ESC European Society of Cardiology

European Journal of Preventive Cardiology (2024) **00**, 1–9
https://doi.org/10.1093/eurjpc/zwae389

FULL RESEARCH PAPER
Sports cardiology

Influence of age and sex on the diagnostic yield of inherited cardiac conditions in sudden arrhythmic death syndrome decedents

Belinda Gray [1,2,3,4†], Elijah R. Behr [1,2*†], Efstathios Papatheodorou[1,2], Athanasios Bakalakos[1,2,5,6], Hariharan Raju [1,2,7], Yanushi D. Wijeyeratne[1,2], Gherardo Finocchiaro[1,2], Aneil Malhotra[1,2,8], Nicola Whiffin [9,10], James S. Ware[9], Maria Tome Esteban[1,2], Mary N. Sheppard[11], Sanjay Sharma [1,2], and Michael Papadakis[1,2†]

[1]Cardiovascular and Genomics Institute, City St George's University of London, Cranmer Terrace, London SW17 0RE, UK; [2]Department of Cardiology, St George's University Hospitals NHS Foundation Trust, Blackshaw Rd, London SW17 OQT, UK; [3]Faculty of Medicine and Health, University of Sydney, Sydney, NSW, Australia; [4]Department of Cardiology, Royal Prince Alfred Hospital, Sydney, NSW, Australia; [5]Institute of Cardiovascular Science, University College London, London, UK; [6]Inherited Cardiovascular Disease Unit, St Bartholomew's Hospital, London, UK; [7]Faculty of Medicine, Health and Human Sciences, Macquarie University, Sydney, Australia; [8]Institute of Sport, Manchester Metropolitan University, Manchester, UK; [9]National Heart and Lung Institute, MRC Laboratory of Medical Sciences, Imperial College, London, UK; [10]Big Data Institute, Centre for Human Genetics, University of Oxford, Oxford, UK; and [11]Department of Cardiovascular Pathology, St George's University of London, London SW17 ORE, UK

Received 4 July 2024; revised 7 September 2024; accepted 14 November 2024; online publish-ahead-of-print 23 December 2024

Aims Sudden arrhythmic death syndrome (SADS) refers to a sudden death, which remains unexplained despite comprehensive post-mortem examination and a toxicological screen. We aimed to investigate the impact of age and sex on the overall diagnostic yield and underlying aetiology in decedents with SADS using a combined approach of familial evaluation (FE) and molecular autopsy (MA).

Methods and results Consecutive referrals to a single centre for FE only, MA only or both, following a SADS death were included. First-degree family members underwent comprehensive FE and decedents with post-mortem DNA were sequenced with a 36 cardiac gene panel for MA. A Bayesian framework for analysis was performed to identify associations. Among 760 SADS decedents (66% male; mean age 31 ± 12 years) the overall diagnostic yield for an inherited cardiac condition was 37% (32–42%) and 9% (6–12%) for FE and MA cohorts. In a subset where both FE and MA were performed the diagnostic yield was 45% (38–61%). The relative risk of an FE diagnosis of long QT syndrome (LQTS) or Catecholaminergic polymorphic ventricular tachycardia (CPVT) vs. remaining unexplained declined by 5.6% [RR 0.94 (0.91–0.98)] and by 11% [RR 0.89 (0.81–0.97)], for each year increase in age. Females were more likely to have a diagnosis by both FE [40% (34–45%) vs. 36% (31–41%)] and MA [15% (10–21%) vs. 6% (3–8%)]. Females [8.1% (4.1–13.4%)], were more likely to be diagnosed with LQTS than males [1.2% (0.2–2.7%)] in the MA cohort.

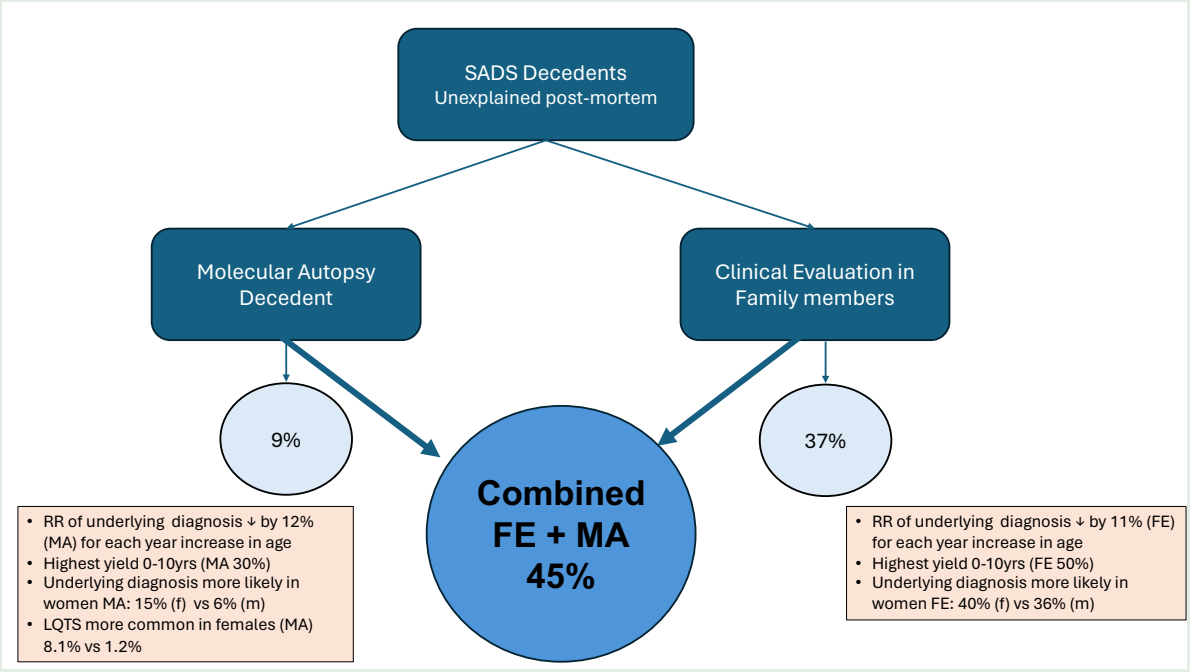
Conclusion After a SADS death, the diagnostic yield of comprehensive FE, MA, or both in an expert setting can be up to 45% with a combined approach. Females had higher diagnostic yield than males, most notable with LQTS. CPVT and LQTS diagnoses declined with increasing age. These data highlight the relative utility of FE and MA depending on age and sex for determining underlying diagnoses following SADS deaths.

Lay summary

- The diagnostic yield of comprehensive FE, MA, or both following a SADS death, in an expert setting can be up to 45%.
- Females are more likely to have an underlying inherited cardiac condition diagnosis following a SADS death.
- Younger age of death is associated with greater likelihood of LQTS and CPVT diagnosis with the highest yield in children and adolescents.
- Female sex in SADS decedents is associated with a higher yield of LQTS in MA.

* Corresponding author. Tel: +442087252994, Email: ebehr@sgul.ac.uk
† These authors contributed equally to the study.

Graphical Abstract



Keywords Unexplained sudden death • SADS • SCD • Age • Gender

Introduction

Sudden arrhythmic death syndrome (SADS) refers to a sudden death,[1,2] which remains unexplained despite comprehensive post-mortem examination and a toxicological screen.[3–5] Contemporary studies suggest that SADS may account for up to 40% of sudden deaths in young (<35 years) individuals.[5] Familial clinical evaluation (FE) and post-mortem genetic testing ['molecular autopsy' (MA)], have the potential to identify an inherited cardiac condition (ICC) in a significant proportion of decedents, with most diagnoses attributed to inherited arrhythmia syndromes, including long QT syndrome (LQTS), Brugada syndrome (BrS), and catecholaminergic polymorphic ventricular tachycardia (CPVT).[1]

The potential to identify underlying ICCs in asymptomatic family members, who may also be at risk of fatal arrhythmias, has led to updated guidelines recommending FE of first-degree relatives of the decedent and MA, following a SADS death, to prevent further tragic sudden deaths in a family.[6,7] Proposed clinical protocols and gene panels have differed between studies historically, leading to disparities in the reported diagnostic yield and the conditions identified in the decedents.[5,8,9] Irrespective of the decedent's demographics and circumstances of death, most families undergo a similar clinical evaluation for underlying ICCs. Though there are some studies reporting differences in baseline demographics for individuals with ICCs, such as a higher prevalence of CPVT in younger patients, no studies to date have reviewed the differences between yield of underlying diagnosis for SADS deaths, according to the age and sex of the decedent. A more personalized approach to the evaluation of SADS families may be more clinically applicable.

We investigated two consecutive cohorts of SADS decedents and their families who were referred to our clinic for evaluation. The objective of this study was to assess the impact of age and sex of the decedents on the diagnostic yield and underlying aetiology of SADS deaths using FE in first degree family members and MA in post-mortem deoxyribonucleic acid (DNA). The diagnostic yield for the combined approach was evaluated in a smaller subset of these cohorts who had undergone both diagnostic methods.

Methods

Case selection

SADS cases were defined as: (i) an unexplained death, (ii) in an individual aged 1–64 years, (iii) with no known prior cardiovascular disease, (iv) who died within 1 h of symptom onset, or an unwitnessed death with the decedent having been seen in good health within 24 h of death, (v) with no cause of death identifiable on comprehensive coronial and/or cardiac autopsy, and (vi) negative toxicology.[3,5,10] This retrospective cohort study included two different cohorts: (i) consecutive referrals for FE at the St George's and Lewisham Hospitals, United Kingdom, tertiary referral Cardiogenetics clinics between 2009 and 2017 and (ii) consecutive referrals for autopsy at the Cardiac Risk in the Young (CRY) Centre for Cardiac Pathology at Royal Brompton Hospital (2007–11) and St George's, University of London (2012–17) with DNA suitable for MA. Cases where cardiac autopsy identified a structural heart disease, such as cardiomyopathy or those with non-specific structural changes on cardiac autopsy, such as idiopathic left ventricular hypertrophy or idiopathic fibrosis were excluded from the study. Consent was obtained from decedent next of kin. The study was approved by St George's University of London ethics (#10/H0803/121, #17/LO/0747).

Familial evaluation

First-degree family members of SADS cases were assessed using a uniform investigation protocol to identify underlying ICCs as previously described.[10] Clinical history, physical examination, baseline electrocardiogram (ECG) with standard (4th intercostal space), and high-lead (2nd and 3rd intercostal

space) placement, echocardiogram and 24-h Holter monitoring were offered to all relatives. Exercise testing was performed in relatives over the age of 16 years, and where possible in younger patients at a paediatric unit. If there was suspicion of underlying structural heart disease, cardiac magnetic resonance imaging was performed. If no diagnosis was established family members were offered drug provocation testing using a class I antiarrhythmic agent [Ajmaline 1 mg/kg (maximum 100 mg) over 5 min]. Further investigations were undertaken at the respective physician's discretion.[11] Standard clinical diagnostic criteria for ICCs were used.[1,12,13] Brugada syndrome diagnoses were also stratified according to the Shanghai consensus diagnostic scoring system.[14] Decedents were allocated the highest score available in their family: definite/probable BrS (score ≥3.5); possible BrS (score 2–3); and low probability (<2).

Molecular autopsy

SADS decedents with appropriate post-mortem DNA samples (in most cases fresh spleen tissue) available were sequenced using next-generation sequencing. Sequencing was performed using the SureSelect system (Aligent Technologies, Santa Clara, CA, USA) or Illumina TruSight Cardio system (Illumina, San Diego, CA, USA). Variants were restricted to those with minor allele frequency <0.001 in genome aggregation database (gnomAD) and non-truncating variants in *TTN* were excluded. Rare variants in 36 major channelopathy and cardiomyopathy genes (see Supplementary material online, *Table S1*) validated to be associated with ICCs and available in the CardioClassifier software (http://www.cardioclassifier.org) were reviewed and manually curated (B.G.) for pathogenicity using the American College of Medical Genetics and Genomics (ACMG) criteria.[15–17] A pathogenic or likely pathogenic variant was deemed diagnostic of the cause of death. Loss of function *SCN5A* variants were deemed diagnostic of BrS.

Statistical analysis

All the variables in the final dataset were explored using graphics, summarized according to their nature and by diagnostic method in *Table 1*. The two cohorts (FE and MA) were analysed separately. There were 407 participants evaluated by FE, and 424 evaluated by MA. A subset of 71 was tested through both diagnostic methods and therefore was part of both cohorts in the analysis.

The outcome of a diagnosis by FE was defined as unexplained, or 'explained', consisting of either BrS, LQTS, CPVT, or cardiomyopathy, in families subjected to FE only, or by FE ± MA in the 71-patient subset subjected to both. The outcome of a diagnosis by MA was defined similarly in decedents subjected to MA only, *or* by MA ± FE in the same 71-patient subset. These

Table 1 Cohort characteristics

Demographic	Overall ($n = 760$)	FE cohort ($n = 336$)	MA cohort ($n = 353$)	Combined MA + FE cohort ($n = 71$)	
Sex					$P = 0.61$
Male	505 (66)	228 (68)	233 (66)	44 (62)	
Female	249 (32)	108 (32)	120 (34)	27 (38)	
Missing	6	4 (1)	2 (1)	–	
Age overall mean	31 ± 12	29 (12)[a]	33(12)[a]	30 (12)[a]	$P < 0.001$
		27(20, 36)[b]	31(23, 41)[b]	29 (22, 37)[b]	
		(1,64)[c]	(1, 64)[c]	(1, 52)[c]	
1–10 years	20 (3)	10	6	4	
11–20 years	154 (20)	86	56	12	
21–30 years	226 (30)	99	105	23	
31–40 years	192 (25)	83	91	18	
41–50 years	109 (14)	32	65	12	
51–64 years	52 (7)	19	30	3	
Missing	7 (0.9)	7	–	–	
Symptoms prior to death	182 (24)				n/a
Palpitations	58 (8)	33	17	8	
Syncope	83 (11)	42	33	8	
Chest pain	45 (6)	17	21	7	
Unknown-missing	97 (13)	–	–	–	
Family history of sudden death	111 (18)	60 (17)	42 (12)	9	n/a
Unknown-missing	141 (19)	–	–	–	
Number of first degree relatives evaluated	3 (2)[a]	2.9 (1.9)[a]	–	3.7 (1.6)	
Number of relatives with a diagnosis	0.6 (1)[a]	0.6 (1)[a]	–	0.6 (1)[a]	
Final cause of death					
Unexplained	578 (76)	215 (64)	324 (92)	39 (55)	$P < 0.0001$
Brugada syndrome	109 (14)	86 (26)	6 (1.6)	17 (24)	$P < 0.0001$
Long QT syndrome	41 (5)	22 (6.5)	12 (3.4)	7 (9.8)	$P = 0.04$
CPVT	13 (2)	5 (1.5)	6 (1.6)	2 (2.8)	$P = 0.73$
Cardiomyopathy	19 (3)	8 (2)	5 (1.4)	6 (8.4)	$P = 0.002$

FE, familial evaluation; MA, molecular autopsy; CPVT, catecholaminergic polymorphic ventricular tachycardia.
Data shown as mean (%) except: [a]mean (SD); [b]median (IQR); [c]range.

outcomes denoted the cause of death in SADS decedents. Continuous and categorical baseline characteristics were analysed using one way ANOVA and χ^2, respectively.

The analytical strategies evaluated the overall yield of FE- and MA-diagnoses using multinomial logistic models on an outcome defined by the cause of death by FE or MA assuming that the group that remained unexplained represented a clinical category. The yields were then investigated for the effects of sex and age, and then by both simultaneously. Circumstances of death were also assessed but incomplete data prevented meaningful analysis and are not presented. The analysis produced age and sex-dependent specific risks of each diagnosis for FE or MA. We opted for a Bayesian framework for statistical inference (non-informative priors were used throughout) using Markov chain Monte Carlo estimation methods which allowed estimation of the posterior distribution of the relative risk ratios (RRR) for the effect of age, sex, or both on the risk of one cause of death vs. another.

The posterior distribution of the estimates has been summarized by their means and their uncertainties were assessed by their corresponding 95% credible intervals (CIs). In the Bayesian framework, a 95% CI associated with a RRR which did not contain 1, was considered a significant result. This approach accommodated missing responses in the data under missing at random assumption. We also derived cause-specific predicted probabilities of a specific diagnosis and all collated 'explained' diagnoses and presented them overall, by age, sex and by both in tables and graphically. Age dependent FE- and MA-estimated yields and their uncertainty were plotted against the corresponding observed proportions in 10 year groups. Elementary data processing, summaries and appropriate tests have been executed using Stata 17 (StataCorp. 2021. *Stata Statistical Software: Release 17*. College Station, TX, USA: StataCorp LLC) but the core of the modelling work has been implemented in OpenBUGS (https://www.mrc-bsu.cam.ac.uk/software/bugs/).

Results

Cohort characteristics

Demographic and clinical characteristics for the SADS cohort are detailed in *Table 1*. A total of 760 SADS decedents were included in the study. There were 407 participants in the FE cohort, 424 in the MA cohort, and 71 in both cohorts. The majority were male (67% FE and 65% MA) with a mean age at death of 29 years in FE cohort and 32 years in MA cohort. The diagnostic flowchart of the study cohort is shown in *Figure 1*. The cohort's breakdown is shown in Supplementary material online, *Figure S1*.

Diagnostic yield and underlying cause of death

The estimated yield of explained FE-diagnoses is 37% (32 and 42%) based on the 407 patients who underwent FE and 149 receiving an explicit cause of death. The estimated yield of explained MA-diagnoses is 8.8% (6 and 12%) based on 424 patients who underwent MA and 35 receiving an explicit cause of death (*Figure 1*). There were 35 pathogenic and likely pathogenic variants (see Supplementary material online, *Table S2*) and 145 variants of uncertain significance identified in the cohort (see Supplementary material online, *Table S3*).

In the 71 decedents, who were evaluated with both the FE and MA modalities, a diagnosis was established in 32 equating to a diagnostic yield of 45% (95% CI; 38%, 61%); 26 (36.6%) diagnosed by *FE only*, 4 (5.6%) diagnosed *by MA only* and 2 (2.8%) diagnosed *by both modalities*. Sensitivity analysis showed no bias attributed to the combined cohort.

Impact of sex on the diagnostic yield of inherited cardiac conditions

An underlying diagnosis of an ICC was more likely to be identified in female SADS decedents compared with males by both FE [40% (34–45%) vs. 36% (31–41)], and MA [15% (10–20) vs. 6% (3–8%)] (*Figure 2A and B*). The increased likelihood of a diagnosis in female decedents was present across most ICCs (see Supplementary material online, *Table S4*) and was most evident with the diagnostic yield of LQTS on MA [8.1% (4.1–13.4%) vs. 1.2% (0.2–2.7%)].

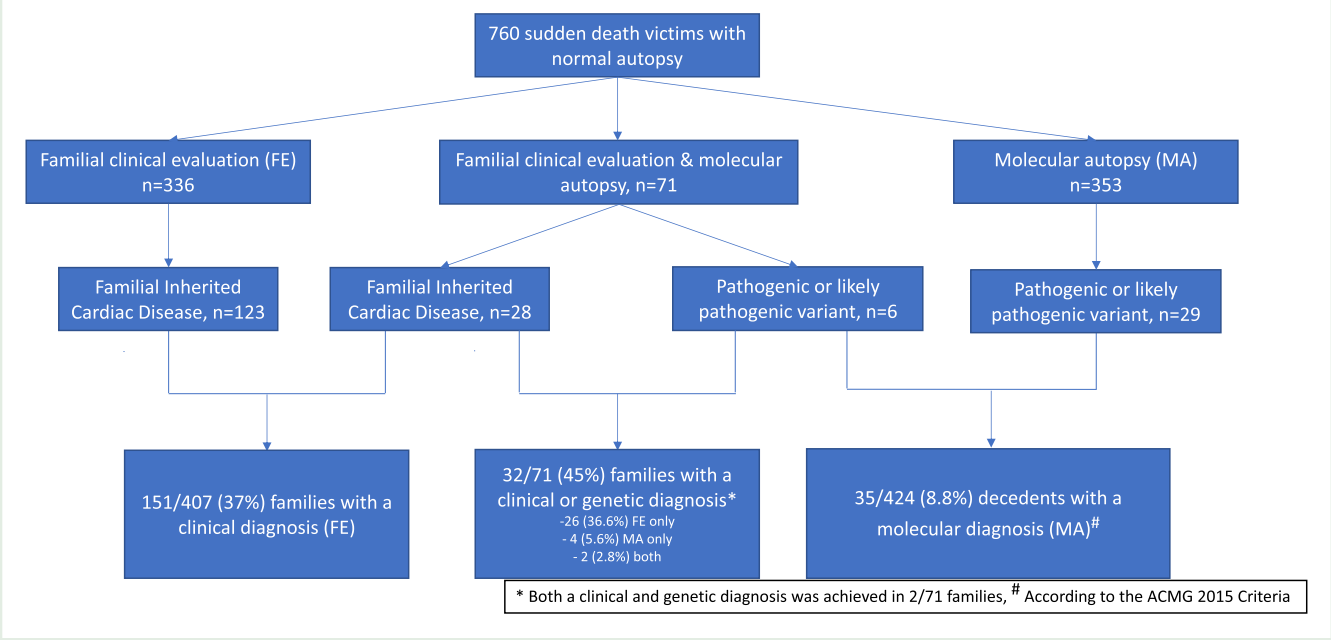


Figure 1 Flowchart of study cohort.

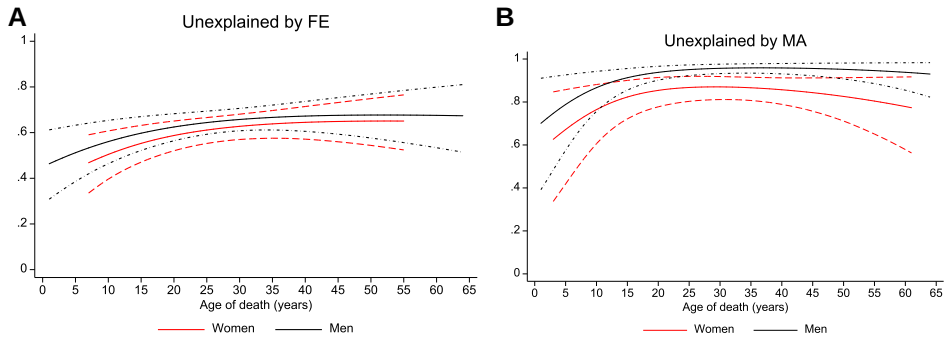


Figure 2 Likelihood of a sudden arrhythmic death syndrome death remaining unexplained is greater in males across all ages by familial evaluation (*A*) and by molecular autopsy (*B*).

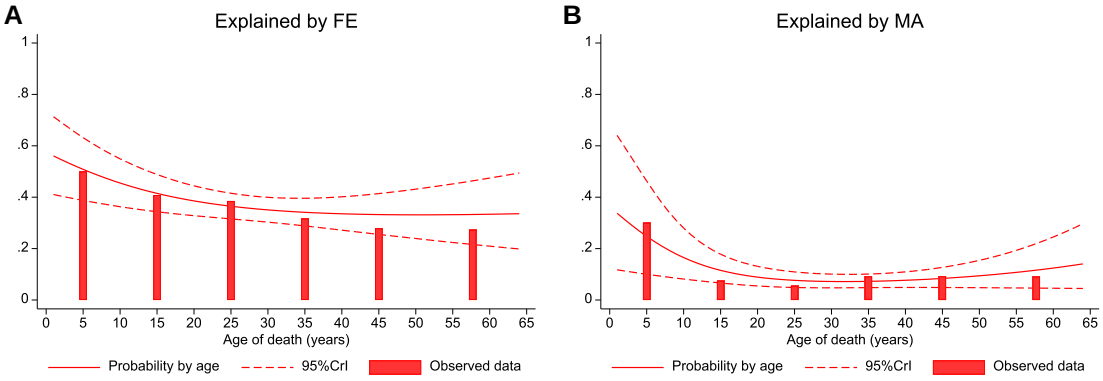


Figure 3 Overall diagnostic yield for sudden arrhythmic death syndrome deaths remaining unexplained after investigation, reduces with increased age at time of sudden death in both familial evaluation cohort (*A*) and molecular autopsy cohort (*B*).

Impact of age on the diagnostic yield of inherited cardiac conditions

The yield of an ICC diagnosis by FE was highest in the youngest decedents (*Figure 3A*), with an observed 50% yield in 0–10 years group. The yield of an ICC diagnosis by MA was also highest in the youngest decedents (*Figure 3B*), with an observed 30% yield in 0–10 years group. Both age-specific diagnostic yields had a decreasing trend with age with the yield of FE being 40% in 11–20 year olds and ~30% in >20 year olds and the MA yield being around 10% in those >10 year old. For each year increase in age, the relative risk ratio of an ICC diagnosis declined by 11% [RRR 0.89 (0.81–0.97)] for FE and by 12% [RRR 0.88 (0.81–0.94)] for MA.

There was an inverse age-related association for the overall diagnostic yield of LQTS (10% for ages 0–10, 7% for ages 11–20, and ~5% for >20 years). This was due to reduced probability of a LQTS diagnosis by FE, with the relative risk ratio declining by 5.6% [RRR 0.94 (0.91–0.98)] for each year increase in age. The relative risk of LQTS diagnosis with MA was similar across age groups (*Figure 4A and B*). There was also an inverse age-related association for the overall diagnostic yield of CPVT, which was consistent across both FE and MA. For each year increase in age, the relative risk of a CPVT diagnosis declined by 11% [RRR 0.89 (0.81–0.97)] for FE and by 12% [RRR 0.88 (0.81–0.94)] for MA (*Figure 4C and D*, see Supplementary material online, *Table S5*).

When adjusting for the combined effect of age and sex, results remained similar with a reduction of the diagnostic yield of LQTS (from FE) and CPVT (from both MA and FE) with advancing age.

Diagnosis of Brugada syndrome and the significance of the Shanghai score

The diagnosis of BrS was similar across all age groups of decedents. When the BrS diagnoses were stratified according to the relatives' highest Shanghai score, 16 were identified with definite/probable BrS (score ≥ 3.5), 45 with possible BrS (score 2–3) and 48 with a low likelihood of BrS. The probability of diagnosing BrS by Shanghai score was associated with advanced age, with a relative risk ratio increase of 7.8% (2.5–14%) for every year for possible vs. low likelihood, and a relative risk ratio increase of 11% (4–19%) for every year for definite vs. low likelihood (*Figure 5*).

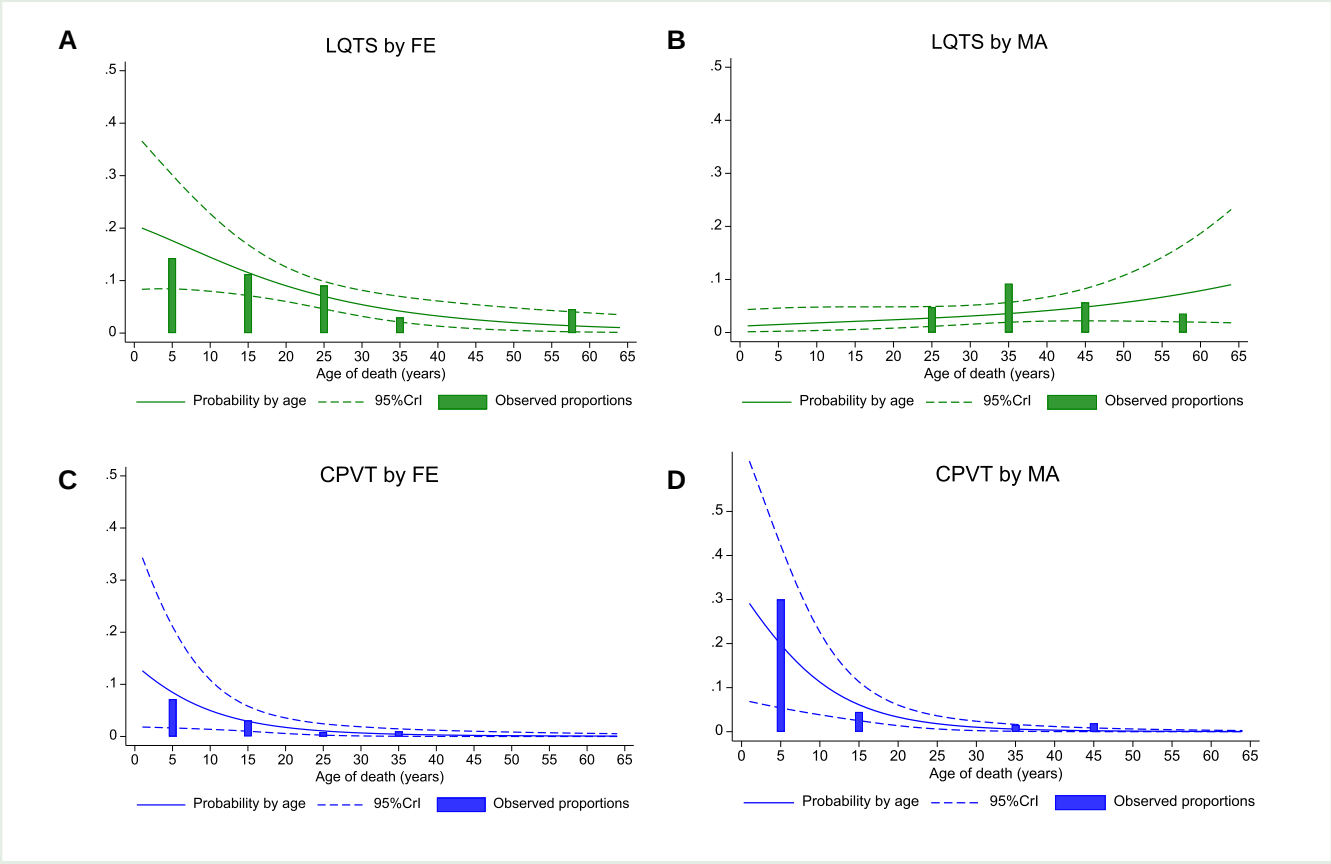


Figure 4 The age-dependent yield of the diagnosis by familial evaluation (*A* and *C*); and by molecular autopsy (*B* and *D*) for the diagnosis of long QT syndrome (*A* and *B*) and catecholaminergic polymorphic ventricular tachycardia (*C* and *D*) (data shown with 95% CI).

Discussion

This study explored the largest cohort of autopsy negative sudden unexplained deaths (SADS) to date, incorporating 760 decedents aged between 1 and 64 years and showed an overall diagnostic yield of an underlying ICC of up to 45% by a combined approach of FE and MA. The yield was 37% by FE alone and 9% by MA alone. Inherited arrhythmia syndromes formed the bulk of the diagnoses with both approaches. Brugada syndrome accounted for 70% of the ICC diagnoses by FE, with LQTS, CPVT, and BrS all identified fairly evenly in the MA cohort. Importantly, our study indicates that a targeted approach to the evaluation of SADS decedents is possible as both age and sex of the decedent had a significant influence on the diagnostic yield by both FE and MA. Younger decedents were more likely to receive a diagnosis of LQTS or CPVT and female sex was associated with a higher diagnostic yield of ICCs, particularly LQTS.

A key finding of this study is that our combined approach of FE + MA provides a diagnostic yield of up to 45% following sudden death in the young. This diagnostic yield is higher than has been suggested in previous studies highlighting the importance of the targeted combined approach. Hayashi *et al.*[18] have reviewed the spectrum of epidemiology of sudden death with their quoted yield up to 29%. They also highlight the age variation with increased yield of ICC in young sudden deaths, and lower yield in sudden death over 35, due to the rapid rise of coronary artery disease associated deaths in this age group. Our study showed a significant inverse association between the age of death and a diagnosis of CPVT or LQTS. This is congruent with patient cohorts where a higher risk of arrhythmic events were observed in younger probands of CPVT.[19,20] Indeed, the relative risk of CPVT diagnosis reduced by 11–12% for each year increase of age and the yield of CPVT diagnosis by either FE or MA was minimal for decedents of both sexes over the age of 25 years. Similarly, the relative risk of LQTS diagnosis by FE reduced by 5.6% for each year increase of age and a diagnosis of LQTS was rare for deaths occurring over the age of 40 years. Whilst CPVT is known to have a higher prevalence in children and adolescent sudden deaths, this is the first study to our knowledge, which shows increased age over 40 years leads to a lower likelihood of a death being due to LQTS.

Our data indicate that after a SADS death there is a greater likelihood of identifying an ICC in women rather than men. This was the case across both investigative approaches and all conditions. The sex discrepancy was more evident in the MA arm where female decedents were threefold more likely (15.3 vs. 5.5%) to exhibit a pathogenic or likely pathogenic variant. The difference was particularly striking for the presence of pathogenic or likely pathogenic variants related to LQTS with a sevenfold difference in females (8.1%) compared with males (1.2%). Our results mirror findings from a large cohort of over 1700 LQTS patients where female sex was an independent risk factor for life-threatening arrhythmias with a hazard ratio of 1.7 compared with male patients.[21] Our results do not support the findings of Priori *et al.*[22] who showed that

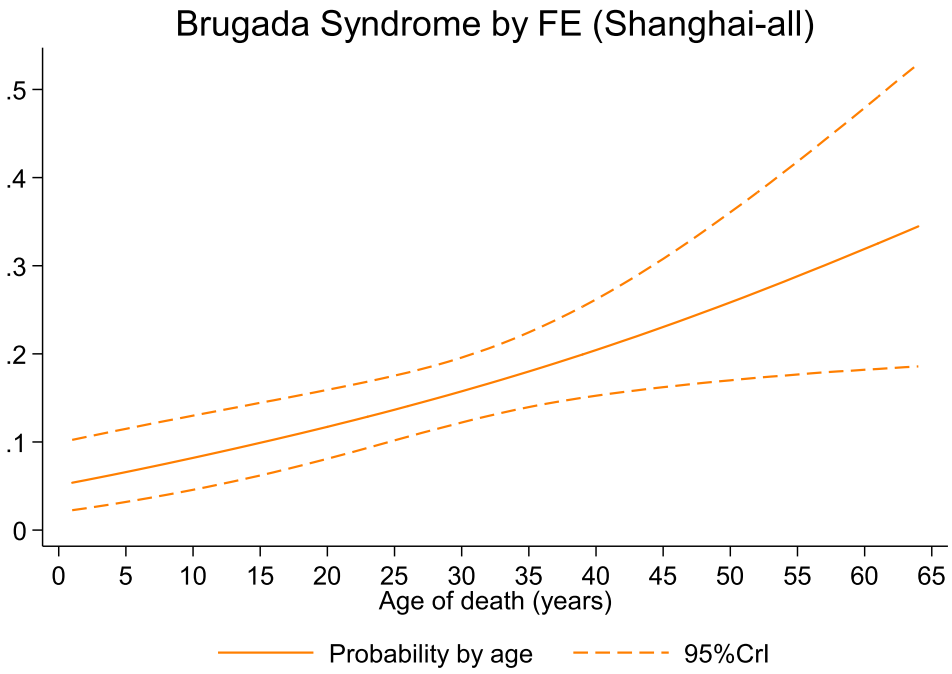


Figure 5 Diagnosis of definite/probable Brugada syndrome according to highest Shanghai score of decedents' relatives increases with increased age at death.

males with *RYR2* variants were more likely to experience events than females but the numbers in the CPVT group were small and results should be interpreted with caution. Our data suggests that MA is an important part of the diagnostic armamentarium following a SADS death in a female decedent given the higher diagnostic yield of up to 15.3% in our cohort.

These results also reflect the larger size and higher frequency of overall unexplained deaths in the male sub-group of SADS decedents. Aetiologies for these unexplained SADS cases where male sex predominates, remain to be defined, including any yield of genetically and clinically inscrutable forms of idiopathic ventricular fibrillation, early repolarization syndrome and short-coupled Torsades-de-Pointes and non-cardiac disorders such as sudden unexpected death in epilepsy (SUDEP).[23–25]

Employing the proposed Shanghai diagnostic scoring system in relatives of SADS decedents diagnosed with a type 1 Brugada pattern resulted in the downgrading of most diagnoses to possible or low likelihood. This is due to the impact of systematic Ajmaline provocation testing on the clinical yield of BrS, as reported in our earlier study.[10] There were, however, no differences in associated decedent characteristics across the three main diagnostic probability groups, other than a younger mean decedent age in the low likelihood sub-group. This may reflect that more severe cases presenting at a younger age are more likely to have greater polygenic or oligogenic susceptiblity,[26] and suggests that adjustments to the Shanghai scoring system will be necessary in order to diagnose and manage these families correctly. Furthermore, given the similar proportion of BrS diagnoses in younger and older age groups, greater recognition of the potential risk of paediatric BrS is required although the absolute numbers of paediatric deaths due to BrS is much lower than in adults.[27]

Our results indicate when the autopsy is unexplained; there is a role for a targeted approach in the evaluation of SADS decedents. Our study adds to the previous literature in unexplained sudden death by providing greater into the underlying cause based on the demographics of the decedent. This data also highlights the relative importance of incorporating a precision medicine approach following a young unexplained sudden death with age and gender targeted investigations a consideration. All but very few cases of CPVT and LQTS diagnoses were in the younger age group (<40 years), with BrS accounting for 14% of the diagnostic yield in decedents ≥40 years. This underscores the importance of high lead ECGs and provocation testing as the main diagnostic tool for BrS in the older age group and suggests that in centres where these tests are not performed there is likely to be limited diagnostic yield for FE in decedents ≥40 years.

The diagnostic yield of MA was 8.8% in our cohort. The value of MA was greatest in younger decedents and in females, largely due to the identification of pathogenic variants related to CPVT and LQTS. This is important when one considers the diagnostic challenges of both conditions and the potential for evidence-based interventions to reduce arrhythmic risk including initiation of beta-blockers. Moreover, especially in CPVT where a significant proportion may be due to *de novo* variants, where first degree family members are unlikely to be at risk, MA provides the opportunity for some degree of psychological closure for other family members.[13,28,29] Thus it is appropriate that MA is now recommended in SADS cases <40 years.[6,7] In countries where post-mortem genetic testing is not readily available, our study suggests the highest diagnostic yield to be in those decedents who are younger, and therefore this study can help guide appropriate resource allocation.

Sadly, in our cohort, 111 (18%) decedents had a family history of sudden death, prior to their own deaths. ICCs are inherited typically in an autosomal dominant manner with first-degree family members having a 50% risk of the same condition. Our results show that with the combined approach of MA and FE the diagnostic yield can be as high as

45%. We therefore hope that this high prevalence of a prior family history of sudden death decreases as families are referred on and investigated appropriately following a first sudden death in their family using our approach.[3,4,8,9]

Limitations

The study is limited by the small numbers of decedents ($n = 71$) who were comprehensively assessed with both MA and FE, the most informative subset. Ideally confirmatory genetic testing would have been performed in relatives diagnosed with a phenotype on FE to provide more evidence to the underlying molecular diagnosis in the deceased. In addition, the low diagnostic yield of MA and the low absolute numbers of decedents with a diagnosis of CPVT limit the overall power of the study despite the large cohort size. Nonetheless, CPVT is a rare condition with a prevalence of 1:10 000 and the prevalence is striking within our cohort of 760 decedents. The number of genes included in our panel was restricted by the availability of contemporaneous sequencing data and the Cardioclassifier software. The addition of more genes may have increased yield of rare variants, but given that many of these would be less definite in their role in SADS, we would not have expected a significant change in yield from that currently reported.

Conclusions

After a SADS death, the diagnostic yield of comprehensive FE and MA in an expert setting can be up to 45% using a combined approach. Younger age of death is associated with greater likelihood of LQTS and CPVT diagnosis with the highest yield in children and adolescents. Female sex in SADS decedents is associated with a higher yield of LQTS in MA. Targeted post-mortem testing can be considered as both age and sex have a significant influence on the diagnostic yield of FE and MA. Based on these data, there was no evidence to suggest an effect of age on the diagnosis of BrS by either FE or MA methods. The incremental value of MA in diagnosing CPVT and LQTS in younger decedents supports routine MA in SADS cases <40 years.

Supplementary material

Supplementary material is available at *European Journal of Preventive Cardiology*.

Acknowledgements

Thanks to Dr Irina Chis-Ster who conducted statistical aspects of this study and to Dr Stuart Cook at Imperial College for his contribution. The data underlying this article will be shared on reasonable request to the corresponding author.

Author contribution

B.G., E.R.B., S.P., and M.P. contributed to design of study, data acquisition, analysis, interpretation, drafting of primary and subsequent manuscripts. A.B., H.R., Y.W., G.F., A.M., M.T.E., M.N.S., and S.S. contributed to data acquisition and manuscript editing. N.W. and J.S.W. contributed to data analysis and manuscript editing. All authors have approved the final version of the manuscript.

Funding

B.G. was the recipient of a National Health and Medical Research Council (NHMRC) Early Career Fellowship (#1122330) and is the recipient of a Heart Foundation Future Leader Fellowship (#107244). E.R.B. receives research funding from the Robert Lancaster Memorial Fund sponsored by McColl's Retail Group Ltd. S.P., M.N.S., G.F., S.S., and M.P. receive funding from Cardiac Risk in the Young. H.R. was funded by British Heart Foundation Fellowship Grant FS/11/71/28918 which part supported the project. J.S.W. was supported by the Sir Jules Thorn Charitable Trust (21JTA), Medical Research Council (UK), British Heart Foundation (RE/18/4/34215), and the NIHR Imperial College Biomedical Research Centre. N.W. was supported by a Sir Henry Dale Fellowship jointly funded by the Wellcome Trust and the Royal Society (220134/Z/20/Z). For the purpose of open access, the authors have applied a Creative Commons Attribution (CC BY) licence to any Author Accepted manuscript version arising.

Conflict of interest: B.G. has participated in advisory board for Bristol Myers Squibb. J.S.W. has received research support from Bristol Myers Squibb, and has acted as a consultant for MyoKardia, Pfizer, Foresite labs, Health Lumen, and Tenaya Therapeutics. N.W. has received research funding from Novo Nordisk and has consulted for ArgoBio. M.T.E. has recieved consultancy fees from Bristol Myers Squibb and Cytokinetics. E.R.B. has acted as a consultant for Boston Scientific and Solid Biosciences. M.P. has consulted for Bristol Myers Squibb.

Data availability

The data underlying this article will be shared on reasonable request to the corresponding author.

References

1. Zeppenfeld K, Tfelt-Hansen J, de Riva M, Winkel B, Behr ER, Blom N, *et al.* 2022 ESC Guidelines for the management of patients with ventricular arrhythmias and the prevention of sudden cardiac death. *Eur Heart J* 2022;**43**:3997–4126.
2. Priori SG, Blomstrom-Lundqvist C, Mazzanti A, Blom N, Borggrefe M, Camm J, *et al.* 2015 ESC Guidelines for the management of patients with ventricular arrhythmias and the prevention of sudden cardiac death: the Task Force for the Management of Patients with Ventricular Arrhythmias and the Prevention of Sudden Cardiac Death of the European Society of Cardiology (ESC). Endorsed by: Association for European Paediatric and Congenital Cardiology (AEPC). *Eur Heart J* 2015;**36**:2793–2867.
3. Behr ER, Wood DA, Wright M, Syrris P, Sheppard MN, Casey A, *et al.* Cardiological assessment of first-degree relatives in sudden arrhythmic death syndrome. *Lancet* 2003;**362**:1457–1459.
4. Behr ER, Dalageorgou C, Christiansen M, Syrris P, Hughes S, Tome Esteban MT, *et al.* Sudden arrhythmic death syndrome: familial evaluation identifies inheritable heart disease in the majority of families. *Eur Heart J* 2008;**29**:1670–1680.
5. Lahrouchi N, Raju H, Lodder EM, Papatheodorou E, Ware JS, Papadakis M, *et al.* Utility of post-mortem genetic testing in cases of sudden arrhythmic death syndrome. *J Am Coll Cardiol* 2017;**69**:2134–2145.
6. Stiles MK, Wilde AAM, Abrams DJ, Ackerman MJ, Albert CM, Behr ER, *et al.* 2020 APHRS/HRS expert consensus statement on the investigation of decedents with sudden unexplained death and patients with sudden cardiac arrest, and of their families. *Heart Rhythm* 2021;**18**:e1–e50.
7. Wilde AAM, Semsarian C, Marquez MF, Sepehri Shamloo A, Ackerman MJ, Ashley EA, *et al.* European Heart Rhythm Association (EHRA)/Heart Rhythm Society (HRS)/Asia Pacific Heart Rhythm Society (APHRS)/Latin American Heart Rhythm Society (LAHRS) expert consensus statement on the state of genetic testing for cardiac diseases. *Heart Rhythm* 2022;**19**:e1–e60.
8. Behr ER, Scrocco C, Wilde AAM, Marijon E, Crotti L, Iliodromitis KE, *et al.* Investigation on sudden unexpected death in the young (SUDY) in Europe: results of the European Heart Rhythm Association Survey. *Europace* 2022;**24**:331–339.
9. Bagnall RD, Weintraub RG, Ingles J, Duflou J, Yeates L, Lam L, *et al.* A prospective study of sudden cardiac death among children and young adults. *N Engl J Med* 2016;**374**:2441–2452.
10. Papadakis M, Papatheodorou E, Mellor G, Raju H, Bastiaenen R, Wijeyeratne Y, *et al.* The diagnostic yield of Brugada syndrome after sudden death with normal autopsy. *J Am Coll Cardiol* 2018;**71**:1204–1214.
11. Gray B, Kirby A, Kabunga P, Freedman SB, Yeates L, Kanthan A, *et al.* Twelve-lead ambulatory electrocardiographic monitoring in Brugada syndrome: potential diagnostic and prognostic implications. *Heart Rhythm* 2017;**14**:866–874.
12. Gray B, Ackerman MJ, Semsarian C, Behr ER. Evaluation after sudden death in the young. *Circ Arrhythm Electrophysiol* 2019;**12**:e007453.

13. Ackerman MJ, Priori SG, Willems S, Berul C, Brugada R, Calkins H, *et al.* HRS/EHRA expert consensus statement on the state of genetic testing for the channelopathies and cardiomyopathies this document was developed as a partnership between the Heart Rhythm Society (HRS) and the European Heart Rhythm Association (EHRA). *Heart Rhythm* 2011;**8**:1308–1339.
14. Antzelevitch C, Yan G-X, Ackerman MJ, Borggrefe M, Corrado D, Guo J, *et al.* J-Wave syndromes expert consensus conference report: emerging concepts and gaps in knowledge. *Heart Rhythm* 2016;**13**:e295–e324.
15. Pua CJ, Bhalshankar J, Miao K, Walsh R, John S, Lim SQ, *et al.* Development of a comprehensive sequencing assay for inherited cardiac condition genes. *J Cardiovasc Transl Res* 2016;**9**:3–11.
16. Richards S, Aziz N, Bale S, Bick D, Das S, Gastier-Foster J, *et al.* Standards and guidelines for the interpretation of sequence variants: a joint consensus recommendation of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology. *Genet Med* 2015;**17**:405–424.
17. Whiffin N, Walsh R, Govind R, Edwards M, Ahmad M, Zhang X, *et al.* CardioClassifier: disease- and gene-specific computational decision support for clinical genome interpretation. *Genet Med* 2018;**20**:1246–1254.
18. Hayashi M, Shimizu W, Albert C. The spectrum of epidemiology underlying sudden cardiac death. *Circ Res* 2015;**116**:1887–1906.
19. Sy RW, Gollob MH, Klein GJ, Yee R, Skanes AC, Gula LJ, *et al.* Arrhythmia characterization and long-term outcomes in catecholaminergic polymorphic ventricular tachycardia. *Heart Rhythm* 2011;**8**:864–871.
20. Tester DJ, Medeiros-Domingo A, Will ML, Haglund CM, Ackerman MJ. Cardiac channel molecular autopsy: insights from 173 consecutive cases of autopsy-negative sudden unexplained death referred for postmortem genetic testing. *Mayo Clin Proc* 2012;**87**:524–539.
21. Mazzanti A, Maragna R, Vacanti G, Monteforte N, Bloise R, Marino M, *et al.* Interplay between genetic substrate, QTc duration, and arrhythmia risk in patients with long QT syndrome. *J Am Coll Cardiol* 2018;**71**:1663–1671.
22. Priori SG, Napolitano C, Memmi M, Colombi B, Drago F, Gasparini M, *et al.* Clinical and molecular characterization of patients with catecholaminergic polymorphic ventricular tachycardia. *Circulation* 2002;**106**:69–74.
23. Hesdorffer DC, Tomson T, Benn E, Sander JW, Nilsson L, Langan Y, *et al.* Combined analysis of risk factors for SUDEP. *Epilepsia* 2011;**52**:1150–1159.
24. Haissaguerre M, Derval N, Sacher F, Jesel L, Deisenhofer I, de Roy L, *et al.* Sudden cardiac arrest associated with early repolarization. *N Engl J Med* 2008;**358**:2016–2023.
25. Mellor G, Raju H, de Noronha SV, Papadakis M, Sharma S, Behr ER, *et al.* Clinical characteristics and circumstances of death in the sudden arrhythmic death syndrome. *Circ Arrhythm Electrophysiol* 2014;**7**:1078–1083.
26. Barc J, Tadros R, Glinge C, Chiang D, Jouni M, Simonet F, *et al.* Genome-wide association analyses identify new Brugada syndrome risk loci and highlight a new mechanism of sodium channel regulation in disease susceptibility. *Nat Genet* 2022;**54**:232–239.
27. Minier M, Probst V, Berthome P, Tixier R, Briand J, Geoffroy O, *et al.* Age at diagnosis of Brugada syndrome: influence on clinical characteristics and risk of arrhythmia. *Heart Rhythm* 2020;**17**:743–749.
28. Ingles J, Semsarian C. The value of cardiac genetic testing. *Trends Cardiovasc Med* 2014;**24**:217–224.
29. Gray B, Behr ER. New insights into the genetic basis of inherited arrhythmia syndromes. *Circ Cardiovasc Genet* 2016;**9**:569–577.