	–	–	RD ACM
E	+	M	–	40	–	+	–	4439	+	60	–	16	+	–	RD ACM
F	+	F	–	43	–	+	+	Aborted SCD	+	66	–	33	+	+	BIV ACM
G	+	M	+	57	+	–	–	nsVT	+	46	–	49	–	+	BIV ACM
H	+	M	–	56	+	+	–	sVT	+	55	–	37	+	–	RD ACM
I	+	M	–	64	+	+	+	sVT	+	44	+	35	+	+	BIV ACM
L, III-1	+	M	+	43	+	–	–	nsVT	–	58	–	61	–	+	LD ACM
L, III-4	+	M	+	25	+	–	+	sVT	+	30*	+	N*	–	N/A	LD ACM
M	+	F	+	35	–	–	+	nsVT	+	39	+	56	–	+	LD ACM
N	+	F	+	42	+	–	+	nsVT	+	34	+	44	–	+	BIV ACM

+ = positive/present; – = negative/absent; ACM = arrhythmogenic cardiomyopathy; BIV = biventricular; CMR = cardiac magnetic resonance; ECG = electrocardiogram; echo = echocardiogram; F = female; FLNC = filamin-C; LD = left dominant; LGE = late gadolinium enhancement; LV = left ventricle; LVEF = left ventricular ejection fraction; M = male; N = normal; N/A = not available; nsVT = nonsustained ventricular tachycardia; PVB = premature ventricular beat; RD = right dominant; RV = right ventricle; RVEF = right ventricular ejection fraction; SCD = sudden cardiac death; sVT = sustained ventricular tachycardia; TWI = T-wave inversion.

*Assessed with echocardiogram.

Table 3 Demographic and clinical profile of patients with *FLNC* cardiomyopathy (pooled analysis)

	Overall sample (n = 145)	ACM (n = 60)	DCM (n = 85)	*P* value
Age (y)	43 ± 16	48 ± 17	40 ± 14	.017
≥35 y	101 (70)	45 (75)	56 (66)	.274
Male sex	89 (61)	40 (67)	49 (58)	.302
Proband	81 (56)	33 (55)	48 (57)	.867
Radical variant	133 (92)	49 (82)	84 (99)	<.001
Electrocardiographic characteristics				
Low (<0.5 mV) QRS voltages in limb leads	40/109 (37)	19/46 (41)	21/63 (33)	.426
TWI in V_1–V_3 ± V_4	11/120 (9)	9/56 (16)	2/64 (3)	.023
TWI in inferolateral/lateral leads	29/120 (24)	19/56 (34)	10/64 (16)	.032
Arrhythmic history				
Frequent PVB (>500/24 h)	94/119 (79)	41/53 (77)	53/66 (80)	.821
Sustained VT	30/107 (28)	15/53 (28)	15/54 (28)	1
Cardiac imaging findings				
LV dilation	69/131 (53)	15/57 (26)	54/74 (73)	<.001
LVEF (%)	42 ± 14	51 ± 11	36 ± 12	<.001
LVEF ≤35%	43/137 (29)	5/57 (9)	38/80 (48)	<.001
RV involvement	36/117 (31)	19/57 (33)	17/60 (28)	.689
LV LGE	50/67 (75)	38/44 (86)	12/23 (52)	.004
Full heart histopathological analysis				
LV fibrosis	11/12 (92)	7/7 (100)	4/5 (80)	.217
Outcome				
SCD composite endpoint	28 (19)	10 (17)	18 (21)	.498
Cardiac transplantation/heart failure death	8 (6)	1 (2)	7 (8)	.088

Values are given as n (%) or mean ± SD unless otherwise indicated.
DCM = dilated cardiomyopathy; VT = ventricular tachycardia; other abbreviations as in Table 2.

disruption of intercellular anchoring, eventually leading to myocyte detachment, cell death, and subsequent fibrofatty replacement. Recent experimental data demonstrated the detrimental effects of filamin C loss in morphology and function in *FLNC* cardiomyocyte-specific knockout models.[20] Based on these findings, it seems reasonable to consider *FLNC* among causative ACM-related genes to be included in clinical gene panels when ACM is investigated.[4]

Cascade genetic screening performed in 5 families of the Padua *FLNC* cohort identified 13 more *FLNC* carriers among family members, 5 of whom fulfilled diagnostic criteria for ACM. Accordingly, ACM disease penetrance was estimated about 70% and was critically age dependent due to a net increase of ACM diagnosis in ≥35-year-old *FLNC* carriers. As such, *FLNC* ACM seems to become clinically overt later in lifetime than what it is expected from desmosomal variant carriers (<35 years).[3,13] This finding has crucial implications in clinical practice because it suggests the need for longer follow-up, especially in family members, to identify disease signs and allow prompt adoption of preventive measures and treatments.

FLNC variant–associated ACM

We found that the most frequent ACM disease phenotype associated with *FLNC* variants was the left-dominant phenotype, diagnosed in >50% of *FLNC*-associated ACM, in keeping with recent studies.[5,6,21] ACM patients from the Padua cohort with left-dominant or biventricular phenotype exhibited nonischemic LGE/fibrosis affecting the subepicardial or midmyocardial layers of the LV free wall at CMR (Figure 2). However, in contrast with recent studies,[21] the so-called "ring-like" pattern, that is, circumferential LGE of the LV free wall and septum in the short-axis view, was observed only in 2 of 11 patients (Family A, IV-8; Family N). This observation together with common identification of a right-dominant ACM phenotype (4/18) in the Padua cohort suggests high variability of *FLNC* variants in disease expressivity.

Among the 12 living ACM patients with LV disease, 50% did not have LV systolic dysfunction and regional LV wall-motion abnormalities (40% of echocardiographic results were unremarkable). This is a key diagnostic aspect and must be considered when dealing with ACM patients, especially family members. In fact, a limited extent of fibrofatty scars to subepicardial or midmyocardial layers of LV may not be sufficient to alter its regional (and global) function and can be undetectable by echocardiography.[22] For this reason, CMR should be always offered to *FLNC*+ family members, irrespective of echocardiographic results.

Clinical phenotype of *FLNC* variant–associated ACM and DCM

FLNC variants have been linked to extracardiac conditions such as distal and myofibrillar skeletal myopathy[23] and other cardiac disorders.[24,25] In our study, we focused on the association of *FLNC* mutations with the wide phenotypic spectrum of ACM, which recognizes not only right-dominant but also biventricular and left-dominant variants.[12] However,

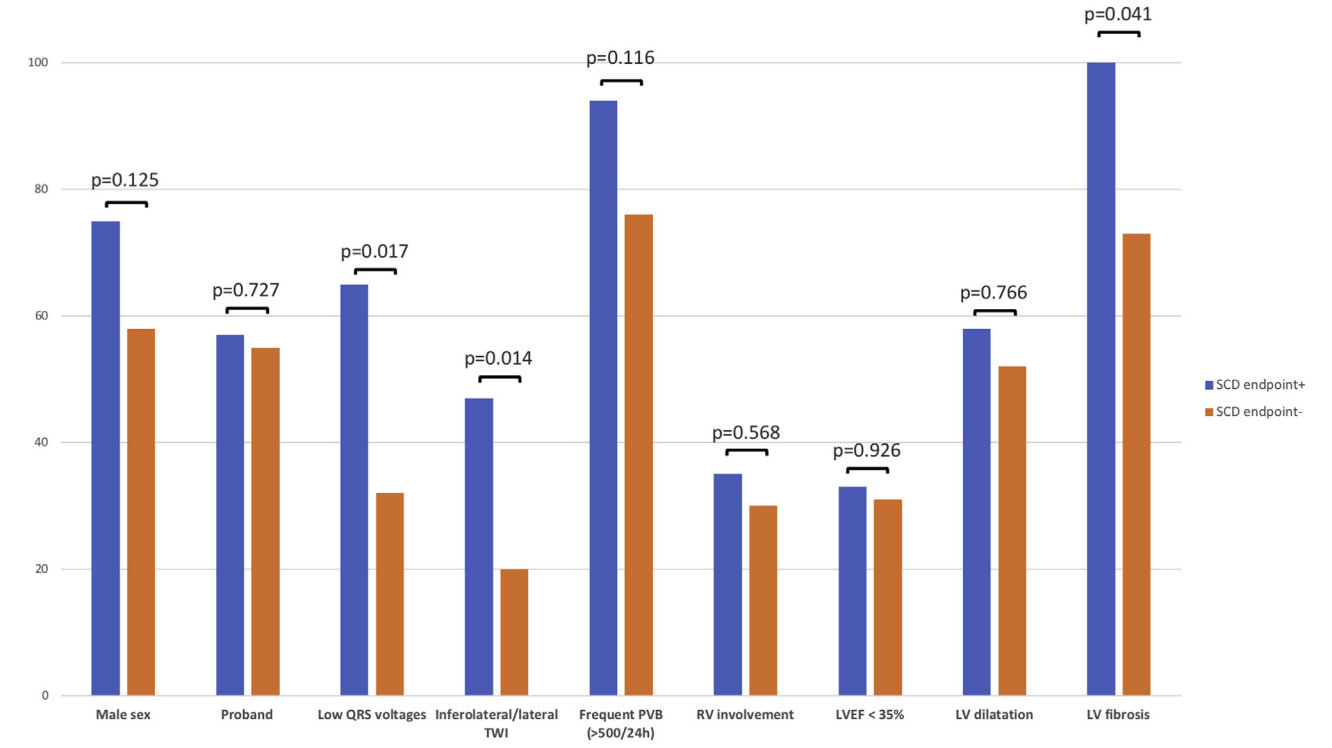

Figure 4 Bar chart showing clinical differences in filamin-C *(FLNC)* cardiomyopathy patients who reached or did not reach the sudden cardiac death (SCD) composite endpoint. LV = left ventricle; LVEF = left ventricular ejection fraction; PVB premature ventricular beat; RV = right ventricle; TWI = T-wave inversion.

these last 2 ACM phenotypes can overlap with that of DCM due to the possible occurrence in both conditions of myocardial scarring, LV systolic dysfunction, and malignant VAs, making differential diagnosis sometimes challenging.[22] For this reason, our pooled analysis included *FLNC* patients with either ACM or DCM phenotype in order to investigate differences and similarities of clinical features involved in the diagnosis, management, and risk stratification strategies. Our data showed that low QRS voltages on ECG, possibly reflecting loss of viable myocardium, were detected in 37% of *FLNC* patients. Electrical instability, represented by frequent PVBs and sustained ventricular tachycardia, was frequently observed, with no significant differences between the 2 phenotypes. LV nonischemic myocardial fibrosis evidenced by CMR was common (75%) and was significantly more prevalent in ACM patients. Noteworthy, due to the dominant LV involvement, RV endomyocardial biopsy can be negative in terms of fibrofatty replacement detection but reveals cardiomyopathic changes that are shared by ACM and DCM.

FLNC and SCD

SCD events have been frequently reported in studies involving *FLNC* cohorts.[5–10,24] In DCM and ACM populations, SCD occurred as the presenting symptom in 5% of cases and during follow-up in 15% of cases.[6] However, risk factors for SCD have not further investigated. In our

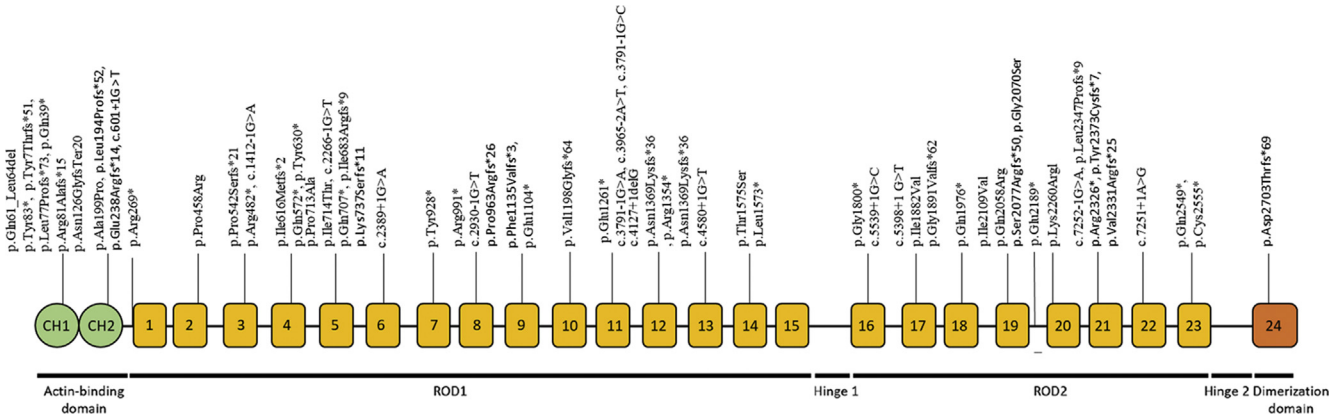

Figure 5 Variants localization in the filamin-C *(FLNC)* gene.

study, we showed that the detection of low QRS voltages in limb leads, ECG TWI in inferolateral/lateral leads, and LV LGE/fibrosis can help identify *FLNC* patient at higher risk for SCD. Importantly, as recently demonstrated for patients with desmoplakin mutations,[26] LVEF <35% also seems not to be a marker of higher SCD risk in *FLNC* patients. Larger multicentric *FLNC* cohorts are needed to extend and improve SCD risk stratification in these patients.

Study limitations

This study is limited by the small number of recruited *FLNC* missense variant carriers, which in part could be linked to the low frequency of *FLNC* variants in ACM and DCM populations and to the high genetic heterogeneity that characterizes these disorders. Indeed, variant types and locations were unable to explain phenotypic variability, and cosegregation studies were limited due to the small size of families.

Conclusion

FLNC-associated cardiomyopathy is characterized by late onset and mostly left-dominant phenotype. Typical ECG abnormalities consist of low QRS voltages and inferolateral/lateral TWI and frequent and complex VAs. Nonischemic LV scar is detectable by CMR or postmortem analysis. The presence of low QRS voltages in limb leads, inferolateral/lateral TWI, and LV LGE/fibrosis, but not LV dilation or severe systolic dysfunction, is associated with SCD.

Appendix

Supplementary data

Supplementary data associated with this article can be found in the online version at https://doi.org/10.1016/j.hrthm.2021.09.029.

References

1. Corrado D, Basso C, Judge DP. Arrhythmogenic cardiomyopathy. Circ Res 2017;121:784–802.
2. Basso C, Thiene G, Corrado D, Angelini A, Nava A, Valente M. Arrhythmogenic right ventricular cardiomyopathy: dysplasia, dystrophy, or myocarditis? Circulation 1996;94:983–991.
3. Pilichou K, Thiene G, Bauce B, et al. Arrhythmogenic cardiomyopathy. Orphanet J Rare Dis 2016;11:33.
4. James CA, Jongbloed JDH, Hershberger RE, et al. An international evidence based reappraisal of genes associated with arrhythmogenic right ventricular cardiomyopathy (ARVC) using the ClinGen Framework. Circ Genom Precis Med 2021;14:e003273.
5. Ortiz-Genga MF, Cuenca S, Dal Ferro M, et al. Truncating FLNC mutations are associated with high-risk dilated and arrhythmogenic cardiomyopathies. J Am Coll Cardiol 2016;68:2440–2451.
6. Hall CL, Akhtar MM, Sabater-Molina M, et al. Filamin C variants are associated with a distinctive clinical and immunohistochemical arrhythmogenic cardiomyopathy phenotype. Int J Cardiol 2020;307:101–108.
7. Oz S, Yonath H, Visochyk L, et al. Reduction in filamin C transcript is associated with arrhythmogenic cardiomyopathy in Ashkenazi Jews. Int J Cardiol 2020;317:133–138.
8. Brun F, Gigli M, Graw SL, et al. FLNC truncations cause arrhythmogenic right ventricular cardiomyopathy. J Med Genet 2020;57:254–257.
9. Begay RL, Graw SL, Sinagra G, et al. Filamin C truncation mutations are associated with arrhythmogenic dilated cardiomyopathy and changes in the cell-cell adhesion structures. JACC Clin Electrophysiol 2018;4:504–514.
10. Akhtar MM, Lorenzini M, Pavlou M, et al. Association of left ventricular systolic dysfunction among carriers of truncating variants in filamin C with frequent ventricular arrhythmia and end-stage heart failure. JAMA Cardiol 2021;6:891–901.
11. Marcus FI, McKenna WJ, Sherrill D, et al. Diagnosis of arrhythmogenic right ventricular cardiomyopathy/dysplasia: proposed modification of the task force criteria. Circulation 2010;121:1533–1541.
12. Corrado D, Perazzolo Marra M, Zorzi A, et al. Diagnosis of arrhythmogenic cardiomyopathy: the Padua criteria. Int J Cardiol 2020;319:106–114.
13. Rigato I, Bauce B, Rampazzo A, et al. Compound and digenic heterozygosity predicts lifetime arrhythmic outcome and sudden cardiac death in desmosomal gene-related arrhythmogenic right ventricular cardiomyopathy. Circ Cardiovasc Genet 2013;6:533–542.
14. Cipriani A, Zorzi A, Sarto P, et al. Predictive value of exercise testing in athletes with ventricular ectopy evaluated by cardiac magnetic resonance. Heart Rhythm 2019;16:239–248.
15. Basso C, Aguilera B, Banner J, et al. Association for European Cardiovascular Pathology. Guidelines for autopsy investigation of sudden cardiac death: 2017 update from the Association for European Cardiovascular Pathology. Virchows Arch 2017;471:691–705.
16. Thompson TG, Chan YM, Hack AA, et al. A muscle-specific sarcoglycan interacting protein. J Cell Biol 2000;148:115–126.
17. Whiffin N, Minikel E, Walsh R, et al. Using high-resolution variant frequencies to empower clinical genome interpretation. Genet Med 2017;19:1151–1158.
18. Mao Z, Nakamura F. Structure and function of filamin C in the muscle Z-disc. Int J Mol Sci 2020;21:2696.
19. Rampazzo A, Calore M, van Hengel J, van Roy F. Intercalated discs and arrhythmogenic cardiomyopathy. Circ Cardiovasc Genet 2014;7:930–940.
20. Zhou Y, Chen Z, Zhang L, et al. Loss of filamin C is catastrophic for heart function. Circulation 2020;141:869–871.
21. Augusto JB, Eiros R, Nakou E, et al. Dilated cardiomyopathy and arrhythmogenic left ventricular cardiomyopathy: a comprehensive genotype-imaging phenotype study. Eur Heart J Cardiovasc Imaging 2020;21:326–336.
22. Cipriani A, Bauce B, De Lazzari M, et al. Arrhythmogenic right ventricular cardiomyopathy: characterization of left ventricular phenotype and differential diagnosis with dilated cardiomyopathy. J Am Heart Assoc 2020;9:e014628.
23. Furst DO, Goldfarb LG, Kley RA, Vorgerd M, Olive M, van der Ven PF. Filamin C-related myopathies: pathology and mechanisms. Acta Neuropathol 2013;125:33–46.
24. Gómez J, Lorca R, Reguero JR, et al. Screening of the filamin C gene in a large cohort of hypertrophic cardiomyopathy patients. Circ Cardiovasc Genet 2017;10:e001584.
25. Tucker NR, McLellan MA, Hu D, et al. Novel mutation in FLNC (filamin C) causes familial restrictive cardiomyopathy. Circ Cardiovasc Genet 2017;10:e001780.
26. Smith ED, Lakdawala NK, Papoutsidakis N, et al. Desmoplakin cardiomyopathy, a fibrotic and inflammatory form of cardiomyopathy distinct from typical dilated or arrhythmogenic right ventricular cardiomyopathy. Circulation 2020;141:1872–1884.