A Novel Calsequestrin 2 Deletion Causing Catecholaminergic Polymorphic Ventricular Tachycardia and Sudden Cardiac Death

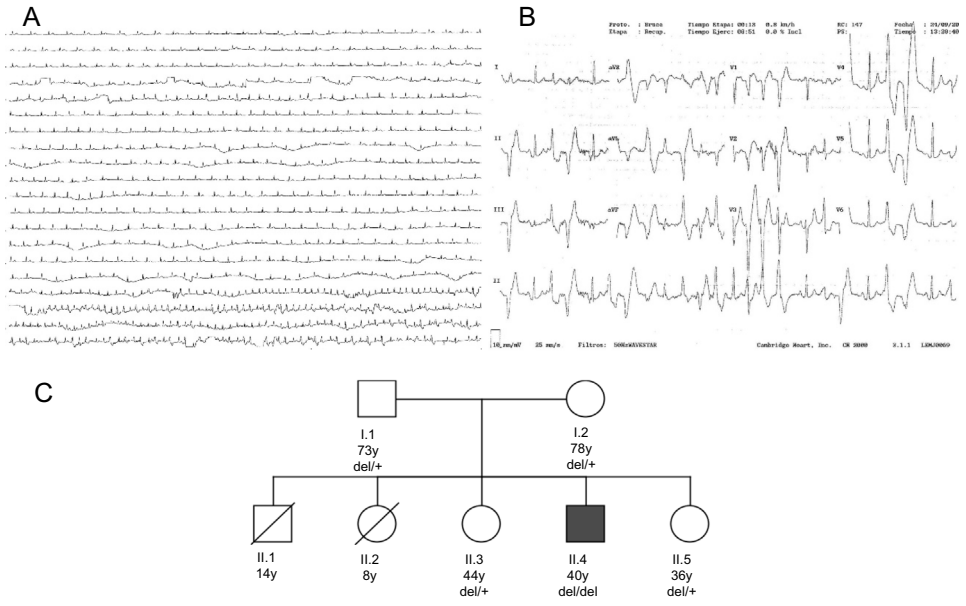
Una nueva delección de calsequestrina 2 que causa taquicardia ventricular polimórfica catecolaminérgica y muerte súbita cardiaca

To the Editor,

We present the case of a 40-year-old man with a history of syncope since childhood. He had been treated with carteolol 2.5 mg bid from the age of 10 years due to syncope and ventricular arrhythmia. When he was aged 17 years, the beta-blocker was changed and nadolol was started at 80 mg qd, which the patient voluntarily discontinued. He was admitted to hospital due to syncope while he was walking. A strong family history of sudden cardiac death was observed: a brother and a sister suddenly died at the age of 8 and 14 years, respectively. The parents were apparently healthy and the grandparents died in old age from tumors. The results of laboratory tests, electrocardiography, QT interval, echocardiographic examination, and cardiac magnetic resonance imaging were normal. A nonsustained polymorphic ventricular tachycardia (PVT) was detected in electrocardiogram recordings. Treadmill testing revealed nonsustained PVT during peak effort that disappeared during rest (Figure 1A and Figure 1B). A diagnosis of catecolaminergic polymorphic ventricular tachycardia (CPVT) was made due to an exercise-induced PVT in the presence of a structurally normal heart and normal electrocardiogram. Beta-blocker treatment was restarted and a treadmill test was repeated. An implantable cardioverter-defibrillator with long delays before shock delivery was implanted due to persistence of nonsustained PVT during exercise. During follow-up, flecainide was added at a dose of 100 mg bid to beta-blocker treatment due to appropriate shocks. After 6 months follow-up, ventricular arrhythmia control was notably improved with no sustained episodes.

A genetic test was performed to confirm the diagnosis.[1–3] We collected a peripheral blood sample from the index case and 4 relatives (Figure 1C). Unfortunately, biological samples from deceased relatives were not available. Massive sequencing for 28 genes associated with arrhythmogenic disease was performed in the index case.[4]

Sequencing showed no causal single nucleotide variant but did show the absence of the sequence for the last 3 exons of the *CASQ2* gene. Long-range polymerase chain reaction was conducted to amplify a fragment of approximately 18 kb, where the deletion is located. The results confirmed the presence of a fragment of approximately 13 kb in the index case and a fragment of approximately 18 kb in a control sample. Long-range polymerase chain reaction product of the index case was sequenced with the Ion Proton System (Thermo Fisher Scientific). The results demonstrated the presence of an approximately 5 kb deletion causing the loss of



Patient	Symptoms	Age at symptom onset, y	Diagnosis	Treatment
I.1	-	-	-	-
I.2	-	-	-	-
II.1	Syncopal events related to emotional stress	?	Sudden unexpected death	-
II.2	Syncopal events	?	Sudden unexpected death	-
II.3	-	-	-	-
II.4	Syncopal events	10	CPVT	ICD, beta-blocker, flecainide
II.5	Syncopal events	33	-	Beta-blocker

Figure 1. A: ECG telemetry recording showing highly frequent ventricular ectopy (bottom). B: 12-lead ECG during treadmill testing with runs of PVT. C: family pedigree. The genotype for the *CASQ2* deletion is indicated. CPVT, catecholaminergic polymorphic ventricular tachycardia; ECG, electrocardiogram; ICD, implantable cardioverter-defibrillator.

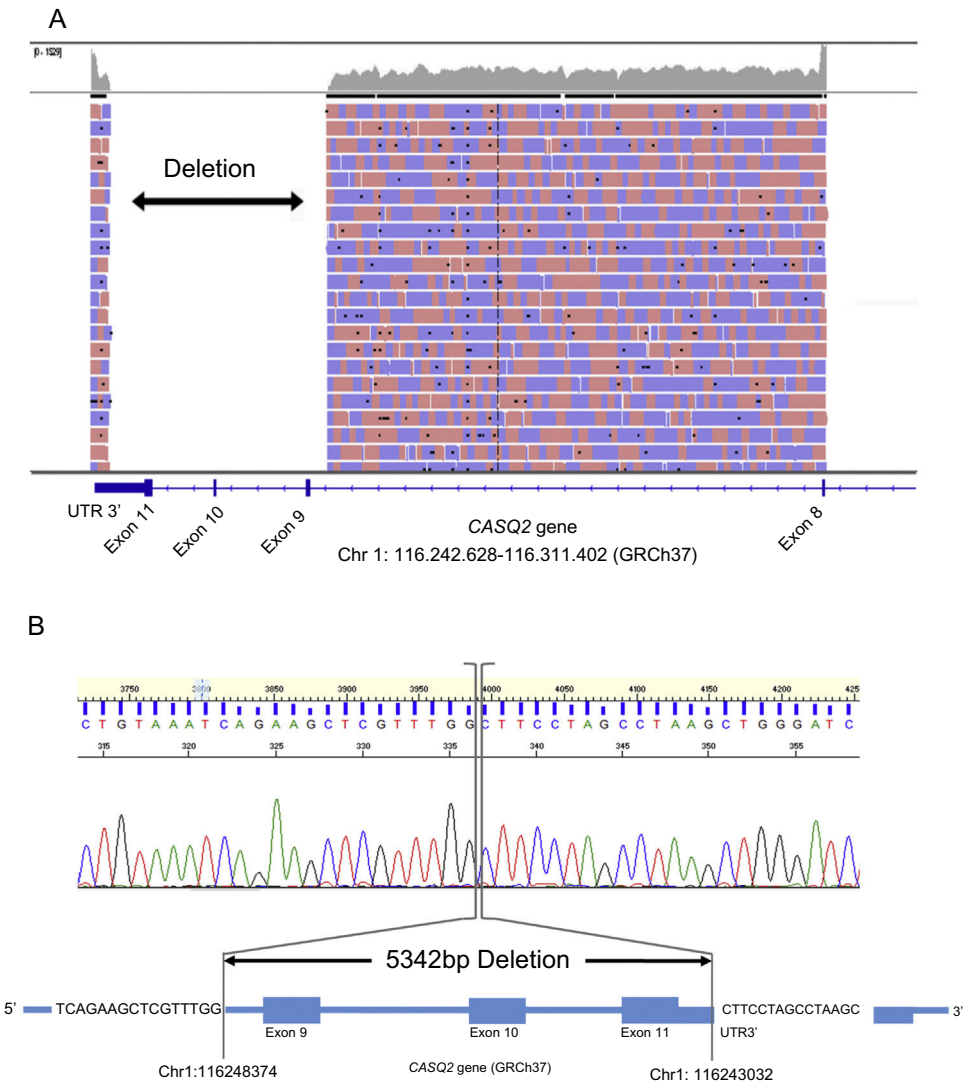


Figure 2. Sequencing results for the *CASQ2* gene. A: Integrative Genomics Viewer (IGV) visualization for long-range PCR product of the index case. B: electropherogram and representation of the exact location of the deletion.

exons 9, 10, and 11 of the *CASQ2* gene (Figure 2). The exact location of the deletion was confirmed by using specific polymerase chain reaction and Sanger sequencing (NM_001232:c.839-461_*830del).

Our family index case showed a homozygote deletion classified as pathogenic according to the American College of Medical Genetics (ACMG),[5] confirming the CPVT diagnosis, whereas unaffected family members were heterozygous carriers of the deletion (Figure 1C). One of the sisters had recently experienced syncope of unknown cause. Based on the hypothesis of a higher risk of arrhythmias associated with heterozygous variants, she was treated with beta-blockers.

Catecolaminergic PVT is an inheritable arrhythmogenic disorder characterized by adrenergic-induced bidirectional ventricular tachycardia or PVT. Two genetic types have been identified: a dominant variant due to mutations in the cardiac ryanodine receptor gene *(RyR2)* and a rare recessive variant caused by mutations in the cardiac calsequestrin gene *(CASQ2)*.[1,3] Mutations in other genes such as *KCNJ2, ANK2, TRDN*, and *CALM1* have been identified in patients with clinical features similar to CPVT but it is not clear whether they are phenocopies of CPVT.[3] Most of the *CASQ2* gene mutations described are truncating or splicing genetic variants and to date there have been no reports of causal copy number variations. Although some copy number variations have been described in channelopathies, they are not common; in the specific case of CPVT, a deletion of exon 3 of *RyR2* has been described.

The clinical manifestations occur in the first decade of life and are triggered by physical activity or emotional stress. First-line therapy consists of exercise restriction and beta-blockers without sympathomimetic activity. Preliminary data suggest that flecainide reduces the ventricular arrhythmia burden in some patients and should be considered as an addition to beta-blockers.[3] Diagnosis is confirmed by genetic testing. Mass sequencing technologies allow rapid screening of single nucleotide or small indel variants. However, the presence of large deletions or insertions may go unnoticed, if we only take into account the lists of variants reported by variant calling programs. The presence of these insertions or deletions must be checked with specific programs, otherwise the sequencing results should be reviewed with genomic visors, checking the coverage of all target regions to be sequenced.[4]

This is the first reported case of a copy number variation as a cause of CVPT in a nonconsanguineous family with poor prognosis, with a severely affected index case carrying the variant in homozygosis and with 2 siblings who died suddenly at very early ages.

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Preconception Assessment of Women of Childbearing Age With Congenital Heart Disease

Valoración preconcepcional de mujeres en edad fértil con cardiopatías congénitas

To the Editor,

Congenital heart disease (CHD) affects approximately 1% of live newborns. Due to improvements in diagnosis and treatment, the vast majority reach adulthood. This large cohort of young adult "survivors" includes a high number of women of childbearing age.

Unfortunately, patients with CHD tend to underestimate the severity of their disease,[1] which is particularly concerning in women of childbearing age. The risks of complications during pregnancy and peripartum in this population depend on the underlying defect, the extent and severity of residual lesions, and comorbidities.[2] With this in mind, the clinical guidelines recommend that all women with congenital heart disease receive advice before conceiving.[3]

Few studies have assessed the perception of women of childbearing age with CHD regarding their heart disease and understanding of the risks,[4] their desire to have children, and their contraceptive options.[5] Our aim was to evaluate these key points with a detailed questionnaire.

A descriptive cross-sectional study was designed, recruiting all women aged 15 to 45 years undergoing follow-up at our adult CHD clinic. The protocol was reviewed and approved by the Ethics Committee of our hospital. The questionnaire included 19 questions for the patient and 8 for an immediate relative.

CHD was classified by severity as mild, moderate, or severe, following the recommended classification of the *Sociedad Española de Pediatría Extrahospitalaria y Atención Primaria* (Spanish Society of Outpatient and Primary Care Pediatrics). The cardiovascular risk of pregnancy was also classified according to the modified World Health Organization classification.[3]

Fifty-one (75%) of the 68 women who were initially identified completed the questionnaire. The types of CHD recorded were ventricular septal defect (n = 14), coarctation of the aorta (n = 5), univentricular heart/Fontan circulation (n = 5), bicuspid aortic valve (n = 4), pulmonary stenosis (n = 4), double outlet right ventricle (n = 4), tetralogy of Fallot (n = 3), partial atrioventricular canal (n = 3), subaortic stenosis (n = 3), prolapsed mitral valve (n = 2), repaired patent ductus arteriosus (n = 1), truncus arteriosus (n = 1), congenitally corrected transposition of the great arteries (n = 1), and ostium primum atrial septal defect (n = 1). In the first part of the questionnaire, the patients were asked to rate the severity of their heart disease: 35.3% responded mild; 45.1%, moderate; and 19.6%, severe. This differed significantly ($P = .001$) from the opinion of the cardiologists and relatives (Figure 1).

Regarding the risks of pregnancy, only 52.9% of the women reported having discussed the health risks of a potential pregnancy with their specialist. When asked to classify the risk associated with a potential pregnancy, 25.5% responded high; 33.3%, low; and 41.2% reported they did not know. Of those who said they did not know, most of the women were from the group deemed low-risk by the specialist (73%). In contrast, in the group of patients that considered pregnancy to be high-risk, the percentage considered high-risk according to the specialist was relatively high (83%) (Figure 2).

Of particular note was that 40% of the women expressed a desire to have children, their mean age being 29.6 ± 6.4 years. Only 11 of the 51 women (21.6%) had had a previous pregnancy, and they reported 9 planned pregnancies, 1 unexpected, and 5 unwanted pregnancies that were terminated (33.3% of the total). None reported that they were using contraception when they became pregnant. Only 44% of all the patients reported having received information from their pediatrician on contraceptive methods.