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ORIGINAL RESEARCH

Risk Prediction in Women With Congenital Long QT Syndrome

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BACKGROUND: We aimed to provide personalized risk estimates for cardiac events (CEs) and life-threatening events in women with either type 1 or type 2 long QT.

METHODS AND RESULTS: The prognostic model was derived from the Rochester Long QT Syndrome Registry, comprising 767 women with type 1 long QT (n=404) and type 2 long QT (n=363) from age 15 through 60 years. The risk prediction model included the following variables: genotype/mutation location, QTc-specific thresholds, history of syncope, and β-blocker therapy. A model was developed with the end point of CEs (syncope, aborted cardiac arrest, or long QT syndrome–related sudden cardiac death), and was applied with the end point of life-threatening events (aborted cardiac arrest, sudden cardiac death, or appropriate defibrillator shocks). External validation was performed with data from the Mayo Clinic Genetic Heart Rhythm Clinic (N=467; type 1 long QT [n=286] and type 2 long QT [n=181]). The cumulative follow-up duration among the 767 enrolled women was 22 243 patient-years, during which 323 patients (42%) experienced ≥1 CE. Based on genotype-phenotype data, we identified 3 risk groups with 10-year projected rates of CEs ranging from 15%, 29%, to 51%. The corresponding 10-year projected rates of life-threatening events were 2%, 5%, and 14%. C statistics for the prediction model for the 2 respective end points were 0.68 (95% CI 0.65–0.71) and 0.71 (95% CI 0.66–0.76). Corresponding C statistics for the model in the external validation Mayo Clinic cohort were 0.65 (95% CI 0.60–0.70) and 0.77 (95% CI 0.70–0.84).

CONCLUSIONS: This is the first risk prediction model that provides absolute risk estimates for CEs and life-threatening events in women with type 1 or type 2 long QT based on personalized genotype-phenotype data. The projected risk estimates can be used to guide female-specific management in long QT syndrome.

Key Words: genetics ■ long QT syndrome ■ QT interval ■ risk prediction ■ sudden cardiac death ■ syncope ■ women

Long QT syndrome (LQTS) is an arrhythmogenic genetic disorder characterized by prolonged ventricular repolarization and is commonly associated with cardiac events (CEs) such as syncope, cardiac arrest, and sudden cardiac death.[1,2] Two of the most common types of LQTS are type 1 long QT (LQT1) and type 2 long QT (LQT2), which together account for ≈70% of all cases of LQTS.[3] LQT1 is caused by mutations in the *KCNQ1* gene that impairs the Kv7.1 potassium channel, which gives rise to slow delayed rectifier potassium current (I_{Ks}). LQT2 is characterized by mutations in the α subunit of the *KCNH2*-encoded Kv11.1 channel that conducts the rapid delayed rectifier potassium current (I_{Kr}) in cardiac myocytes.[3,4]

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CLINICAL PERSPECTIVE

What Is New?

- This is the first risk prediction model that provides absolute risk estimates for cardiac and life-threatening events for women with congenital long QT syndrome based on both genetic and clinical data.
- Our prediction model identified 3 risk groups with distinct 10-year predicted risks of cardiac and life-threatening events in women after the onset of adolescence.

What Are the Clinical Implications?

- These projected risk estimates can help further refine and facilitate sex-specific risk stratification and clinical decision-making in congenital long QT syndrome.

Nonstandard Abbreviations and Acronyms

CE	cardiac event
C loop	cytoplasmic loop
LQT1	type 1 long QT
LQT2	type 2 long QT
LQTS	long QT syndrome
LTE	life-threatening event

Notably, mutation location is an important risk factor in patients with LQT1 and LQT2, with mutations located in the cytoplasmic loop (C loop) mutation domain of Kv7.1 and in the pore loop region of Kv11.1 exhibiting a significantly higher arrhythmic risk.[5,6]

Prior studies indicate that there are important sex differences in the clinical course of patients with LQT1 and LQT2, wherein women experience a pronounced increase in the risk of CEs after the onset of adolescence or during the postpartum and perimenopause periods, particularly for women with LQT2.[7–12] These findings may possibly be attributable to the modulating effects of estrogen and progesterone on the cardiac potassium channels affected by *KCNQ1* and *KCNH2* mutations.[13,14] Of note, similar modulating effects of sex hormones have not been shown for LQT3-causative mutations in the *SCN5A*-encoded Nav1.5 cardiac sodium channel.[2–4,12]

The unique clinical course experienced by women with LQT1 and LQT2 after the onset of adolescence suggests that female-specific risk prediction models are required to more optimally guide management in these inherited arrhythmic disorders, rather than current, age, and sex-adjusted approaches to LQTS population-based risk stratification.[7–16]

Accordingly, the present study was performed in a population of 767 genetically confirmed women from the Rochester Long-QT Syndrome Registry with either LQT1 or LQT2, and was designed to: (1) model CE risk in women with either LQT1 or LQT2 after the onset of adolescence based on genotype and mutation location; (2) incorporate female-specific QTc thresholds in an effort to develop personalized QTc cutoff values by mutation location risk; (3) assess the applicability of the risk prediction model for the end point of life-threatening events (LTEs); and (4) validate the model performance by using an external cohort from the Mayo Clinic Windland Smith Rice Genetic Heart Rhythm Clinic, comprising 467 women with LQTS including 286 with LQT1 and 181 with LQT2.

METHODS

Study Population

The data that support the findings of this study are available from the corresponding author upon reasonable request. The study population consisted of patients with either LQT1 or LQT2 enrolled in the Rochester Long-QT Syndrome Registry (n=1585). Patients were excluded from the study if there were missing mutation and location types (n=18), missing QTc duration (n=182), or multiple LQTS mutations (n=70). From the remaining 1315 patients, men were excluded. Thus, the final analysis population consisted of 767 genetically confirmed women with LQTS: 404 with LQT1 and 363 with LQT2, derived from 297 proband identified families. Patients were drawn primarily from the Rochester, NY, enrolling center (center 1) of the registry (n=727), as well as from data submitted by other investigators specifically for this collaborative mutation analysis project, including Israel (n=23) and Salt Lake City, UT (n=17). Patients from the Italian, Dutch, and Japanese enrolling centers of the Rochester Long-QT Syndrome Registry were not included in the study because of incomplete follow-up data on β-blocker treatment in women with LQT1 and LQT2 after the onset of adolescence. External validation was performed in an additional group of 467 women with LQTS derived from the Mayo Clinic Windland Smith Rice Genetic Heart Rhythm Clinic: 286 with LQT1 and 181 with LQT2. The Rochester and Mayo Clinic Long-QT Syndrome Registries are approved by institutional review boards of the University of Rochester and Mayo Clinic, respectively. All patients provided informed consent for participation.

Data Collection and Management

For each patient, information on personal history, including CEs, ECGs, and therapies, as well as

family history, were obtained at enrollment. Clinical data were then collected yearly on prospectively designed forms with information on demographic characteristics, personal and family medical history, ECG findings, medical therapies, left cardiac sympathetic denervation, implantation of a pacemaker or an implantable cardioverter-defibrillator, and the occurrence of LQTS-related CEs. The QT interval was corrected for heart rate using the Bazett formula to derive the patient's QTc value.[17] The routine approach for QTc assessment in both registries is based on a paper copy of baseline ECG findings. Lead II is reported using manual measurements to the nadir between the T wave and the isoelectric line or between the T wave and the U wave if present. Lead V5 is used as an alternative when QT measurement cannot be performed in lead II.

Beginning in 2010, information on menstruation, oral contraceptive use, pregnancy, and menopause were obtained from all women in the Long-QT Syndrome Registry using a specific questionnaire. To date, 299 women with LQT1 or LQT2, who were alive or enrolled after 2010, have completed this questionnaire. Data captured for the present analysis included follow-up through March 2019.

Genotype Characterization

The *KCNQ1* and *KCNH2* mutations were identified with the use of standard genetic tests conducted in academic molecular genetic laboratories including the Functional Genomics Center, University of Rochester Medical Center, Rochester, NY; Baylor College of Medicine, Houston, TX; and Boston Children's Hospital, Boston, MA.

Mutation definition and categorization is provided in Data S1, and the specific mutations included in the present study, by location, type, and number of patients, are detailed in Table S1. The distribution of the mutations in the *KCNQ1* and *KCNH2* genes by their frequency among study patients is shown in Figure S1A and S1B, respectively.

Based on our prior studies,[5,6,15,16] we prespecified 4 genotype and mutation location groups: higher risk mutation locations were defined as missense C loop for LQT1 and pore loop for LQT2; and lower risk mutation locations were defined as non–C loop for LQT1 and non–pore loop for LQT2 for each respective genotype.

End Points

The primary end point was CE, defined as first occurrence of syncope, aborted cardiac arrest requiring defibrillation as part of resuscitation, or LQTS-related sudden cardiac death (abrupt in onset without evident cause, if witnessed, or death that was not explained by any other cause if it occurred in a nonwitnessed setting such as sleep). The secondary end point was LTE, defined as first occurrence of aborted cardiac arrest, LQTS-related sudden cardiac death, or appropriate implantable cardioverter-defibrillator shock.

The age for initiation of follow-up was established specifically at 15 years. This age cutoff was selected based on the fact that in women with LQT1 and LQT2, the rates of CEs and LTEs are essentially low before the age of 15 years, with a pronounced increase after the age of 15 years (Figure S2A and S2B, respectively). Furthermore, these data are consistent with the fact that the mean (±SD) age of menses onset was 13 (±2) years in the analysis sample.

Statistical Analysis

Patient baseline characteristics, prior treatments and cardiac episodes, and treatments and cardiac episodes during the follow-up period were summarized as mean±SD for continuous variables and frequency (percentage) for categorical variables among 4 prespecified genotype and mutation location groups. Continuous variables were compared by genotype and mutation location using Wilcoxon rank-sum tests (2-sample test) or Kruskal-Wallis tests (>2-sample test), and categorical variables were compared using chi-square tests.

To explore the association between CEs and genotype and mutation location groups, Kaplan-Meier curves were then generated to estimate the probability of CEs by 4 genotype and mutation location groups, and the log-rank test was used to assess whether the differences between the outcome curves were statistically significant at $P<0.05$. Furthermore, to evaluate the independent association of clinical and genetic factors with first occurrence of a CE, multivariable Cox proportional hazards regression modeling was performed in which robust standard errors were used to account for clustering of patients within a family.[18] Covariates used in the model included the following: the 4 genotype and mutation location groups, QTc value, a history of syncope before the age of 15 years, and time-dependent β-blocker therapy. The effect of β-blocker therapy within each mutation location category was assessed by performing an interaction test.

To determine QTc-specific thresholds within each of the 4 genotype and mutation location groups, Kaplan-Meier curves by quartiles of QTc durations among each genotype and mutation location groups were generated. The log-rank tests were used to identify patients with similar observed probability of CEs (eg, Kaplan-Meier curves overlapped across follow-up time) in which were grouped together. Next, these groups were entered in the Cox models

for CE outcome adjusting for time-dependent β-blocker therapy and history of syncope before the age of 15 years and Wald test statistics were used to further combine the groups with similar estimated β coefficients. Thus, 3 risk groups based on QTc-specific thresholds, genotype, and mutation location were formed. In addition, a risk prediction model for CEs was developed using a Cox model with robust standard errors adjusting for 3 risk groups, a history of syncope before the age of 15 years, and time-dependent β-blocker therapy. Last, this risk prediction model, developed for the CE outcome and including the 3 risk groups, a history of syncope before the age of 15 years, and time-dependent β-blocker therapy covariates, was estimated for the LTE outcome. Adjusted probability curves of a CE and LTE based on the average of covariate vectors among the 3 risk groups was displayed across age in year. Furthermore, the 10-year observed and predicted risk of a CE was calculated for these 3 risk groups. For the adjusted curves and 10-year predicted risk, baseline β-blocker therapy at age 15 years was used instead of time-dependent β-blocker therapy.[19,20]

Internal validation of the predictive model involved creating 100 bootstrap samples by sampling records with replacement from the studied population. The discriminative performance of the model was quantified by Harrell concordance C statistics.[21] For the external validation, variable definitions and data rules were provided to the Mayo Clinic Windland Smith Rice Genetic Heart Rhythm Clinic, comprising 467 women with LQTS: 286 with LQT1 and 181 with LQT2. From this independent cohort, the C statistics were calculated using the same set of covariates (eg, 3 risk groups, a history of syncope before age 15 years, and time-dependent β-blocker therapy) by re-estimating the parameters. All statistical analyses were performed using SAS version 9.4 software (SAS Institute Inc).

RESULTS

Clinical Characteristics of the Study Population

The clinical characteristics of study patients by genotype and mutation location are shown in Table 1. Overall, both women with C loop–associated, LQT1-causative mutations and women with pore loop localizing, LQT2-causative mutations exhibited a higher rate of baseline risk factors, including a more prolonged baseline QTc duration, increased likelihood of prior β-blocker therapy, and a higher rate of syncope before age 15 years (Table 1).

Female-Specific Information Relating to Sex hormones

The mean±SD age of menses onset was 13±2 years (median, 12; interquartile range, 11–14). A total of 142 women (48%) used oral contraceptives at any time during follow-up, with a mean age of 33±14 years at first use. Pregnancy at any time during follow-up occurred in 262 women (88%) at a mean age of 26±5 years at first pregnancy; and 190 women reported menopause (64%) at a mean age of 48±5 years (Table 2). Female-specific information relating to sex hormones was similar by genotype/mutation location (Table 2).

Mutation-Specific Risk in Women With LQTS

During the total accumulated follow-up time of 22 243 patient-years, 323 of 767 women (42%) experienced a CE. The cumulative probability of CEs from the age of 15 to 60 years was initially assessed based on genotype and mutation location (Figure 1A). This analysis showed that LQT2 women with pore loop mutations exhibited the highest unadjusted rate of CEs. The rate of CEs was intermediate in women with LQT2 who had non–pore mutations and in women who had LQT1 with C loop mutations, and was lowest in women with LQT1 who had non–C loop mutations ($P<0.001$ for the overall difference during follow-up). Kaplan-Meier survival analysis for the end point of LTE showed consistent findings (Figure 1B).

Multivariate analysis showed that, compared with women with LQT1 who had non–C loop mutation, women with LQT2 who had pore loop mutations had a 2.2-fold ($P<0.001$) increase in the risk of CE; women with LQT2 who had non–pore loop mutations had a corresponding 1.9-fold ($P<0.001$) increased risk; and women with LQT1 with C loop mutations did not show a significant risk increase compared with women with LQT1 who had non–C loop mutations after multivariate adjustment (Table S2). Similarly, analysis within the LQT1 genotype did not show a risk difference between women with C loop and non–C loop mutations (data not shown).

Similar results were shown for the end point of LTEs (Table S3). Compared with women with LQT1 who had non–C loop mutations, women with LQT2 who had pore loop mutations had an adjusted 3.3-fold ($P<0.001$) increase in the risk of LTEs; women with LQT2 who had non–pore loop mutations had a corresponding 2.2-fold ($P=0.002$) increased risk; and women with LQT1 who had C loop mutations did not show a significant increase in the risk of LTEs compared with women with LQT1 who had non–C loop mutations.

Table 1. Clinical Characteristic of the Study Population by Genotype and Mutation Location

Clinical Characteristics	LQT1		LQT2		*P* Value		
	Non–C Loop (n=332)	C Loop (n=72)	Non–Pore Loop (n=283)	Pore Loop (n=80)	LQT1	LQT2	All 4 Groups
ECG parameters							
QTc, ms	483±43	501±53	481±52	506±61	0.007	0.001	<0.001
QTc >500 ms	74 (22)	32 (44)	67 (24)	30 (38)	<0.001	0.014	<0.001
RR, ms	859±189	831±230	862±214	844±175	0.30	0.62	0.70
Prior treatment before 15 y							
β-Blockers	54 (16)	21 (29)	43 (15)	26 (33)	0.011	<0.001	<0.001
Pacemaker	3 (1)	1 (1)	3 (1)	6 (8)	0.55	0.005	0.005
ICD	6 (2)	3 (4)	6 (2)	2 (3)	0.20	0.69	0.55
LCSD	0 (0)	0 (0)	0 (0)	1 (1)		0.22	0.20
Prior cardiac events							
Syncope	70 (21)	30 (42)	41 (14)	26 (33)	<0.001	<0.001	<0.001
ACA	3 (1)	0 (0)	2 (1)	3 (4)	1.00	0.07	0.15
Appropriate ICD shocks	1 (0)	1 (1)	0 (0)	0 (0)	0.33		0.27
Therapies during follow-up after 15 y							
β-Blockers	214 (64)	49 (68)	208 (73)	72 (90)	0.56	0.002	<0.001
Pacemaker	9 (3)	1 (1)	31 (11)	10 (13)	1.00	0.70	<0.001
ICD	56 (17)	8 (11)	85 (30)	37 (46)	0.23	0.007	<0.001
LCSD	4 (1)	0 (0)	7 (2)	3 (4)	1.00	0.46	0.21
CEs during follow-up							
Syncope	98 (30)	29 (40)	132 (47)	45 (56)	0.08	0.13	<0.001
ACA	11 (3)	7 (10)	29 (10)	7 (9)	0.026	0.69	0.005
SCD	3 (1)	2 (3)	5 (2)	4 (5)	0.22	0.11	0.07
Appropriate ICD shocks	12 (4)	2 (3)	14 (5)	10 (13)	1.00	0.016	0.021

Data are presented either as mean±SD or number (percentage). ACA indicates aborted cardiac arrest; C loop, cytoplasmic loop; CE, cardiac event; ICD, implantable cardioverter-defibrillator; LCSD, left cardiac sympathetic denervation; LQT1, long QT type 1; LQT2, long QT type 2; and SCD, sudden cardiac death.

Time-dependent β-blocker therapy was associated with a significant 46% reduction in the risk of CE and with a corresponding 44% reduction in LTE risk in women with LQTS. The effect of β-blocker therapy on the risk of CE was similar within each mutation location category (β-blocker by mutation location interaction=0.74).

Table 2. Female-Specific Information Related to Sex Hormones

Clinical Characteristic	LQT1		LQT2		*P* Value		
	Non–C Loop (n=135)	C-Loop (n=16)	Non–Pore Loop (n=121)	Pore-Loop (n=27)	LQT1	LQT2	All 4 Groups
Menarche							
Age at first occurrence of menstruation, y	13±2	14±2	13±3	12±2	0.39	0.52	0.79
Oral contraceptives							
Oral contraceptive use	69 (51)	9 (56)	54 (45)	10 (37)	0.70	0.47	0.43
Age at first oral contraceptive use, y	33±14	30±12	33±15	30±14	0.71	0.76	0.96
Pregnancy							
History of pregnancy	120 (89)	15 (94)	103 (85)	24 (89)	1.0	0.77	0.78
Age at first pregnancy, y	26±6	24±5	26±5	26±6	0.16	0.90	0.51
Menopause							
Menopause	89 (66)	12 (75)	74 (61)	15 (56)	0.47	0.59	0.52
Age at menopause, y	48±5	46±10	49±5	48±5	0.69	0.31	0.63

Data are presented as mean±SD or number (percentage). C loop indicates cytoplasmic loop; LQT1, long QT type 1; and LQT2, long QT type 2.

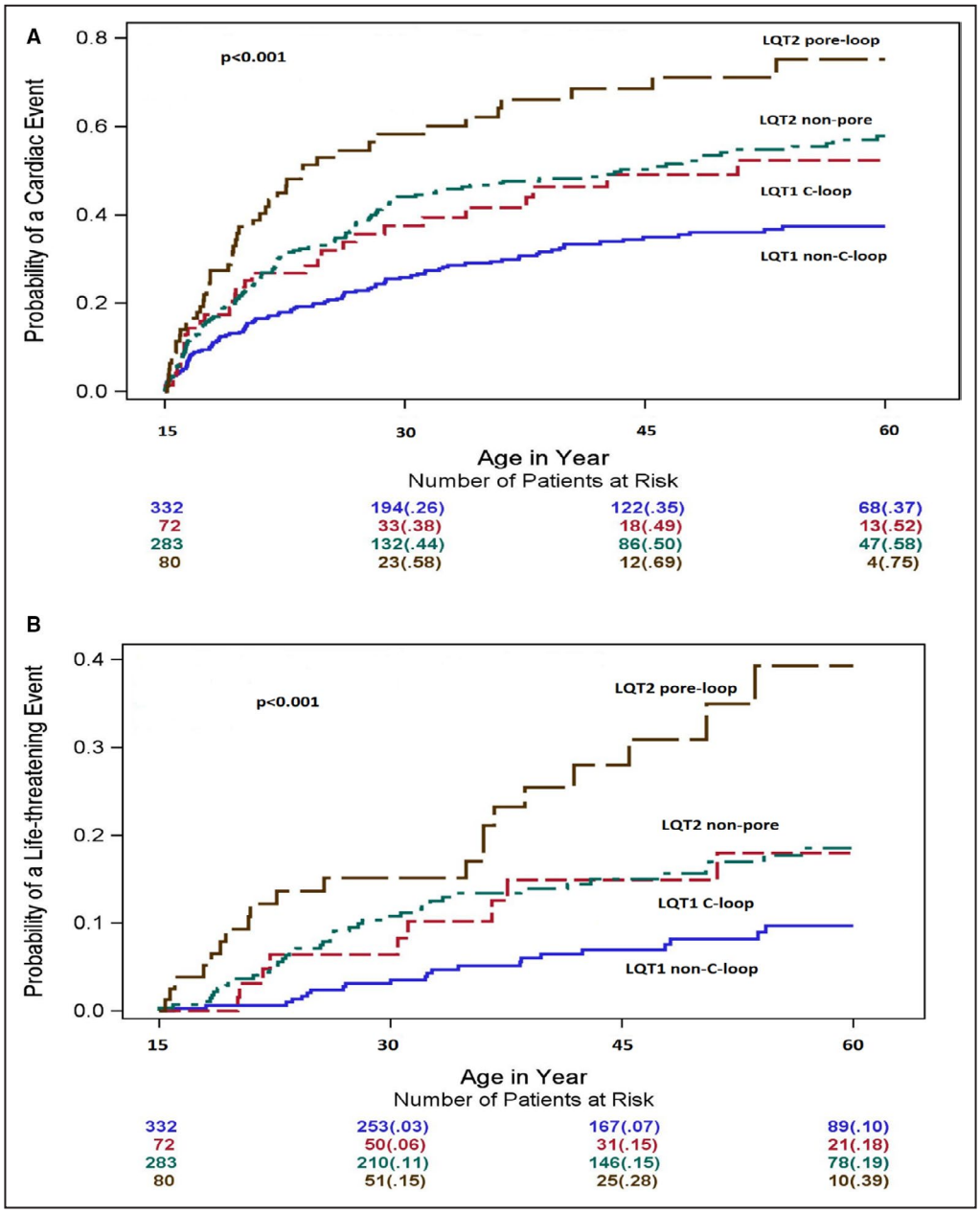

Figure 1. Kaplan-Meier estimates of outcomes from age 15 through 60 years by genotype and mutation location end point: cardiac events (A) end point: life-threatening events (B).
C loop indicates cytoplasmic loop; LQT1, long QT type 1; and LQT2, long QT type 2.

Development of the Genotype-Phenotype Risk Prediction Model

QTc-specific thresholds were explored for each genotype and mutation location groups for CE, which revealed 3 risk groups that are summarized in Figure 2: (1) low risk for CE (including women with LQT1 who had C loop mutations and QTc ≤450 ms; women with LQT1 who had non–C loop mutations and QTc <480 ms; and women with LQT2 who had non–pore loop mutations and QTc ≤450 ms); (2) intermediate risk for CE (including women with LQT1 who had C loop mutations and QTc >450 ms; women with LQT1 who had non–C loop mutations and QTc ≥480 ms; women with LQT2 who had non–pore loop mutations and QTc 451 to 500 ms; and women with LQT2 who had pore loop mutations and QTc ≤460 ms); and (3) high risk for CE (including women with LQT2 who had non–pore loop mutations and QTc >500 ms; and women with LQT2 who had pore loop mutations and QTc >460 ms). The prediction model showed that, compared with the low-risk group, high-risk women had a 4-fold increased risk of CE and intermediate-risk women had a 2-fold increased risk of CE (Figure 2). The C statistic of the prediction

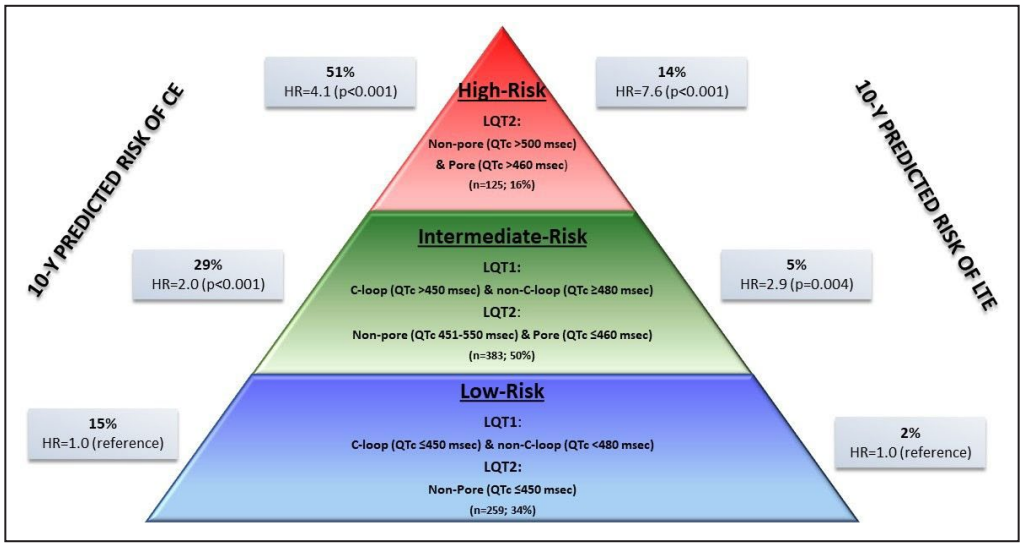

Figure 2. Risk groups for cardiac events (CEs) and life-threatening events (LTEs) in women with long QT syndrome.*

Risk groups were identified based on combined assessment of genotype-phenotype data in women with either long QT type 1 (LQT1) or long QT type 2 (LQT2). *Findings are further adjusted for time-dependent β-blocker therapy and a history of syncope before age 15 years. Risk estimates may vary based on medical treatment and/or clinical history, which should be taken into account in decision-making. C loop indicates cytoplasmic loop; and HR, hazard ratio.

model was 0.682 (95% CI 0.653, 0.711), where the C statistic of 100 bootstrap samples was 0.680 (95% CI 0.651–0.709).

Performance of the Prediction Model

Covariate-adjusted risk prediction of CE from age 15 to 60 years for the 3 risk groups are presented in Figure 3A.

To evaluate the performance of the risk prediction model, we compared the 10-year predicted risk of CE with the corresponding observed Kaplan-Meier rates. The projected 10-year CE risks were 15% in the low-risk group, 29% in the intermediate-risk group, and 51% in the high-risk group, which were similar to the observed rates (Figure S3A).

Applicability of the Prediction Model for the End Point of Life-Threatening Events

When applied to the end point of LTEs, high-risk women had a 7.6-fold increased risk for LTEs (hazard ratio [HR], 7.6; 95% CI, 3.6–16.3) and intermediate-risk women had a 2.9-fold increased risk for LTEs (HR, 2.9; 95% CI, 1.4–6.1) when compared with low-risk women (Figure 2). The C statistic was 0.713 (95% CI 0.662–0.764), where the C statistic of 100 bootstrap samples was 0.725 (95% CI 0.682–0.769).

Covariate-adjusted risk prediction of LTEs from age 15 to 60 years for the 3 risk groups are presented in Figure 3B.

The 10-year projected rates of LTEs after age 15 years were 2% in low-risk women, 5% in intermediate-risk women, and 14% in high-risk women. Projected rates were similar to observed rates in each risk group (Figure S3B).

External Independent Validation of the Risk Prediction Model

External validation was conducted in 467 women with LQTS from the Mayo Clinic Windland Smith Rice Genetic Heart Rhythm Clinic that included 286 with LQT1 and 181 with LQT2. Models using the same set of covariates as those with the Rochester, NY–based data were estimated with the Mayo Clinic LQTS patient data. The clinical characteristics of Mayo Clinic's validation cohort are provided in Table S4.

Based on the re-estimating parameters using the Mayo Clinic LQTS data, external validation confirmed model performance with C statistics of 0.649 (95% CI 0.597–0.700) and 0.766 (95% CI 0.695–0.838) for the 2 CEs and LTEs end points, respectively. The prediction model in the external validation Mayo Clinic cohort showed that, compared with the low-risk group, women classified as high risk had a 4.6-fold increased risk of CEs (adjusted HR, 4.61; $P<0.001$) and intermediate-risk women had a 2.5-fold increased risk of CE (adjusted HR, 2.46; $P<0.001$). For the end point of LTEs, the respective adjusted HRs were 12.4 ($P<0.001$) and 4.00 ($P=0.002$).

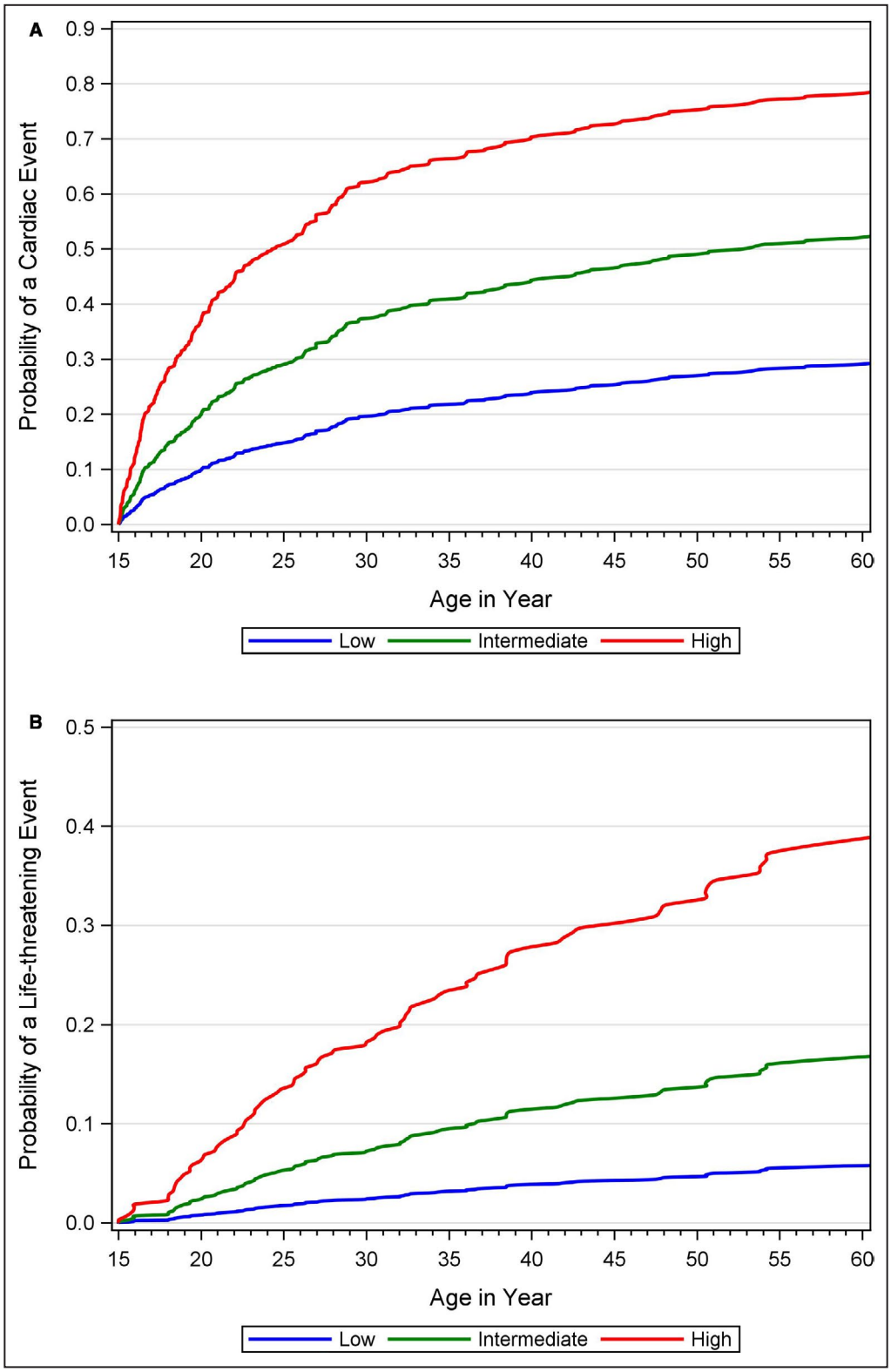

Figure 3. Adjusted risk prediction of outcomes from age 15 to 60 years for cardiac events (A) and (B) life-threatening events (B).

DISCUSSION

Women with either LQT1 or LQT2 experience hormonal changes that may predispose to QT prolongation and vulnerability to arrhythmias after the onset of adolescence.[13,14] These effects were suggested to be caused by a modulating effect of female sex hormones on mutations in the potassium channels (Kv7.1 and Kv11.1) affected in LQT1 and LQT2, respectively.[14] We therefore aimed to develop a prediction model focused on women with LQTS during the high-risk adolescence and postadolescence periods by utilizing a novel approach of integrating mutation-related risk with QTc-specific thresholds. Our prediction model identified 3 risk groups with distinct 10-year predicted risks of CE and LTE that can be used to guide female-specific decision-making in LQTS, specifically LQT1 and LQT2. Importantly, model validation in the external cohort derived from Mayo Clinic's Windland Smith Rice Genetic Heart Rhythm Clinic confirmed model performance with C statistics of 0.65 and 0.77 for the respective end points of CEs and LTEs.

Female-Specific Risk in LQTS

Prior studies have shown significant age-related differences in the risk for CEs between men and women with congenital LQTS, wherein women experience an increased risk for CEs after the onset of adolescence,[7–9] during the postpartum period,[10,22] and during the perimenopause period.[11] In contrast, the risk of LTE among men is attenuated after the onset of adolescence.[7–9] The mechanisms underlying the influence of sex hormones on cardiac repolarization are complex and are not completely understood.

Sex hormones have varying effects on I_{Ks} (Kv7.1) and I_{Kr} (Kv11.1) currents.[14] Testosterone increases the potassium channel currents, resulting in a shorter QTc in both animal and human studies. Progesterone increases the I_{Ks} current and may therefore shorten the QTc, whereas estrogen decreases the potassium channel currents and may lengthen the QTc through this mechanism.[14] Thus, the genotype and mutation-specific risk identified for women after the onset of adolescence is not applicable for men, who exhibit a reduction in the risk of CEs and LTEs after the onset of adolescence and increased risk associated with LQT1 C loop mutations.[15,16] These data suggest a need for sex-specific risk modeling in patients with LQTS rather than general models that merely incorporate sex as a covariate.

The present study establishes a comprehensive approach to risk stratification for women with either LQT1 or LQT2 who are affected by changes in sex hormones. A recent study by Mazzanti et al[23] similarly developed a prediction model for LTEs that incorporated the interplay between QTc duration and the genotype. We extend this individualized approach to women with LQTS by combining mutation-related risk with QTc-specific thresholds (Figure 1).

Management Implications

Current guidelines for the management of patients with congenital LQTS provide recommendations for β-blocker therapy or treatment intensification based mainly on the presence of clinical symptoms and QTc duration, categorized as high risk at >500 ms and as lower risk at <470 ms.[24,25] However, these general recommendations do not reflect individualized risk of women with either LQT1 or LQT2 after the onset of adolescence. Our personalized risk estimates can be used for shared decision-making on possible management strategies to reduce the risk of CEs and LTEs in women with LQTS after the onset of adolescence. For example, based on our prediction score, the 10-year risk of LTE in women with LQT2 with a pore variant and a QTc of 470 to 490 ms is 15%. Thus, women with LQT2 who have a pore loop mutation should be considered to be at a high risk for a life-threatening event after the onset of adolescence even with a QTc <500 ms, which is the traditional cutoff for risk assessment in LQTS.

Our prediction model shows that the 10-year predicted risk of LTEs increases from 2% and 5% in low- and intermediate-risk women, respectively, to 14% in high-risk women even after adjustment for β-blocker treatment. These risk estimates suggest that careful follow-up and more advanced therapies, such as videoscopic left cardiac sympathetic denervation or prophylactic implantable cardioverter-defibrillator implantation should be considered in women with high-risk LQT1 and LQT2 who do not tolerate β-blocker therapy or experience ongoing LQTS-associated CEs despite β-blocker therapy. In contrast, among asymptomatic low-risk women, shared decision-making can be used to agree on the need for preventive medical management.

Limitations and Strengths

Several limitations of this study should be noted. First, we did not conduct in vitro expression studies to assess the effects of estrogen and testosterone on ion channel mutations by their location in the potassium channels. Further studies are necessary to evaluate the underlying mechanisms related to the observed female-specific risk related to mutation location. Second, the presented risk prediction model was developed for the end point of CEs that is dominated by nonfatal syncopal events. However, when assessing the applicability of the CE risk prediction modeling for the more severe end point of LTEs, our findings remained

consistent with somewhat improved C statistics, further supporting the applicability of the prediction model for LTEs. Third, we have previously shown that women with LQTS, particularly LQT2, also experience a pronounced increase in the risk of CEs during the postpartum and perimenopause periods.[10,11] However, these higher risk time periods were not specifically evaluated and incorporated in the present prediction model. Of note, the rate of occurrence of these higher risk periods was similar among the genotype/mutation location subgroups (Table 2). Conversely, the strengths of this study are its inclusion of a large cohort of patients, systematic acquisition of phenotypic and genotypic data, and validation of its predictive findings utilizing an independent Registry data set.

It should also be noted that in the present study we used a minor allele frequency criterion of <0.0002 in large healthy population databases, combined with QTc prolongation observed in the proband patient as the definition of a pathogenic variant (with the exception of 1 patient with a minor allele frequency of 0.006 for the R148W variant [man with QTc=480 ms] known to be associated with current reduction).[26] Using minor allele frequency as a criterion for pathogenicity may not take into account a rare variant "background noise" in *KCNQ1* and *KCNH2*.

We also performed sensitivity analysis by excluding the R148W variant, which yielded virtually identical results (Table S5).

CONCLUSIONS AND CLINICAL IMPLICATIONS

This is the first risk prediction model that provides absolute risk estimates for CEs and LTEs for women with congenital LQTS based on both genetic and clinical data. These projected risk estimates can help further refine and facilitate clinical decision-making in this population. Future studies are needed to further investigate mechanisms relating to modulating effects of sex hormones on the phenotypic expression of LQTS and to evaluate potential hormonal-based interventions in this population.

ARTICLE INFORMATION

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Disclosures

MJA is a consultant for Abbott, ARMGO Pharma, Boston Scientific, Daiichi Sankyo, Invitae, LQT Therapeutics, Medtronic, and UpToDate. MJA and Mayo Clinic have an equity/royalty relationship with AliveCor and Anumana. Howveer, none of these entities participated in this study in any way. No other disclosures relevant to this study are reported from the other coauthors.

Supplementary Material

Data S1
Tables S1–S5
Figures S1–S3

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SUPPLEMENTAL MATERIAL

Data S1.

Mutation definitions and categorization

In the present study we used a minor allele frequency (MAF) criterion of <0.0002 in large healthy population databases, combined with QTc prolongation observed in the proband patient as the definition of a pathogenic variant (with the exception of one patient with a MAF of 0.0006 for the R148W variant (male with QTc = 480 msec) known to be associated with current reduction.[27] Pathogenicity of either mutations or rare variants was, when possible, confirmed using phenotype segregation in families, ion channel functional studies and computer based algorithms that predict functional effect of the variant.

Based on prior data regarding mutation-location/function and arrhythmic risk in LQT1,[5] mutations were categorized by their location and type in the *KCNQ1*-encoded Kv7.1 channel subunit as follows: (1) missense mutations in the C-loops; defined as the coding sequence involving amino acid residues from 171 to 195 (S2-S3 linker) and from 242 to 262 (S4-S5 linker); and (2) other LQT1 mutations as the reference group (i.e. missense mutations not in the C-loops, splice sites, in-frame insertions, in-frame deletions, nonsense [stop codon], and frameshift).[5,14]

KCNH2 mutations were characterized by location in the channel protein and by the type of mutation (missense, splice site, in-frame insertions/deletions, nonsense, and frameshift). Based on prior data in LQT2,[6,15] the transmembrane region of the *KCHNH2*-encoded Kv11.1 protein was defined as the coding sequence involving amino acids residues from 404 through 659 (pore region: 548-659), with the N-terminus region defined before residue 404, and the C-terminus region after residue 659. “Non-pore-loop” mutations are defined as C- or N- terminus missense, and transmembrane beta-barrel (TMB).

Table S1. Specific mutations included in the present study, by location, type, and number of patients.

	Number of Patients
Individual Location	**(n=767)**
C-Loop	87
C-term	246
N-term	124
Pore	80
TMB	230
Individual Mutation Type	**(n=767)**
NCR	1
In-frame deletion	3
Frameshift	67
In-frame insertion	4
Missense	587
Splice site	32
Stop codon	73

C-loop = cytoplasmic-loop; C-term = C-terminus; NCR = Non coding region; N-term = N-terminus; TMB = transmembrane beta-barrel.

Table S2. **Adjusted risk of a first cardiac event by genotype and mutation-location.**

Variable	HR	(95% CI)	P-value
LQT2 pore-loop	2.21	1.47 – 3.33	<0.001
LQT2 non-pore-loop	1.90	1.47 – 2.45	<0.001
LQT1 C-loop	1.15	0.74 – 1.78	0.53
LQT1 non-C-loop	1.00	(Reference Group)	
QTc duration (per 10-msec increase)	1.08	1.05 – 1.10	<0.001
History of syncope prior to age 15 years	2.64	2.01 – 3.45	<0.001
Beta-blocker use*	0.54	0.38 – 0.78	0.001

*Assessed as a time-dependent covariate

CI = confidence interval; HR = hazard ratio; All other abbreviations are defined in Table 1.

Table S3. Adjusted risk of a first life-threatening event by genotype and mutation-location.

Variable	HR	(95% CI)	P-value
LQT2 pore-loop	3.29	1.72 – 6.31	<0.001
LQT2 non-pore-loop	2.20	1.35 – 3.59	0.002
LQT1 C-loop	1.33	0.64 – 2.79	0.45
LQT1 non-C-loop	1 – (Reference Group)		
QTc duration (per 10-msec increase)	1.08	1.04 – 1.11	<0.001
History of syncope prior to age 15 years	2.06	1.26 – 3.37	0.004
Beta-blocker use*	0.66	0.39 – 0.93	0.03

*Assessed as a time-dependent covariate

CI = confidence interval; HR = hazard ratio; All other abbreviations are defined in Table 1.

Table S4. Clinical characteristics of the study population by genotype and mutation-location from the Mayo Clinic Windland Smith Rice Genetic Heart Rhythm Clinic validation cohort.

	LQT1		LQT2		P-value		
Clinical Characteristics[+]	Non-C-loop (n=258)	C-loop (n=28)	Non-pore-loop (n=135)	Pore-loop (n=46)	LQT1	LQT2	4 groups
ECG Parameters							
QTc, msec	471±29	474±38	467±34	498±73	0.80	<0.001	0.006
QTc > 500 msec	31 (12)	6 (21)	17 (13)	13 (28)	0.23	0.014	0.025
Prior Treatment before 15 yrs							
Beta-blockers	49 (19)	9 (32)	33 (24)	18 (39)	0.10	0.056	0.015
Pacemaker	0 (0)	0 (0)	1 (1)	2 (4)		0.16	0.021
ICD	4 (2)	0 (0)	4 (3)	5 (11)	1.00	0.048	0.017
LCSD	3 (1)	1 (4)	4 (3)	3 (7)	0.34	0.37	0.088
Prior Cardiac Events							
Syncope	22 (9)	5 (18)	11 (8)	7 (15)	0.11	0.25	0.19
ACA	0 (0)	0 (0)	1 (1)	1 (2)		0.44	0.12
App ICD Shocks	2 (1)	0 (0)	1 (1)	3 (7)	1.00	0.051	0.040
Therapies During Follow-up after 15 yrs							
Beta-blockers	206 (80)	24 (86)	114 (84)	42 (91)	0.62	0.33	0.25
Pacemaker	2 (1)	0 (0)	3 (2)	0 (0)	1.00	0.57	0.55
ICD	27 (11)	3 (11)	42 (31)	26 (57)	0.97	0.002	<0.001
LCSD	46 (18)	2 (7)	12 (9)	10 (22)	0.19	0.021	0.032
Cardiac Events During Follow-up							
Syncope	40 (16)	4 (14)	23 (17)	14 (30)	0.87	0.052	0.12
ACA/SCD	4 (2)	0 (0)	10 (7)	4 (9)	1.00	0.76	0.005
App ICD Shocks	9 (3)	0 (0)	16 (12)	13 (28)	0.61	0.018	<0.001

[+]Data are presented either as mean±SD or No (%).

All other abbreviations are defined in Table 1.

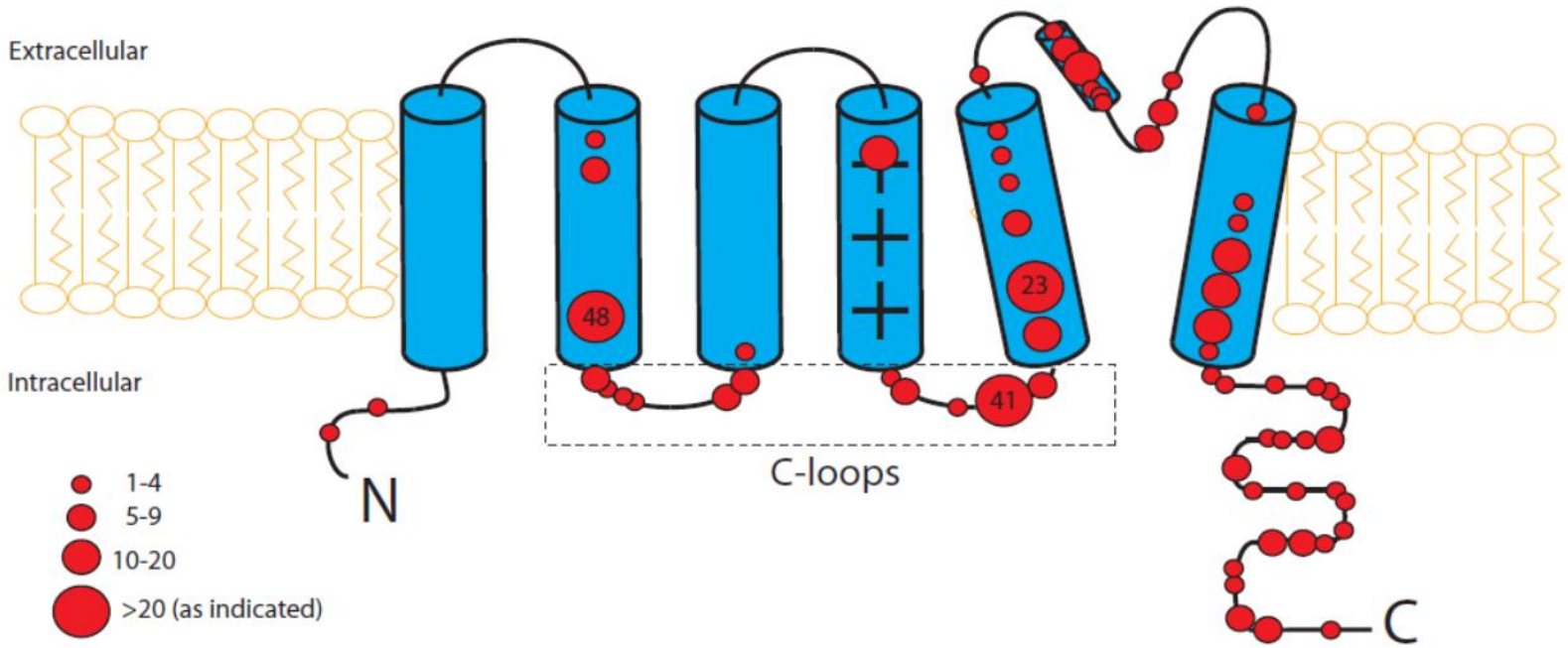
Table S5. Sensivity analysis of main model of cardiac event by excluding patients who were classified as pathogenic based on MAF >0.0002 *

Variable	Adjusted[+] HR	95% CI	p-value
High-Risk	4.11	2.88-5.86	<0.001
Intermediate-Risk	2.04	1.52-2.75	<0.001
Low-Risk	1.00 (Reference)		

*One patient with the R148W variant and a MAF of 0.0006 was excluded

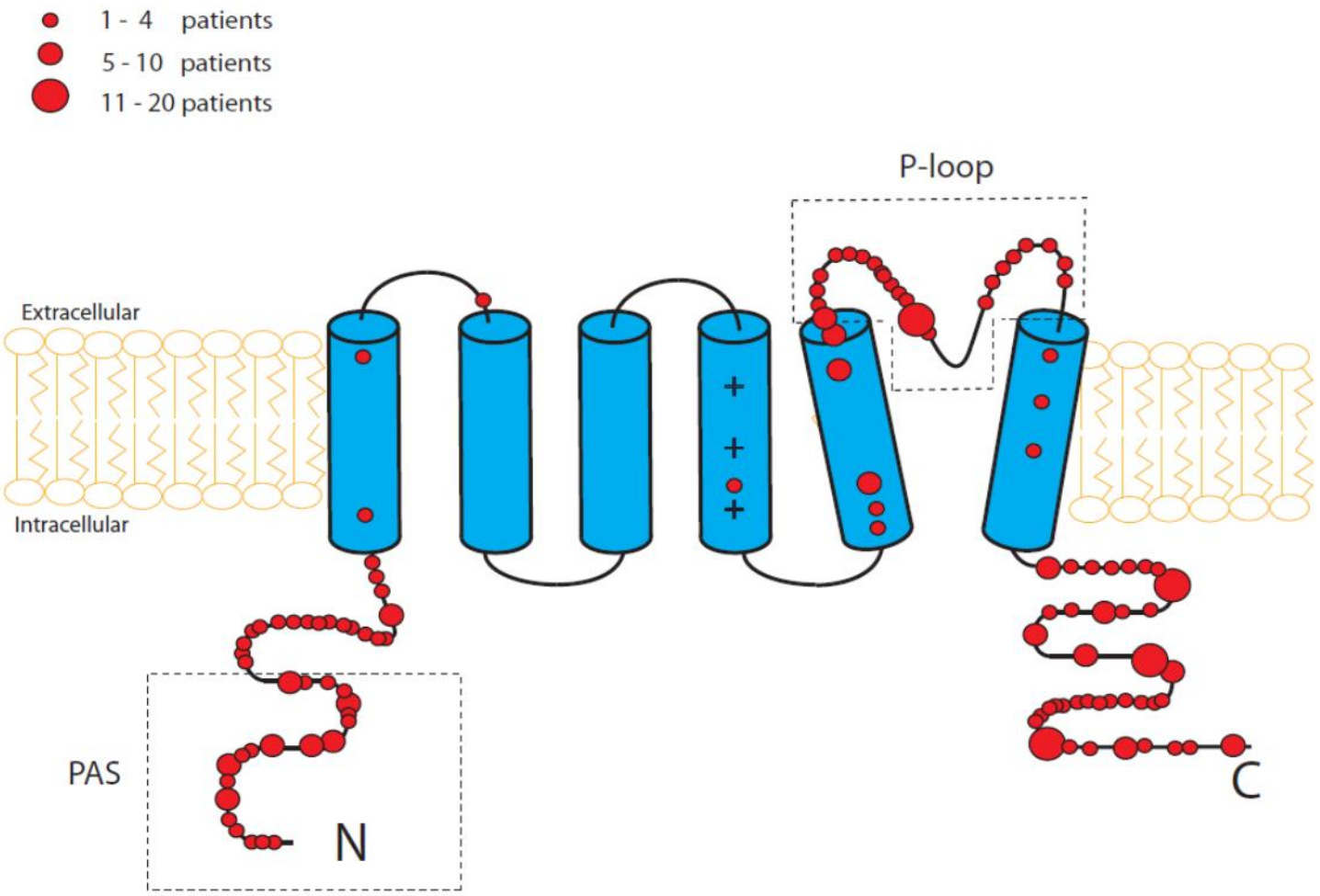
Figure S1. Distribution of mutations among study patients.

A. LQT1 women with mutations in the *KCNQ1*-encoded Kv7.1 potassium channel



C-loop = cytoplasmic-loops; LQT1 = long QT syndrome type 1

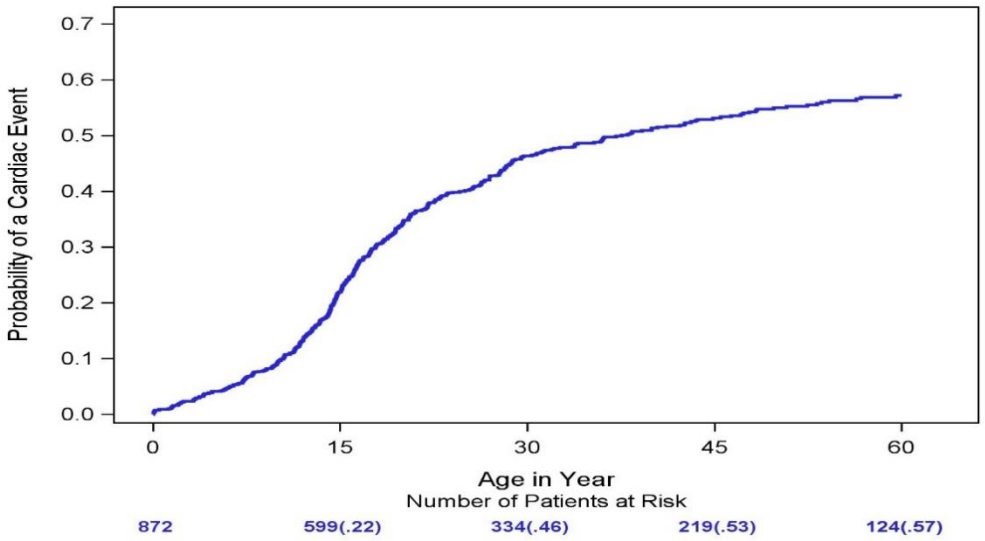
B. LQT2 women with mutations on the *KCNH2*-encoded Kv11.1 potassium channel



LQT2 = long QT syndrome type 2; PAS = Per-Arnt-Sim

Figure S2.

A. Cumulative probability of a first cardiac event from birth through age 60 years in women with either LQT1 or LQT2



B. Cumulative probability of a life-threatening event from birth through age 60 years in women with either LQT1 or LQT2

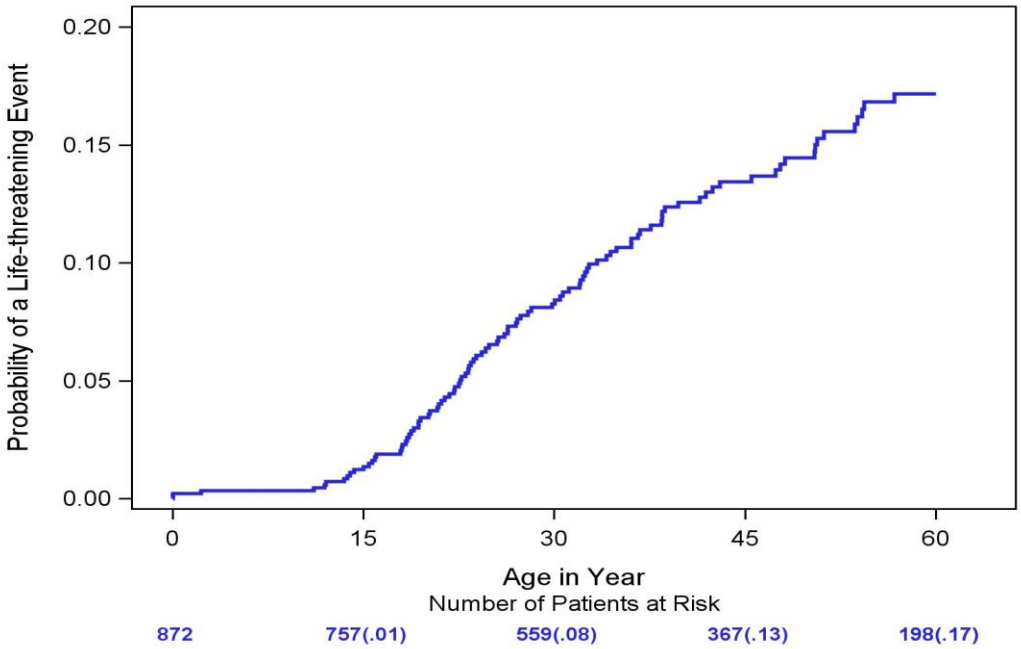
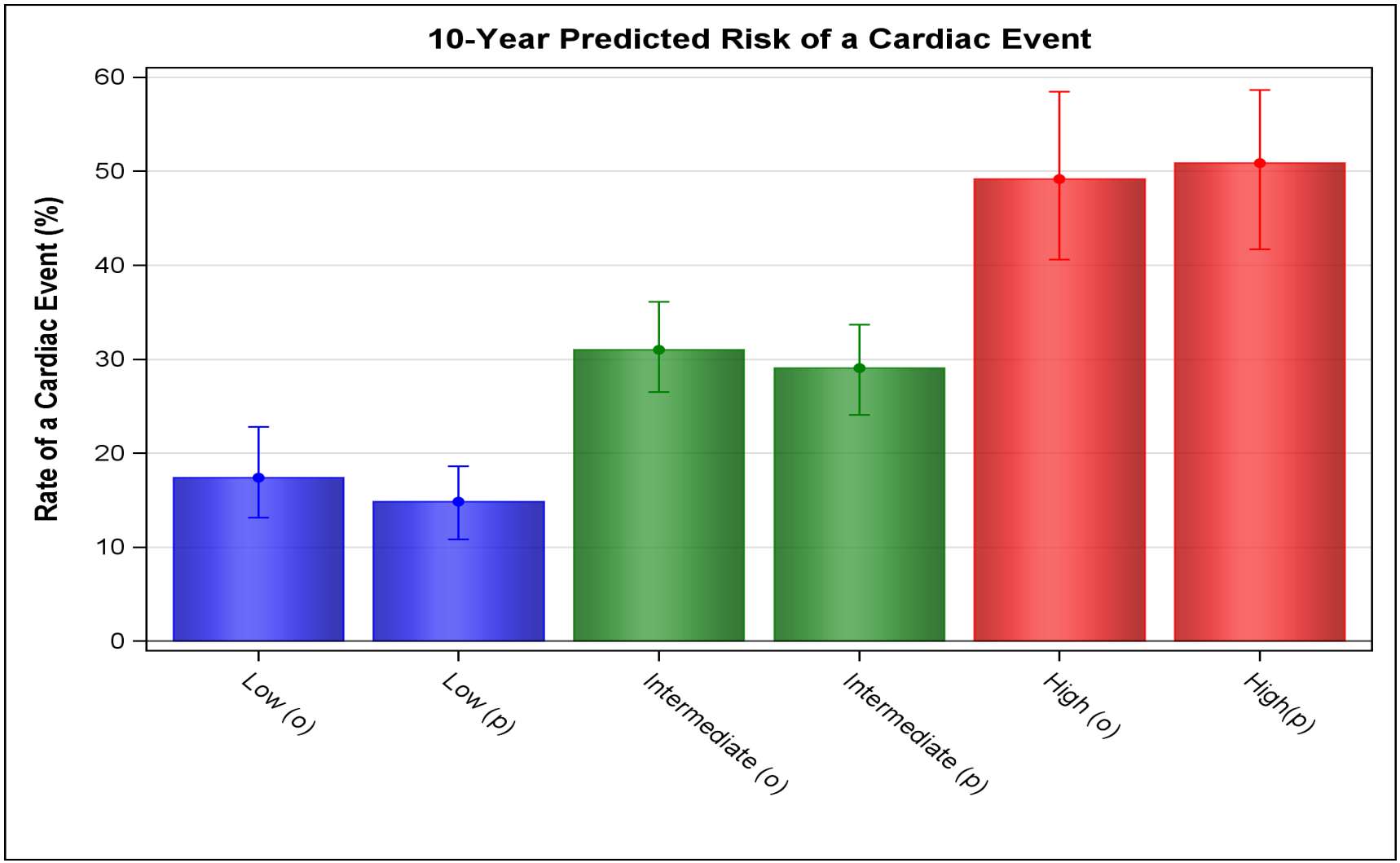
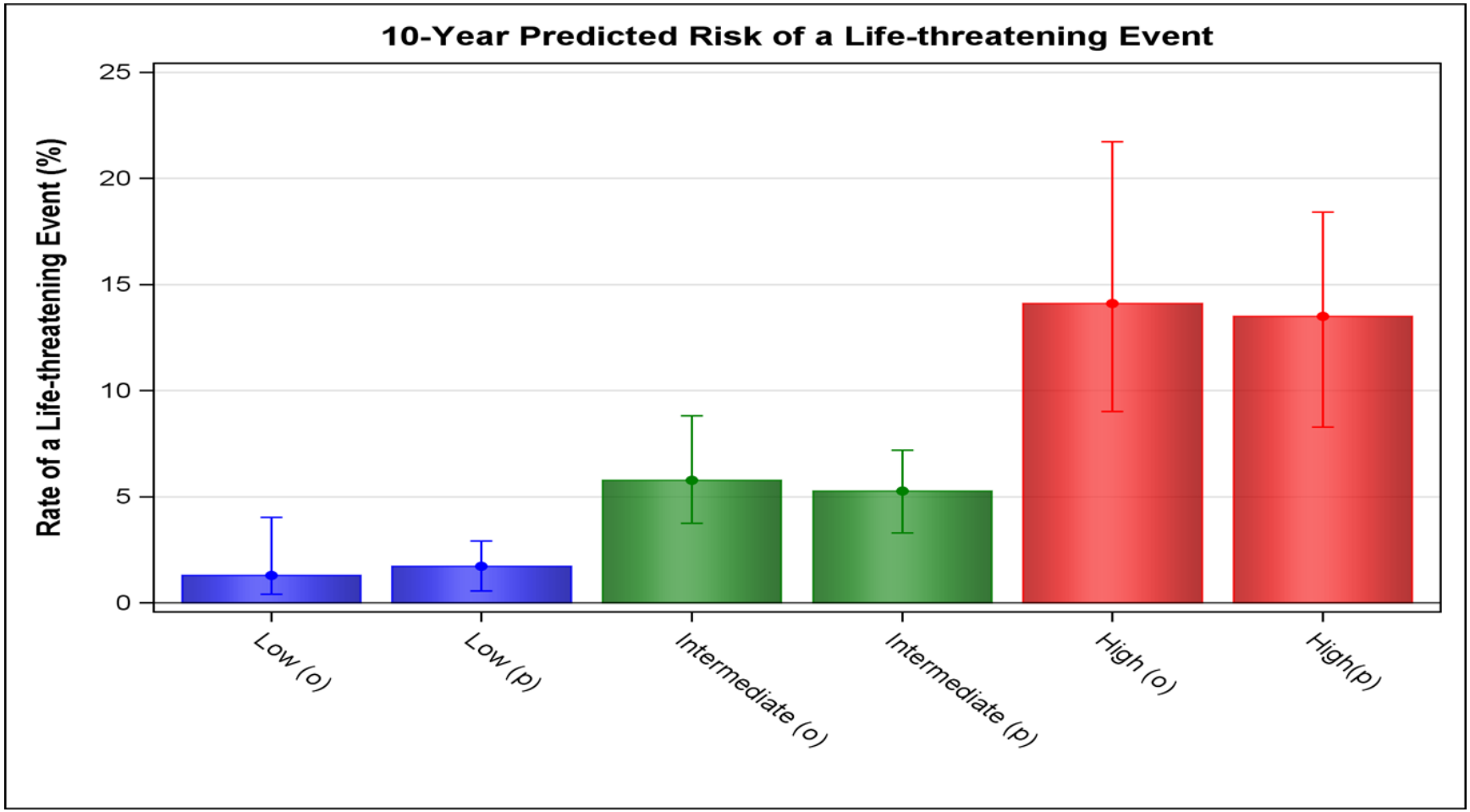


Figure S3. 10-year predicted (p) risk of outcomes with the corresponding observed (o) Kaplan-Meier rates*

A. Endpoint: Cardiac events (CEs)



B. Endpoint: Life-threatening events (LTEs)



O = observed; P = predicted.

*whisker represents 95% confidence intervals.