Common Genetic Variant Risk Score Is Associated With Drug-Induced QT Prolongation and Torsade de Pointes Risk

A Pilot Study

Editorial, see p 1321

BACKGROUND: Drug-induced QT interval prolongation, a risk factor for life-threatening ventricular arrhythmias, is a potential side effect of many marketed and withdrawn medications. The contribution of common genetic variants previously associated with baseline QT interval to drug-induced QT prolongation and arrhythmias is not known.

METHODS: We tested the hypothesis that a weighted combination of common genetic variants contributing to QT interval at baseline, identified through genome-wide association studies, can predict individual response to multiple QT-prolonging drugs. Genetic analysis of 22 subjects was performed in a secondary analysis of a randomized, double-blind, placebo-controlled, crossover trial of 3 QT-prolonging drugs with 15 time-matched QT and plasma drug concentration measurements. Subjects received single doses of dofetilide, quinidine, ranolazine, and placebo. The outcome was the correlation between a genetic QT score comprising 61 common genetic variants and the slope of an individual subject's drug-induced increase in heart rate–corrected QT (QTc) versus drug concentration.

RESULTS: The genetic QT score was correlated with drug-induced QTc prolongation. Among white subjects, genetic QT score explained 30% of the variability in response to dofetilide (r=0.55; 95% confidence interval, 0.09–0.81; P=0.02), 23% in response to quinidine (r=0.48; 95% confidence interval, −0.03 to 0.79; P=0.06), and 27% in response to ranolazine (r=0.52; 95% confidence interval, 0.05–0.80; P=0.03). Furthermore, the genetic QT score was a significant predictor of drug-induced torsade de pointes in an independent sample of 216 cases compared with 771 controls (r^2=12%, $P=1\times10^{-7}$).

CONCLUSIONS: We demonstrate that a genetic QT score comprising 61 common genetic variants explains a significant proportion of the variability in drug-induced QT prolongation and is a significant predictor of drug-induced torsade de pointes. These findings highlight an opportunity for recent genetic discoveries to improve individualized risk-benefit assessment for pharmacological therapies. Replication of these findings in larger samples is needed to more precisely estimate variance explained and to establish the individual variants that drive these effects.

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Clinical Perspective

What Is New?

- We demonstrated that a genetic risk score comprising multiple independent genetic variants that have previously been found to be associated with QT interval duration is collectively associated with the degree of drug-induced QT prolongation.
- In addition, the genetic risk score was associated with drug-induced torsade de pointes in a case-control cohort.

What Are the Clinical Implications?

- If our results are confirmed in real-world collections of drug-exposed patients with larger sample sizes, the genetic risk score (updated as new variants are discovered) could potentially be used to individualize assessment of risks and benefits of drugs with high risk for drug-induced arrhythmias.

The US government recently launched a Precision Medicine Initiative to move away from a "one size fits all approach" for medical therapies and instead take into account specific characteristics of individual patients.[1] Outside of oncology, advances in pharmacogenomics have been limited, with the exception of the genetic basis of drug absorption, distribution, metabolism, and excretion (pharmacokinetics), which are traits often controlled by 1 or a few genetic mechanisms rather than the many mechanisms responsible for most complex traits and diseases. Drug-induced QT prolongation (reflecting delayed ventricular repolarization), which is a risk factor for torsade de pointes, is a potential side effect of many marketed and withdrawn medications through their direct actions on the heart (pharmacodynamics).[2]

We previously performed genome-wide association studies (GWASs) of the electrocardiographic QT interval identifying many common genetic variants that contribute a modest increment to resting QT interval (eg, ≈1 to 3 milliseconds per allele) when considered individually.[3–5] We demonstrated that a genetic QT score is a strong predictor of baseline QT interval, with individuals in the top quintile having a 15-millisecond-higher QT interval compared with individuals in the bottom quintile,[6] explaining up to 10% of QT variation (≈25% of its heritability).[4] In the present study, we test the hypothesis that a weighted combination of common genetic variants contributing to QT at baseline will predict individual response to multiple QT-prolonging drugs and risk of torsade de pointes in a case-control study.

METHODS

Clinical Study Design

The study was approved by the US Food and Drug Administration Research Involving Human Subjects Committee and local institutional review boards. All subjects gave written informed consent. The study design and primary results (not including genetic analysis) have been previously published.[7,8] The study was a randomized, double-blind, crossover study of healthy subjects (Figure 1) at a phase 1 clinical research unit (Spaulding Clinical, West Bend, WI) to differentiate the effects of individual versus multichannel block on the ECG. The inclusion and exclusion criteria were similar to those for thorough QT studies. Subjects were 18 to 35 years old, 50 to 85 kg, and without a family history of cardiovascular disease or unexplained sudden cardiac death. Subjects also had to have a baseline heart rate–corrected QT (QTc) of <450 milliseconds for men (470 milliseconds for women) with the Fridericia correction and <12 ventricular ectopic beats during a 3-hour continuous recording at screening.

There was a 7-day washout period between each 24-hour treatment period. In the morning of each period, subjects received a single dose of 500 μg dofetilide (Tikosyn, Pfizer, New York, NY), 400 mg quinidine sulfate (Watson Pharma, Corona, CA), 1500 mg ranolazine (Ranexa, Gilead, Foster City, CA), 120 mg verapamil hydrochloride (Heritage Pharmaceuticals, Edison, NJ), or placebo. As previously reported,[7] verapamil did not prolong QTc at the dose administered and is not included in this analysis of the association of genetic variants with QTc prolongation.

Continuous ECGs were recorded at 500 Hz with an amplitude resolution of 2.5 μV. From the continuous recording, triplicate 10-second ECGs were extracted before dose and at 15 predefined time points over 24 hours after dose, during which the subjects were resting in a supine position for 10 minutes. ECGs were extracted with stable heart rates and maximum signal quality with Antares software (AMPS-LLC, New York, NY) at each of the 16 time points.[9] All postdose time points were time matched with blood samples for pharmacokinetic analysis. Plasma drug concentration was measured with a validated liquid chromatography with tandem mass spectroscopy method by Frontage Laboratories (Exton, Philadelphia, PA).[7]

Semiautomatic adjudication of the ECG intervals of the upsampled ECGs was carried out by investigators blinded to treatment and time as previously described.[7] For identification of the peak of the T wave (T_{peak}) and end of the T wave (T_{end}), 2 ECG readers identified the global peak and end of the T wave in the vector magnitude lead derived from the Guldenring transformation matrix.[10] T_{peak} was located by fitting a parabola through the T-wave peak. In the presence of a notch, the T_{peak} was defined as the first discernible peak. T_{end} was determined with the tangent method, which involves locating the intersection between the line through the terminal descending part of the T wave and isoelectric line. This approach of using the global vector magnitude lead to identify T_{peak} and the tangent method for T_{end} is not the same as T_{peak}-T_{end} measured in a precordial lead but produces more consistent measurements. In cases of low-amplitude, flat T waves, this results in longer QT intervals. Disagreements on a T wave being measureable, the presence of a notch, or a difference of >5 milliseconds in either T_{peak} or T_{end} were rereviewed and adjudicated by an expert ECG reader. This was the case for only ≈1.4% of ECGs.[7] QT was corrected for heart rate with the Fridericia formula (QTc), and J-T_{peak} was corrected with the Johannesen formula (J-T_{peak}c=J-T_{peak}/$RR^{0.58}$, with RR in seconds), whereas T_{peak}-T_{end} was not

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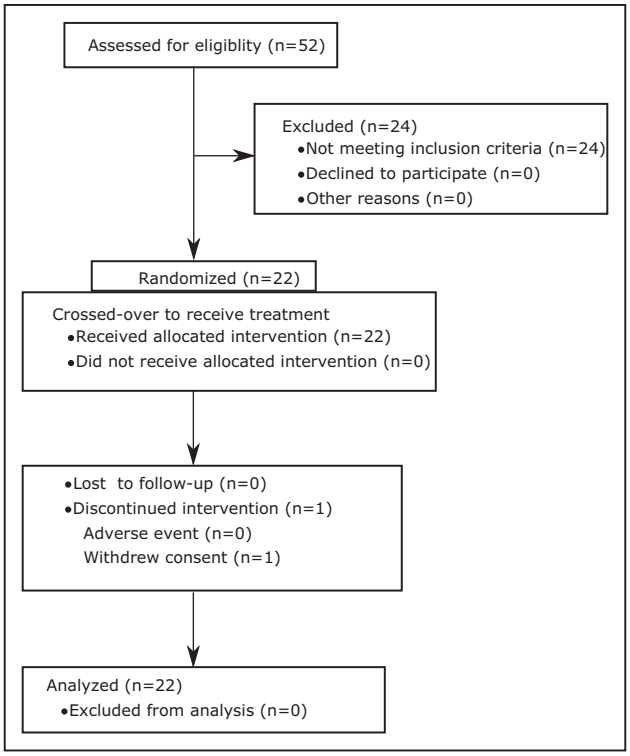

Figure 1. CONSORT (Consolidated Standards of Reporting Trials) diagram for the study as reported in Vicente et al.[8]
Twenty-four of the 52 screened subjects did not meet the inclusion criteria. Twenty-two of the 28 subjects who met the inclusion criteria were randomized. All subjects completed the study except 1 subject who withdrew before the last treatment period.

corrected for heart rate because it has minimal heart rate relationship at rest as previously described.[11] The annotated ECG median beats are available on Physionet at https://physionet.org/physiobank/database/ecgrdvq/.[12] A fully automated algorithm for T_{peak} and T_{end} is also now available at https://github.com/FDA/ecglib.[13]

DNA Extraction

Blood samples for isolation of DNA and genetic testing were collected and spotted onto Whatman FTA blood spot cards (Whatman Inc, Clifton, NJ) by a research team member at check-in of the first period. DNA was extracted from Whatman FTA blood spot cards with Promega Tissue and Hair Extraction (Promega, Inc, Madison, WI) kits. For samples with comparatively low yield, whole genome amplification was performed with the Qiagen REPLI-g Midi Kit (Qiagen, Inc, Venlo, Limburg, the Netherlands). Samples were plated in duplicate from both raw extracted DNA and amplified DNA.

Primer Selection and Design

Sixty-eight single-nucleotide polymorphisms (SNPs) with established independent effects on QT interval from a large GWAS in 76 061 individuals of European descent, all meeting the $P<5\times10^{-8}$ threshold for statistical significance,[4] were targeted for design in 3 multiplex assays with Sequenom custom software. When assays for specific SNPs could not be designed, alternative SNPs that were highly correlated ($r^2>0.90$ to the index SNP) and known to be equally associated with QT interval were attempted. In total, 63 SNPs were designed into 3 multiplexed pools; 5 SNPs could not be designed because of multiplexing limitations.

Genotyping and Quality Control

Sixty-three SNPs were attempted on the Sequenom matrix-assisted laser desorption/ionization time-of-flight platform. DNA with and without whole-genome amplification was tested in duplicate (88 wells for 22 individuals) on 384-well plates, with DNA from an additional 200 individuals genotyped for a separate study. For a given individual in whom 2 samples were genotyped, the sample with the highest genotyping call rate was selected for analysis. Sixty-one SNPs with a call rate >90% and Hardy-Weinberg equilibrium $P>0.001$ across all plated samples (22+200) were retained for further analysis; 2 SNPs failed. The average genotyping success rate across 61 SNPs among 22 study subjects was 95.0%.

Genetic QT Score

A genotype score was calculated as previously described.[6] Briefly, the effects of 61 common variants on QT interval in individuals of European and of African descent were previously estimated in the Arking et al[4] GWAS. We use European or African descent when describing analyses that included genetic inference of continental ancestry and black or white when describing study subject self description. We oriented the coded allele (the allele coded 0, 1, or 2) to be the QT-raising allele for each SNP, regardless of allele frequency. A "simple" score just adding up the QT-increasing alleles across the 61 variants would have a theoretical minimum of 0 QT-prolonging alleles to a maximum of 122 QT-prolonging alleles because everyone has 2 alleles. This approach ignores the fact that not all genetic variants have equal effects on QT interval. Our approach (taken by most others in the genetics community) is to weight each allele by the observed effect on QT from the original 2014 GWAS. This changes the scale of the score from the number of QT-prolonging alleles to the predicted QT increase on the millisecond scale; predicted, not observed, because the weights are taken from the original GWAS, not the present study. The contribution of a given SNP to the QT score was weighted according to the effect estimate per coded allele. For example, rs12143842 is a C/T SNP of which the T allele has a frequency of 0.24 in individuals of European ancestry and is associated with a 3.5-millisecond-longer QT interval per allele copy. An individual homozygous for the major allele (CC) would have 0 copies of the QT-raising allele, and the contribution in that individual for that SNP to the QT score would be 0 (=3.5×0) milliseconds. An individual homozygous for the minor allele (TT) would have 2 copies of the QT-raising allele, and the contribution for that SNP to the QT score would be +7.0 (=3.5×2) milliseconds. This process is then repeated for all 61 SNPs, and the individual SNP contributions are summed. For SNPs with missing genotypes in a given individual, the contribution to the score was imputed from the allele frequency in the general population (twice the allele frequency because every individual has 2 copies of each gene). For example, for

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rs12143842, the coded allele frequency is 0.24, and the average number of coded alleles in individuals in the general population would be 0.24×2=0.48, and thus the contribution of a missing genotype for this SNP would be 1.68 (=3.5×0.48) milliseconds. The effect of such imputation biases the genotype score toward the null.

In self-described white individuals in the present study, we used the allelic effects estimated from the prior GWAS in individuals of European ancestry (Table I in the online-only Data Supplement). As reported in the Arking et al[4] study, an independent African descent had a smaller sample size, and therefore, fewer SNPs reached stringent statistical significance ($P<5\times10^{-8}$), accounting for the genome-wide multiple testing burden. However, we observed high correlation among the effects of SNPs identified in European-derived individuals with effects for the same SNPs estimated in a GWAS in 13105 black individuals (r=0.60).[14] We cannot tell which SNPs among these are truly associated and which are not because of limitations of power; however, the estimates in African descent individuals for null SNPs (not truly associated) will tend to cancel each other out. Therefore, in self-described black individuals in the present study, we used the allelic effects estimated for 60 of the 61 SNPs (1 SNP was unavailable) in the earlier African descent GWAS.[14] The European-derived and African-derived genetic QT scores were calculated in all individuals, regardless of self-described ancestry, for comparison purposes, but ancestry-specific scores were tested as the primary analysis. The PLINK version 1.07 statistical package was used in all QT score calculations. Genotyping, quality control, and genetic QT score calculation were performed by coinvestigators blinded to all clinical data, including race, sex, and QTc response to drug.

Case-Control Analysis of Torsade de Pointes

A GWAS was previously performed on 216 individuals of European descent, with drug-induced torsade de pointes collected as part of the Trans-Atlantic Alliance Against Sudden Death supported by the Fondation Leducq and the DARE study (Drug-Induced Arrhythmia Risk Evaluation) compared with 771 ancestry-matched controls.[15] The control group included a sample of drug-exposed, ancestry-matched controls free of excessive QT prolongation and population-based controls. In the study of rare diseases such as rare adverse drug events, with incidence well below 1%, the frequencies of common variants among population-based controls and among drug-exposed QT nonprolongers are expected to be broadly similar. In the original study of torsade cases, a diversity of potential offending drugs was observed, albeit enriched for users of quinidine, sotalol, and amiodarone. Considering the small number of cases, we used combined sets of drug-exposed and population-based controls to maximize the control size. Using the methods developed by Johnson and reported in Ehret et al,[16] we applied an instrumental variable approach based on the weighted effects from the QT Interval–International GWAS Consortium[4] on the risk of drug-induced torsade de pointes for 60 of the 68 total SNPs that were directly genotyped or imputed with imputation quality >0.90 in the torsade study. In a sensitivity analysis, we repeated the risk score analysis using only 1 SNP per locus (31 index SNPs from 35 possible loci). These analyses were performed in R (R Foundation for Statistical Computing, Vienna, Austria) with the "gtx" package (version 0.0.8) available at https://cran.r-project.org/web/packages/gtx/index.html.

Statistical Analysis

Personalized ECG response to drug was defined as the slope of an individual subject's drug-induced change in ECG biomarker (Figure 2A–2C). This was calculated by inputting individual-subject baseline (triplicate ECG measurements obtained immediately before dosing of a specific drug) and placebo-corrected (time of day–matched ECG measurement from the placebo day) change (ΔΔQTc) for each of the ECG biomarkers and plasma drug concentrations into PROC MIXED in SAS 9.3 (SAS Institute Inc, Cary, NC), with concentration as a fixed effect and subject as a random effect on concentration (ie, with each subject having his or her own slope with an intercept set to 0). The association between biomarkers (eg, ΔΔQTc/drug concentration slope versus genetic QT score) was tested with the Pearson product-moment correlation coefficient in R 3.1.2. The crossover design was not formally accounted for in the statistical analysis, except for calculating placebo-corrected change from baseline for all ECG biomarker measurements. Values of $P<0.05$ were considered statistically significant.

RESULTS

The drug study included 17 self-described white subjects, 4 black subjects, and 1 Asian subject free of electrolyte abnormality, concomitant medication use, or clinically apparent cardiovascular disease (Table 1). The white group included 8 men and 9 women with a mean age of 26 years. The European genetic score explained 27% of the variability in baseline QTc in white subjects (P=0.03; Figure 2F). The black genetic score was also correlated with baseline QTc in African subjects (P=0.03), although the small sample size limits precise estimation of the effect (Table II in the online-only Data Supplement).

Baseline QTc was not a significant predictor of drug-induced QTc prolongation for any of the drugs in 17 white subjects, potentially as a result of limited power (Figure 2G and Figure I in the online-only Data Supplement). However, there was a significant correlation between the genetic QT score and drug-induced QTc prolongation (Table 2 and Figure I in the online-only Data Supplement). Among white subjects, European genetic score explained 30% of the variability (P=0.02) in response to dofetilide (Figure 2H), 23% in response to quinidine (P=0.06), and 27% in response to ranolazine (P=0.03). Among 4 black subjects, a significant correlation existed between baseline QTc and response to dofetilide (P=0.04; Table II in the online-only Data Supplement) and between the African genetic score and response to dofetilide (P=0.03, Table 2) but not for quinidine or ranolazine.

We next investigated how response to 1 QT-prolonging drug predicted the response to other QT-prolonging drugs, combining subjects of all races together. There were significant correlations between all drug-drug rela-

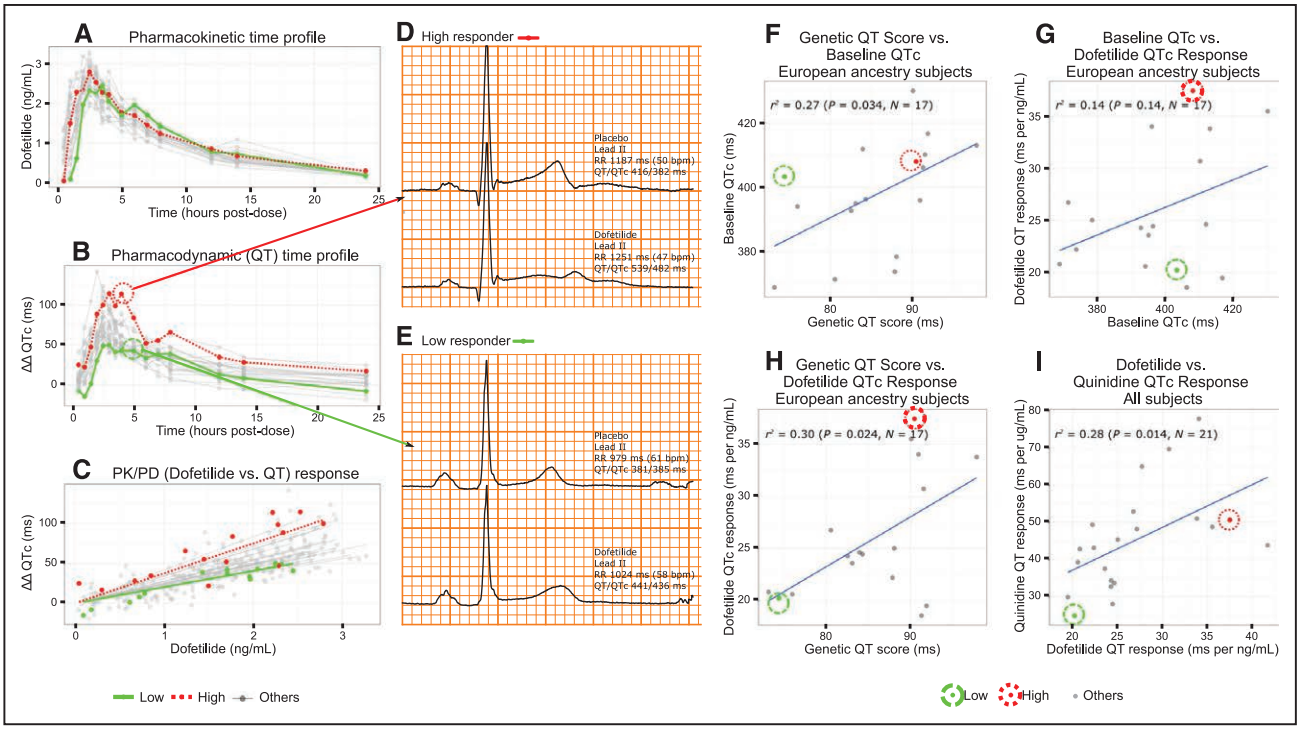

Figure 2. Pharmacokinetic/pharmacodynamic (PK/PD) response and genetic QT score.
A, Pharmacokinetic time profile shows plasma dofetilide concentration at each of the 15 time points after dose (dots) for each subject (lines). Example subjects are shown in red (dofetilide high responder) and green (low responder) throughout. **B**, Pharmacodynamic time profile shows baseline- and placebo-corrected changes from baseline in heart rate–corrected QT (ΔΔQTc) at 15 time points (dots) after a single oral dose of dofetilide for each subject (lines). **C**, PK/PD response plot showing the measures of ΔΔQTc from the ECGs and the corresponding time-matched dofetilide plasma concentration. Solid lines show each subject's QTc concentration-dependent response, the slope of which was tested in genetic QT score analyses. ECG examples show lead II and QT/QTc measures of (**D**) a high responder subject (red line and dots in **A**–**C**) during placebo (**top** ECG) and dofetilide (**bottom** ECG) and (**E**) a low responder subject (green line and dots in **A**–**C**) during placebo (**top** ECG) and dofetilide (**bottom** ECG). Note that although lead II is shown, QT measurements are from the global vector magnitude lead as described in Methods. Correlations between (**F**) genetic QT score and baseline QTc in white subjects, (**G**) baseline QTc and dofetilide QTc response in white subjects, (**H**) genetic QT score and dofetilide QTc response in white subjects, and (**I**) dofetilide QTc response and quinidine QTc response in all subjects are shown. Each dot represents a subject's value. The scale of the QT genetic score is in milliseconds of predicted QT effect for the variants in aggregate, as described in Methods.

tionships, with response to each drug explaining 24% to 29% of the variability in response to each of the other drugs (Figure 2I and Table 3).

Although hERG potassium channel block prolongs both J-T_{peak}c and T_{peak}-T_{end} intervals, additional inward current block from L-type calcium or late sodium current block can shorten the J-T_{peak}c interval.[8,17] Thus, T_{peak}-T_{end} may be a more specific marker for hERG potassium channel block than the entire QT interval.[7,11] Genetic QT score was not associated with baseline T_{peak}-T_{end} or drug-induced change in T_{peak}-T_{end} (Table III in the online-only Data Supplement). Response to each of the 2 strongest hERG potassium channel–blocking drugs (dofetilide and quinidine) explained 52% of the variability in the response to the other (P<0.001; Table 3). Baseline T_{peak}-T_{end} was also correlated with drug-induced QTc prolongation for dofetilide and quinidine but not ranolazine (Table IV in the online-only Data Supplement), and baseline T_{peak}-T_{end} was correlated with drug-induced T_{peak}-T_{end} prolongation for all 3 drugs (Table V in the online-only Data Supplement).

To test the relevance of the impact of the genetic risk score on quantitative drug-induced QT response to the outcome for which QT response is a surrogate, we examined a previously published GWAS of drug-induced torsade de pointes.[15] From a GWAS in 216 individuals with drug-induced torsade de pointes of European descent compared with 771 ancestry-matched controls, 60 of 68 possible QT SNPs had adequate imputation quality or were directly genotyped and available for analysis (Table VI in the online-only Data Supplement). Increasing genetic QT risk score was associated with significantly increased risk of drug-induced torsade de pointes (P=1.3×10^{-7}), explaining 12.1% of variation in risk (Figure 3).

In a sensitivity analysis restricted to 1 SNP per locus, for which 31 SNPs at 35 loci were available, the genetic risk score explained a smaller proportion of variance in

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Table 1. Baseline Characteristics

	All	White	Black	Asian
Age, y	26.9±5.5	25.7±5.3	30.3±3.8	35.0
BMI, kg/m^2	23.1±2.7	22.5±2.7	25.3±1.0	23.1
QTc, ms	395.9±17.1	398.0±17.2	389.5±19.0	385.5
European genetic QT score, ms	86.3±6.4	85.8±6.9	88.8±5.2	84.2
African genetic QT score, ms	53.1±4.8	53.4±5.2	51.9±3.4	51.3
Total subjects, n	22	17	4	1
Female, n	11	9	2	0

Age, body mass index (BMI), heart rate–corrected QT (QTc), and genetic QT score values are reported as mean±SD.

drug-induced QT prolongation, and significance was attenuated (Table VII in the online-only Data Supplement), but it remained a significant predictor of torsade risk (P=3×10^{-6}, r^2=9.6%; Figure II in the online-only Data Supplement).

DISCUSSION

Drug-induced QT prolongation and torsade de pointes have resulted in the withdrawal of several drugs from the market, and >150 are listed on CredibleMeds.org as being associated with QT prolongation and/or torsade de pointes.[18] However, the incidence of torsade de pointes is low, and only a small number of patients develop drug-induced long-QT syndrome. The present pilot study provides a link between common genetic variants and drug-induced QT prolongation and demonstrates how GWAS results can be leveraged to define personalized pharmacodynamic response to drugs. Moreover, our finding that these same common genetic variants influence risk of drug-induced torsade de pointes confirms the potential clinical relevance of the genetic QT score.

A genetic component of long-QT syndrome has been recognized since the 1950s,[19] with the molecular basis of rare genetic variants causing congenital long-QT syndrome first identified in the 1990s. However, not all individuals with congenital long-QT syndrome variants have prolonged QT intervals at baseline, a hallmark of incomplete penetrance or expression of the genetic abnormality. Recent GWASs have identified >60 common genetic variants that individually have small effects on QT at baseline (eg, 1 to 3 milliseconds) but in aggregate may have a larger effect. Individual SNPs at the *NOS1AP* locus and at *KCNE1* have been associated with increased risk of acquired long-QT syndrome.[20,21] Indeed, in the present study, we demonstrated that a weighted combination of 61 common genetic variants explained 27% of the variability in baseline QTc. This common genetic variability may help explain not only the incomplete penetrance of congenital long-QT syndrome[22–24] but also why only certain individuals without recognized congenital long-QT syndrome develop drug-induced long-QT syndrome and torsade de pointes.

Previous reports have suggested that patients developing drug-induced long-QT syndrome with 1 drug are more likely to develop drug-induced long QT syndrome with exposure to other drugs.[25] In addition, Kannankeril et al[26] studied the effects of quinidine on drug-induced QTc and T_{peak}-T_{end} prolongation in first-degree relatives of patients who developed drug-induced long-QT syndrome, including torsade de pointes, compared with relatives of patients who tolerated QT-prolonging therapy. Having a relative with drug-induced long-QT syndrome was associated with exaggerated T_{peak}-T_{end} prolongation, but not QTc prolongation, compared with having a drug-tolerant

Table 2. Correlations Between Common Genetic Variant QT Score and Drug-Induced QTc Response

	r (95% CI)	*P*	n	r^2
European genetic QT score vs treatment (white subjects)				
Genetic score vs baseline QTc	0.52 (0.05 to 0.80)	0.03	17	0.27
Genetic score vs dofetilide QTc slope	0.55 (0.09 to 0.81)	0.02	17	0.30
Genetic score vs quinidine QTc slope	0.48 (−0.03 to 0.79)	0.06	16	0.23
Genetic score vs ranolazine QTc slope	0.52 (0.05 to 0.80)	0.03	17	0.27
African genetic QT score vs. treatment (black subjects)				
Genetic score vs baseline QTc	0.97 (0.11 to 1.00)	0.03	4	0.94
Genetic score vs dofetilide QTc slope	0.97 (0.12 to 1.00)	0.03	4	0.94
Genetic score vs quinidine QTc slope	0.18 (−0.94 to 0.97)	0.82	4	0.03
Genetic score vs ranolazine QTc slope	0.55 (−0.87 to 0.99)	0.45	4	0.30

CI indicates confidence interval; and QTc, heart rate–corrected QT. Figure I in the online-only Data Supplement shows the corresponding correlation plots.

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Table 3. Correlations Between Responses to Different Drugs

Drug A vs Drug B, by ECG Measure (All Subjects)	*r* (95% CI)	*P*	n	r^2
QTc				
Dofetilide vs quinidine	0.53 (0.13–0.78)	0.01	21	0.28
Dofetilide vs ranolazine	0.49 (0.09–0.76)	0.02	22	0.24
Quinidine vs ranolazine	0.53 (0.13–0.78)	0.01	21	0.29
J-T_{peak}c				
Dofetilide vs quinidine	0.46 (0.03–0.74)	0.04	21	0.21
Dofetilide vs ranolazine	0.54 (0.15–0.78)	0.009	22	0.29
Quinidine vs ranolazine	0.51 (0.11–0.77)	0.02	21	0.26
T_{peak}-T_{end}				
Dofetilide vs quinidine	0.72 (0.41–0.88)	<0.001	21	0.52
Dofetilide vs ranolazine	0.44 (0.03–0.73)	0.04	22	0.20
Quinidine vs ranolazine	0.57 (0.19–0.80)	0.007	21	0.33

CI indicates confidence interval. Drug A versus drug B correlations were computed by comparing the slopes of each measure.

relative, although the sample size was limited.[26] This is consistent with our recent findings that global T_{peak}-T_{end} measured in the vector magnitude lead may be a more specific biomarker than QT prolongation for hERG potassium channel block,[7,8,11] and in the present study, response to the 2 strongest hERG blockers (dofetilide and

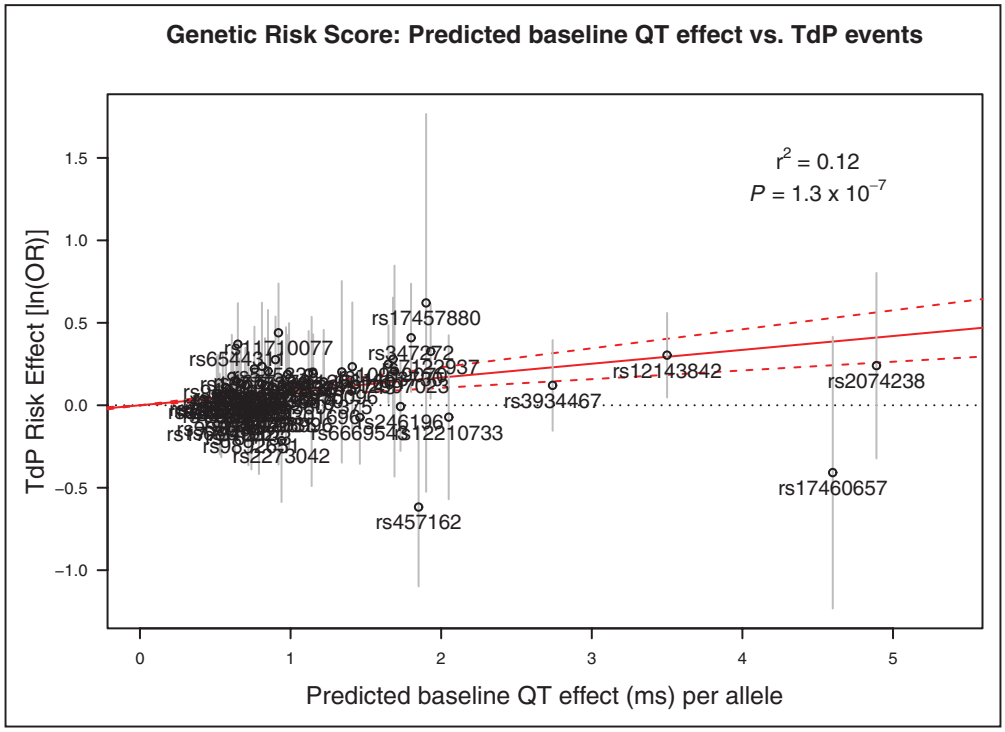

Figure 3. Validation of genotype score in cases of drug-induced torsade de pointes (TdP).
Instrumental variable analysis of effect of 60 single-nucleotide polymorphisms (SNPs) associated with resting QTc using effect estimates from the QT Interval–International GWAS Consortium (QT-IGC) genome-wide association study (*x* axis) in milliseconds of predicted QT interval per allele as a predictor of log odds ratio of drug-induced TdP (*y* axis). Individual labels represent SNPs used in the analysis, and error bars correspond to the standard error of the log odds ratio [ln(OR)] of drug-induced TdP. For example, the QT-raising allele of SNP rs12143842 is associated with a 3.5-millisecond-longer QT interval (Table I in the online-only Data Supplement) and a ln(OR) of 0.30, corresponding to an OR of 1.35 for TdP risk (Table VI in the online-only Data Supplement). The overall r^2 and *P* value reflect the effect on TdP risk of all variants combined in the score.

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quinidine) explained 52% of the variability in response to the other. However, the genetic QT score was not associated with baseline or drug-induced T_{peak}-T_{end} prolongation. This is not surprising because the common genetic variants were selected for association with the whole QT interval, not just the T_{peak}-T_{end} component. Nonetheless, the relationship between T_{peak}-T_{end} measurements at baseline and individual-subject drug response suggests that further study should investigate the relationship between T_{peak}-T_{end}, common genetic variants and risk.

Repolarization reserve, as originally proposed,[27,28] suggests that multiple redundant mechanisms contribute to repolarization such that minor alterations (eg, from genetic variants) may not be detectable at baseline. However, in the presence of additional insults such as hypokalemia or exposure to a drug, reduced repolarization reserve can be unmasked, resulting in an extreme drug response that can lead to ventricular arrhythmias.[29] This model has been considered largely in the context of mendelian long-QT syndromes, in which some ion channel mutation carriers only manifest life-threatening arrhythmia after drug exposure. Although cases of subclinical Mendelian long-QT syndrome exposed by the development of torsade de pointes on drug challenge are well recognized, they appear to represent a minority of cases of drug-induced long-QT syndrome.[30–32] Our genetic and drug A versus drug B response findings strongly support that a significant proportion of repolarization reserve[27,28] in apparently healthy subjects has a genetic basis and that a relatively modest number of common variants—many in genes without an established role in mendelian long-QT syndromes—in aggregate play a substantial role. That the genetic QT score is associated with increased risk of drug-induced torsade de pointes supports the clinical relevance of these variants and confirms the established relationship between QT prolongation after drug exposure and torsade de pointes risk. However, precise quantification of risk of torsade de pointes will be challenging because of the rarity of the outcome and the modest sample size of existing case-control collections.

The present study is limited by the small sample size, especially for blacks, and attempted replication is needed to confirm the findings in individuals of European descent, to provide more precise estimates of effects, and to perform adequately powered tests in individuals of African and other non-European ancestries. The study was conducted in healthy volunteers as opposed to patients, in whom sources of variation in QT response may be greater. However, the study represents a proof of principle that common genetic variants in aggregate influence QT response by administering multiple QT-prolonging drugs to the same subjects in a phase 1 clinical trial unit with pharmacokinetic/pharmacodynamic modeling to precisely define personalized response. We have imputed results for missing genotypes, although this is expected to bias results to the null. In addition, aggregating individual effects of variants in genes in diverse pathways does not establish which variants drive the risk of QT prolongation and torsade. We took a genetic risk score approach to maximize power under a model in which QT-prolonging alleles generally increase QT prolongation after drug exposure. Ultimately, much larger sample sizes, including, for example, individuals with the underlying cardiovascular diseases for which antiarrhythmic medications such as those examined here are prescribed, will be required to establish which variants contribute to the predictive ability of the score and the relative explanatory power of a genetic risk score when set against other clinical predictors of QT interval response.

Individualized prediction of risk of adverse response to medication is needed. Our finding that a simple genetic risk score comprising 61 common variants explains a substantial proportion of variation in QT response to multiple drugs highlights the opportunity to translate GWAS findings to clinical care. Genetic risk scores will be expanded as more genetic variants are identified. The present study highlights the value of genetic studies of continuous, quantitative cardiovascular traits measured in very large sample sizes to identify variants that have meaningful effects on clinical outcomes captured in much smaller samples. Studies to examine whether preemptive, prepresCription genotyping leads to a reduction in serious adverse events are warranted.

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DISCLOSURES

None.

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FOOTNOTES

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Common Genetic Variant Risk Score Is Associated With Drug-Induced QT Prolongation and Torsade de Pointes Risk: A Pilot Study

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Supplemental Material

Common Genetic Variants Explain Variability in Drug-Induced QT Prolongation

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Supplementary Table 1: Common genetic variants for QT score

SNP	Chr.	Nearest Gene	QT Raising Allele	European genetic QT score weight	African American genetic QT score weight	0 copies (%)	1 copy (%)	2 copies (%)	Missing (%)
rs10919070	1	ATP1B1	A	1.668	-2.063	0	13.6	86.4	0
rs11809180	1	ATP1B1	C	1.163	-1.574	0	18.2	77.3	4.5
rs12061601	1	ATP1B1	T	1.38	-1.888	9.1	13.6	72.7	4.5
rs1983546	1	ATP1B1	A	0.8174	-0.6946	36.4	31.8	31.8	0
rs545833	1	ATP1B1	T	0.8518	0.4236	59.1	18.2	18.2	4.5
rs12025136	1	NOS1AP	C	1.449	0.0336	50	18.2	27.3	4.5
rs12143842	1	NOS1AP	T	3.489	3.144	50	31.8	0	18.2
rs164133	1	NOS1AP	C	0.706	-0.2965	50	40.9	4.5	4.5
rs16857031	1	NOS1AP	G	2.365	0.7958	68.2	9.1	4.5	18.2
rs17460657	1	NOS1AP	A	4.997	0.3507	0	13.6	81.8	4.5
rs347272	1	NOS1AP	A	1.804	-0.3486	59.1	22.7	0	18.2
rs3934467	1	NOS1AP	T	2.759	1.688	59.1	22.7	0	18.2
rs4656345	1	NOS1AP	G	4.845	-4.709	4.5	0	86.4	9.1
rs2273042	1	RNF207	A	0.9223	-0.6483	77.3	13.6	0	9.1
rs846111	1	RNF207	C	1.69	0.6535	59.1	27.3	0	13.6
rs2298632	1	TCEA3	T	0.7924	0.6809	45.5	31.8	18.2	4.5
rs12997023	2	SLC8A1	T	1.694	-1.675	4.5	4.5	77.3	13.6
rs6544311	2	SLC8A1	A	0.6505	-0.9235	50	40.9	9.1	0
rs938291	2	SP3	G	0.5482	0.3565	27.3	36.4	13.6	22.7
rs295140	2	SPATS2L	T	0.5534	0.358	31.8	40.9	13.6	13.6
rs7561149	2	TTN-CCDC141	T	0.5287	-0.3299	18.2	22.7	45.5	13.6
rs17784882	3	C3ORF75	C	0.5342	0.1658	13.6	22.7	54.5	9.1
rs11708996	3	SCN5A-SCN10A	G	0.9123	-2.919	0	18.2	77.3	4.5
rs6793245	3	SCN5A-SCN10A	G	1.107	-0.5121	13.6	27.3	45.5	13.6
rs6801957	3	SCN5A-SCN10A	C	0.6181	-0.9547	4.5	31.8	50	13.6
rs9851710	3	SCN5A-SCN10A	C	0.6628	0.1167	45.5	31.8	9.1	13.6
rs2363719	4	SLC4A4	A	0.9567	-0.5002	86.4	9.1	0	4.5
rs3857067	4	SMARCAD1	T	0.5091	-0.1312	18.2	31.8	27.3	22.7
rs10040989	5	GFRA3	G	0.8571	0.0051	4.5	9.1	86.4	0
rs7765828	6	GMPR	G	0.6208	-0.0931	40.9	27.3	27.3	4.5
rs10499087	6	SLC35F1-PLN	C	0.7001	0.1828	72.7	9.1	4.5	13.6
rs11153730	6	SLC35F1-PLN	C	1.647	-0.565	40.9	9.1	45.5	4.5

rs12210733	6	SLC35F1-PLN	G	2.036	0.6448	0	4.5	95.5	0
rs17349133	6	SLC35F1-PLN	C	0.857	-0.4496	0	36.4	54.5	9.1
rs465226	6	SLC35F1-PLN	T	1.844	0.1849	0	0	100	0
rs9920	7	CAV1	C	0.8447	-0.1587	63.6	4.5	0	31.8
rs1805121	7	KCNH2	C	1.278	NA	22.7	40.9	36.4	0
rs2072413	7	KCNH2	C	1.673	-1.343	9.1	36.4	40.9	13.6
rs1961102	8	AZIN1	T	0.5836	0.3118	59.1	18.2	13.6	9.1
rs16936870	8	NCOA2	A	0.9739	0.6432	77.3	18.2	4.5	0
rs2485376	10	GBF1	G	0.5629	-0.0445	22.7	36.4	36.4	4.5
rs174583	11	FADS2	C	0.6575	-0.6151	4.5	27.3	54.5	13.6
rs2074238	11	KCNQ1	C	4.94	-3.112	0	9.1	90.9	0
rs7122937	11	KCNQ1	T	1.928	1.347	45.5	18.2	31.8	4.5
rs3026445	12	ATP2A2	C	0.5717	0.474	40.9	36.4	13.6	9.1
rs728926	13	KLF12	T	0.5746	0.3524	45.5	22.7	27.3	4.5
rs2273905	14	ANKRD9	T	0.6938	1.096	59.1	27.3	13.6	0
rs3105593	15	USP50-TRPM7	T	0.67	0.9802	54.5	31.8	13.6	0
rs246258	16	CNOT1	C	1.732	-1.392	0	31.8	50	18.2
rs4784934	16	CNOT1	A	0.6815	0.3654	63.6	18.2	9.1	9.1
rs1296720	16	CREBBP	C	0.834	0.5728	81.8	4.5	9.1	4.5
rs12444261	16	LITAF	G	0.7988	-0.888	4.5	22.7	68.2	4.5
rs735951	16	LITAF	G	1.156	-1.436	27.3	36.4	22.7	13.6
rs246185	16	MKL2	C	0.7205	0.373	50	27.3	13.6	9.1
rs10775360	17	KCNJ2	C	0.7672	-0.2469	22.7	13.6	50	13.6
rs1396515	17	KCNJ2	G	0.9762	-0.4522	13.6	13.6	54.5	18.2
rs17763769	17	KCNJ2	A	0.8944	-0.5354	77.3	13.6	4.5	4.5
rs236586	17	KCNJ2	G	0.6408	1.002	45.5	36.4	13.6	4.5
rs1052536	17	LIG3	C	0.9715	0.8081	22.7	22.7	27.3	27.3
rs9892651	17	PRKCA	T	0.7387	-0.7171	31.8	27.3	22.7	18.2
rs1805128	21	KCNE1	T	1.014	14.18	100	0	0	0

Supplementary Table 2: Baseline QTc vs. drug slope response by race

Group	*r* [95% CI]	*P*	*N*	r^2
All Subjects				
Baseline QTc vs. Dofetilide QTc slope	0.45 [0.03 to 0.73]	0.04	22	0.20
Baseline QTc vs. Quinidine QTc slope	<0.01 [-0.46 to 0.40]	0.89	21	<0.01
Baseline QTc vs. Ranolazine QTc slope	0.18 [-0.26 to 0.56]	0.43	22	0.03
White				
Baseline QTc vs. Dofetilide QTc slope	0.38 [-0.13 to 0.73]	0.14	17	0.14
Baseline QTc vs. Quinidine QTc slope	<0.01 [-0.53 to 0.46]	0.86	16	<0.01
Baseline QTc vs. Ranolazine QTc slope	0.03 [-0.46 to 0.50]	0.92	17	<0.01
Black				
Baseline QTc vs. Dofetilide QTc slope	0.96 [-0.02 to 1.00]	0.04	4	0.92
Baseline QTc vs. Quinidine QTc slope	0.42 [-0.91 to 0.98]	0.58	4	0.17
Baseline QTc vs. Ranolazine QTc slope	0.64 [-0.83 to 0.99]	0.36	4	0.41

Asian group not reported because only 1 subject was Asian. Supplementary Figure I shows the corresponding correlation plots.

Supplementary Table 3. Correlations between common genetic variant QT score and drug-induced T_{peak}-T_{end} slope response

Genetic QT score vs. treatment (white subjects)	***r* [95% CI]**	***P***	***N***	r^2
Genetic score vs. Baseline T_{peak}-T_{end}	0.27 [-0.24 to 0.67]	0.29	17	0.07
Genetic score vs. Dofetilide T_{peak}-T_{end} slope	0.13 [-0.37 to 0.58]	0.61	17	0.02
Genetic score vs. Quinidine T_{peak}-T_{end} slope	0.27 [-0.26 to 0.68]	0.31	16	0.07
Genetic score vs. Ranolazine T_{peak}-T_{end} slope	0.38 [-0.12 to 0.73]	0.13	17	0.14
Genetic QT score vs. treatment (black or African American subjects)	***r* [95% CI]**	***P***	***N***	r^2
Genetic score vs. Baseline T_{peak}-T_{end}	0.87 [-0.56 to 1.00]	0.13	4	0.76
Genetic score vs. Dofetilide T_{peak}-T_{end}	0.86 [-0.58 to 1.00]	0.14	4	0.74
Genetic score vs. Quinidine T_{peak}-T_{end} slope	<0.01 [-0.99 to 0.86]	0.41	4	0.35
Genetic score vs. Ranolazine T_{peak}-T_{end} slope	0.94 [-0.24 to 1.00]	0.06	4	0.88

Supplementary Table 4: Baseline T_{peak}-T_{end} vs. drug QTc slope response by race

Group	*r* [95% CI]	*P*	*N*	r^2
All Subjects				
Baseline T_{peak}-T_{end} vs. Dofetilide QTc slope	0.59 [0.23 to 0.81]	<0.01	22	0.35
Baseline T_{peak}-T_{end} vs. Quinidine QTc slope	0.78 [0.52 to 0.91]	<0.001	21	0.61
Baseline T_{peak}-T_{end} vs. Ranolazine QTc slope	0.37 [-0.06 to 0.69]	0.09	22	0.14
White				
Baseline T_{peak}-T_{end} vs. Dofetilide QTc slope	0.65 [0.24 to 0.86]	<0.01	17	0.42
Baseline T_{peak}-T_{end} vs. Quinidine QTc slope	0.80 [0.50 to 0.93]	<0.001	16	0.64
Baseline T_{peak}-T_{end} vs. Ranolazine QTc slope	0.41 [-0.09 to 0.74]	0.11	17	0.17
Black				
Baseline T_{peak}-T_{end} vs. Dofetilide QTc slope	0.96[-0.07 to 1.00]	0.05	4	0.91
Baseline T_{peak}-T_{end} vs. Quinidine QTc slope	0.17 [-0.95 to 0.97]	0.83	4	0.03
Baseline T_{peak}-T_{end} vs. Ranolazine QTc slope	0.11 [-0.95 to 0.97]	0.89	4	0.01

Asian group not reported because only 1 subject was Asian.

Supplementary Table 5: Baseline T_{peak}-T_{end} vs. drug T_{peak}-T_{end} slope response by race

Group	*r* [95% CI]	*P*	*N*	r^2
All Subjects				
Baseline T_{peak}-T_{end} vs. Dofetilide T_{peak}-T_{end} slope	0.72 [0.43 to 0.88]	<0.001	22	0.52
Baseline T_{peak}-T_{end} vs. Quinidine T_{peak}-T_{end} slope	0.66 [0.33 to 0.85]	<0.01	21	0.44
Baseline T_{peak}-T_{end} vs. Ranolazine T_{peak}-T_{end} slope	0.50 [0.10 to 0.76]	0.02	22	0.25
White				
Baseline T_{peak}-T_{end} vs. Dofetilide T_{peak}-T_{end} slope	0.69 [0.31 to 0.88]	<0.01	17	0.47
Baseline T_{peak}-T_{end} vs. Quinidine T_{peak}-T_{end} slope	0.69 [0.30 to 0.89]	<0.01	16	0.48
Baseline T_{peak}-T_{end} vs. Ranolazine T_{peak}-T_{end} slope	0.40 [-0.10 to 0.74]	0.12	17	0.16
Black				
Baseline T_{peak}-T_{end} vs. Dofetilide T_{peak}-T_{end} slope	1.00[0.88 to 1.00]	0<0.01	4	0.99
Baseline T_{peak}-T_{end} vs. Quinidine T_{peak}-T_{end} slope	<0.01 [-0.98 to 0.92]	0.65	4	0.13
Baseline T_{peak}-T_{end} vs. Ranolazine T_{peak}-T_{end} slope	0.89 [-0.50 to 1.00]	0.11	4	0.79

Asian group not reported because only 1 subject was Asian.

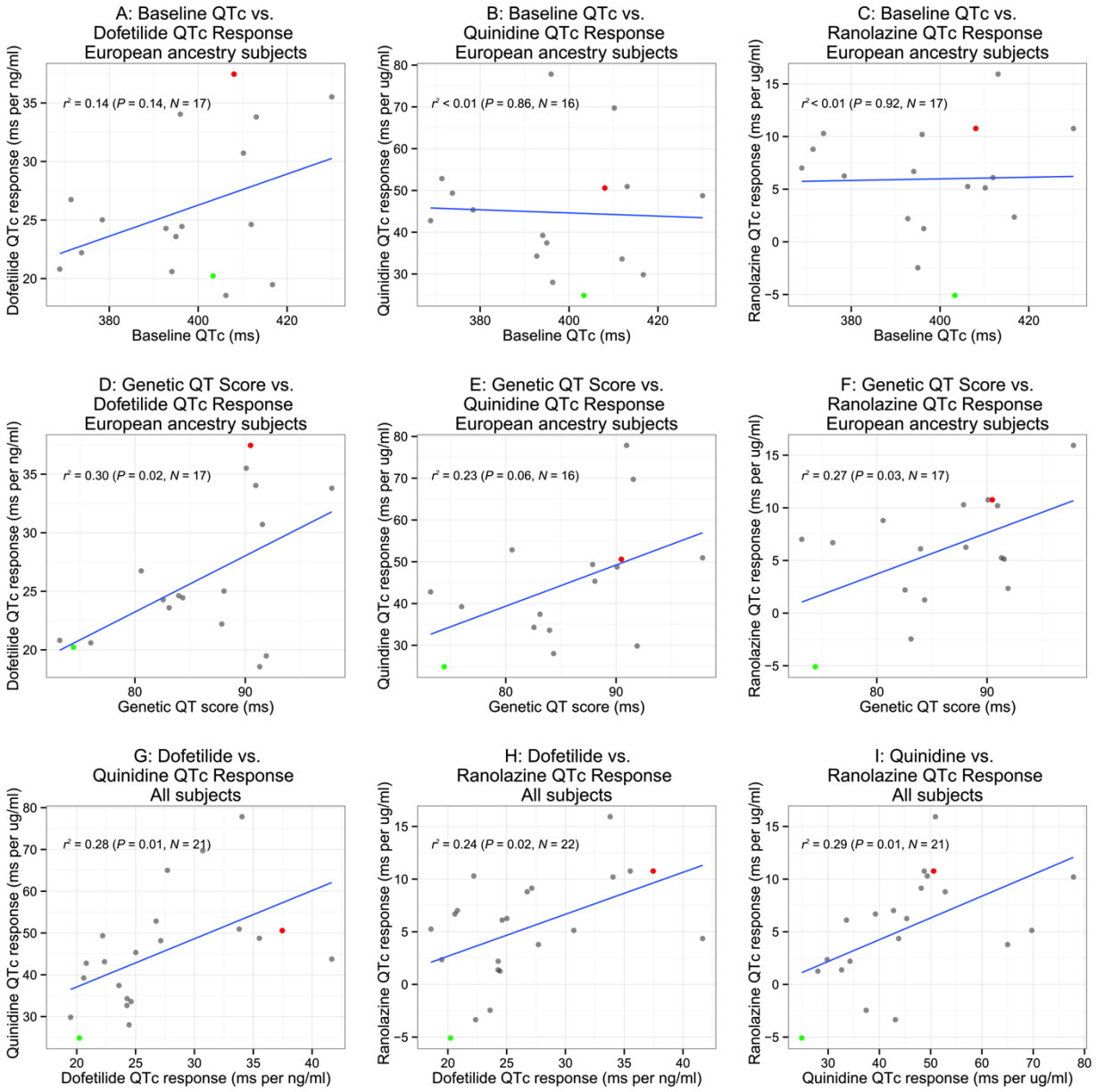
Supplementary Table 6. Association results for individual torsade de pointes variants from the QT interval score. Variants with imputation quality scores > 0.9 were included in the QT score analysis.

SNP	CHR	position (hg19)	coded allele	coded allele freq	effect (ln(OR))	SE	P	imputed	imputation quality
rs2273042	1	6149122	A	0.12	-0.22	0.19	0.256	0	1.00
rs846111	1	6279370	C	0.19	-0.18	0.22	0.422	1	0.76
rs2298632	1	23710475	C	0.49	-0.12	0.12	0.289	0	1.00
rs6669543	1	161981025	T	0.24	-0.07	0.14	0.608	1	0.94
rs4656345	1	161991237	NA	NA	NA	NA	NA	NA	NA
rs12143842	1	162033890	T	0.27	0.30	0.13	0.020	0	0.98
rs16857031	1	162112910	G	0.15	0.11	0.16	0.493	1	0.58
rs17457880	1	162168154	A	0.01	-0.62	0.58	0.288	1	1.00
rs4657172	1	162179632	C	0.11	-0.24	0.20	0.231	1	0.99
rs3934467	1	162182677	T	0.22	0.12	0.14	0.390	1	0.99
rs7545047	1	162191103	A	0.05	-0.11	0.28	0.684	1	0.59
rs17460657	1	162261826	C	0.02	0.41	0.42	0.331	1	0.99
rs347272	1	162318498	A	0.14	0.41	0.17	0.015	1	1.00
rs164133	1	162381288	C	0.27	-0.11	0.13	0.418	1	1.00
rs545833	1	168689940	T	0.28	0.28	0.13	0.039	0	1.00
rs12061601	1	169070450	C	0.11	-0.23	0.20	0.242	0	1.00
rs10919070	1	169099037	C	0.13	-0.28	0.19	0.143	1	0.99
rs12079745	1	169101060	A	0.05	-0.20	0.28	0.471	1	1.00
rs1983546	1	169446183	G	0.35	-0.13	0.12	0.289	0	0.99
rs6544311	2	40353277	A	0.39	0.37	0.13	0.003	1	1.00
rs12997023	2	40752982	C	0.04	-0.21	0.33	0.525	1	0.99
rs938291	2	174742608	G	0.38	-0.05	0.12	0.671	1	1.00
rs7561149	2	179689856	C	0.40	-0.07	0.12	0.551	1	1.00
rs295140	2	201160699	T	0.43	0.07	0.12	0.552	0	0.99
rs6793245	3	38599037	A	0.31	-0.20	0.13	0.134	1	1.00
rs11708996	3	38633923	C	0.14	0.02	0.17	0.884	1	0.94
rs11710077	3	38657899	T	0.20	0.44	0.15	0.004	1	0.98
rs6599234	3	38715300	A	0.30	0.03	0.13	0.801	1	1.00
rs6801957	3	38767315	T	0.41	0.08	0.12	0.525	1	1.00
rs17784882	3	47544003	A	0.41	0.08	0.12	0.504	0	1.00
rs2363719	4	72138216	A	0.11	0.10	0.19	0.581	0	1.00
rs3857067	4	95026434	A	0.49	-0.05	0.12	0.670	1	1.00
rs10040989	5	137573725	A	0.13	-0.21	0.19	0.275	0	0.95
rs7765828	6	16294722	G	0.37	0.06	0.13	0.624	1	1.00
rs457162	6	118535983	T	0.05	0.62	0.25	0.012	1	1.00
rs12210733	6	118653075	A	0.06	0.07	0.25	0.775	1	1.00
rs11153730	6	118667522	C	0.49	0.25	0.12	0.038	1	1.00

rs3902035	6	119000232	C	0.32	0.03	0.12	0.835	1	1.00
rs9489510	6	119043898	G	0.32	0.18	0.13	0.172	0	1.00
rs9920	7	116200092	C	0.10	-0.03	0.20	0.885	0	1.00
rs2072413	7	150647969	NA	NA	NA	NA	NA	NA	NA
rs3807375	7	150667210	T	0.37	0.08	0.12	0.495	1	0.99
rs16936870	8	71189342	A	0.10	0.12	0.20	0.552	1	1.00
rs11779860	8	98850330	C	0.46	-0.06	0.12	0.624	0	1.00
rs1961102	8	103932845	T	0.35	0.08	0.12	0.537	1	0.52
rs2485376	10	104050006	A	0.37	-0.05	0.13	0.674	1	0.82
rs2301696	11	2426984	C	0.47	0.02	0.26	0.930	1	0.98
rs2074238	11	2484803	T	0.06	-0.24	0.29	0.403	1	0.98
rs7122937	11	2486550	T	0.19	0.33	0.15	0.026	1	1.00
rs174583	11	61609750	T	0.36	-0.03	0.12	0.804	0	1.00
rs3026445	12	110723203	C	0.36	0.07	0.13	0.550	0	0.98
rs728926	13	74513122	T	0.39	0.11	0.12	0.384	1	1.00
rs2273905	14	102974999	T	0.35	0.05	0.12	0.655	0	1.00
rs3105593	15	50845018	T	0.47	0.04	0.12	0.736	0	0.95
rs1296720	16	3873642	C	0.21	0.12	0.15	0.402	1	1.00
rs12930096	16	11670758	T	0.17	-0.14	0.16	0.379	0	0.95
rs735951	16	11693536	A	0.46	-0.19	0.12	0.114	1	0.96
rs12444261	16	11734642	T	0.24	0.02	0.15	0.872	1	0.99
rs246185	16	14395432	C	0.32	0.06	0.13	0.662	1	1.00
rs4784934	16	58459926	A	0.28	0.13	0.13	0.311	1	1.00
rs246196	16	58574253	C	0.26	0.01	0.14	0.954	1	1.00
rs1052536	17	33331575	T	0.48	-0.19	0.12	0.125	0	0.99
rs9892651	17	64303793	C	0.42	0.15	0.12	0.199	1	1.00
rs236586	17	68203546	G	0.48	0.15	0.12	0.219	1	1.00
rs10775360	17	68325868	T	0.29	-0.22	0.13	0.091	1	1.00
rs1396515	17	68430993	G	0.48	-0.05	0.12	0.679	0	1.00
rs17763769	17	68560789	A	0.14	0.04	0.17	0.822	0	1.00
rs1805128	21	35821680	NA	NA	NA	NA	NA	NA	NA

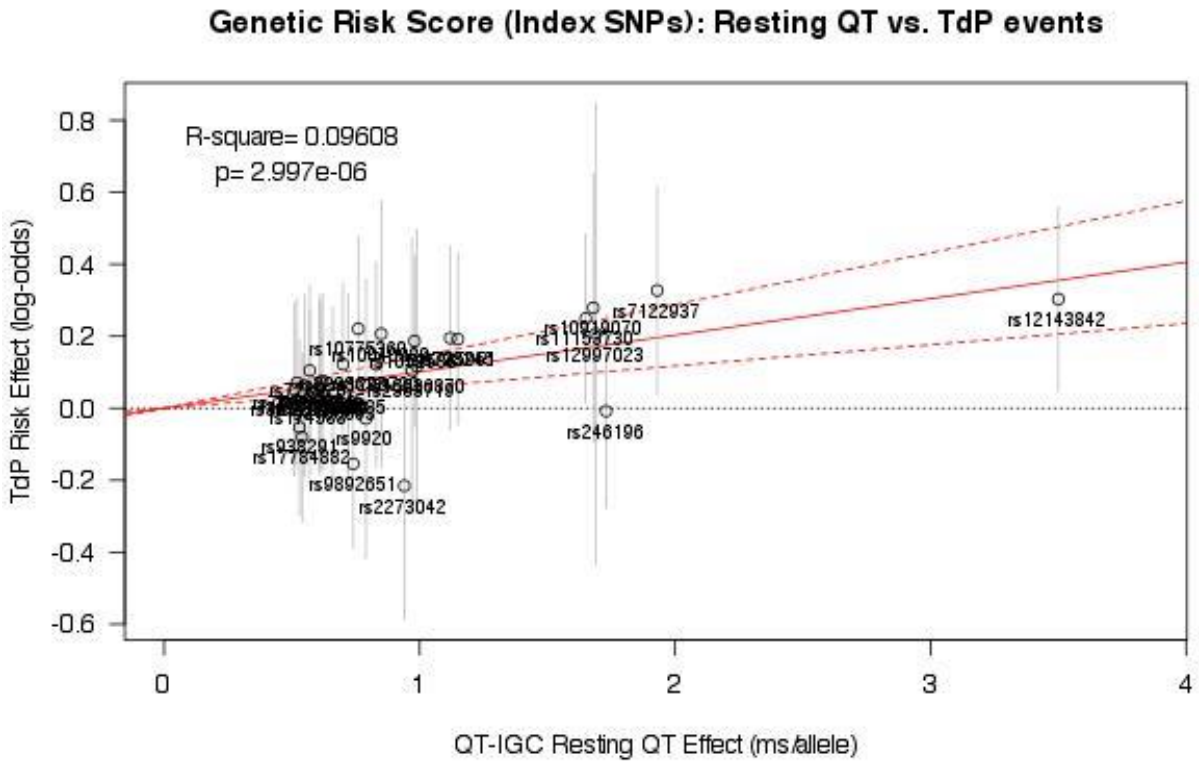
Supplementary Table 7. Effects on QTc slope of genetic QT score using 61 SNPs at 31 loci (full) and restricted to 1 SNP per locus (index only)

		Genetic QT score (white subjects)			
		Full		*Index only*	
Treatment	***N***	***P***	***r^2***	***P***	***r^2***
Baseline QTc	*17*	*0.03*	*0.27*	*0.10*	*0.17*
Dofetilide QTc slope	*17*	*0.02*	*0.30*	*0.19*	*0.11*
Quinidine QTc slope	*16*	*0.06*	*0.23*	*0.45*	*0.04*
Ranolazine QTc slope	*17*	*0.03*	*0.27*	*0.42*	*0.04*



Supplementary Figure 1: Correlations between baseline QTc and QTc slope, genetic score and QTc slope as well as QTc slope between drugs. Correlation between baseline QTc and QTc slope of dofetilide (A), quinidine (B) and ranolazine (C) in white subjects. Correlation between European ancestry genetic QT score and QTc slope of dofetilide (D), quinidine (E) and ranolazine (F) in white subjects. Correlation between dofetilide and quinidine QTc slopes (G), dofetilide and ranolazine QTc slope (H) and quinidine and ranolazine QTc slopes (I). All correlations computed in white subjects. Each dot corresponds with a subject. Example subjects are shown in red (dofetilide high responder) and green (low responder).

Supplementary Figure 2. Sensitivity analysis of genotype score in cases of drug-induced torsade de pointes. Instrumental variable analysis of effect of 31 SNPs associated with resting QTc (restricted to only one SNP per locus), using effect estimates from the QT-IGC GWA study (x axis) in milliseconds of QT interval per allele as a predictor of log odds ratio of diTdP (y axis). Individual labels represent SNPs used in the analysis, and error bars correspond to the standard error of the log odds ratio of drug-induced torsade de pointes.



Carolyn: Welcome to Circulation on the Run, your weekly podcast summary and backstage pass to The Journal and its editor's. I'm Dr. Carolyn Lam, associate editor from the National Heart Center and Duke National University of Singapore. Our Journal this week features important new data telling us that a common genetic variant risk score is associated with risk of drug induced QT prolongation and torsades de pointes.

First, let's give you your summary of this week's journal. The first paper provides both clinical and experimental data to show that the adipokine, retinal binding protein four promotes atherosclerosis. First author, Dr. Liu, corresponding author, Dr. Xia and colleagues from Sun Yat Sen University in Guangzhou, China first evaluated the association between serum retinal binding four levels and the incidents of adverse cardiovascular events in a community based prospective cohort and then examined the effects of retinal protein four gain or loss of function on macrophage foam cell formation and atherogenesis in an apple lipase protein E deficient mouse model. They found, in the clinical cohort study, that base line serum retinal binding protein four level was an independent predictor of incidents of adverse cardiovascular events after adjustment for traditional risk factors.

In the experimental study's, they showed that retinal binding protein four promoted macrophage derived foam cell formation through the activation of scavenger receptor CD36 mediated cholesterol uptake. In turn dependent on June and terminal kinase and signal transducer and activator of transcription one, as well as upstream regulation by the tracing kinase CSRC. These findings, therefore, support the use of retinal binding protein four as a novel biomarker for the prediction of cardiovascular risk. The data also provide insight into the mechanism of action of retinal binding protein four in the path of physiology of atherosclerosis.

The next paper is the first clinical trial, looking at remote ischemic pre conditioning prior to carotid artery stinting in patients with severe carotid artery stenosis. Remote ischemic pre conditioning is a protective, systemic strategy by which cycles of bilateral limb ischemia are applied briefly to confer protection from subsequent severe ischemia and distant organs. First author, Dr. Zhao, corresponding authors, Dr. Ji, and colleagues from Xuanwu Hospital, Capital Medical University in Beijing, China performed a proof of concept, single center, prospective, randomized control trial to assess whether remote ischemic preconditioning was safe and effective in attenuating ischemic injury related to carotid artery stinting in 189 patients with severe carotid artery stenosis. Results show that daily remote ischemic pre conditioning for two weeks, prior to carotid artery stenting, was feasible, safe, well tolerated, and may effectively attenuate secondary brain injury as evidence by a decreased incidence and reduced volumes of new ischemic legions on magnetic residence imaging performed within 48 hours post operation. The clinical implications are that if results are confirmed by future, larger studies, remote ischemic preconditioning may evolve into a nonpharmacological, neuro protective method for inhibiting carotid artery stenosis related cerebral ischemic events.

This potential for clinical translation in discussed in an accompanying editorial by Doctors Bell and Yellen, from University College, London.

The final paper discusses firefighting and the heart. What's the link? Well, cardiovascular events are the leading cause of death amongst firefighters and the risk is known to be substantially increased during fire suppression duties. In the current study, first author Dr. Hunter, corresponding author, Dr. Mills, and colleagues from University of Edinburgh in United Kingdom sought to understand this link better by assessing the effects of simulated fire suppression on measures of cardiovascular health in an open label, randomized cross over study of 19 healthy firefighters. These firefighters performed a standardized training exercise in a fire simulation facility or like duties for 20 minutes. Following each exposure, ex vivo thrombus formation, fibrinolysis, platelet activation and for armed blood flow in response to intra-arterial infusions of endothelium dependent and independent vasodilators were all measured. The authors found that exposure to extreme heat and physical exertion during fire suppression activated platelets, increased thrombus formation, impaired vascular function, and promoted myocardial ischemia and injury in healthy fire fighters. These finding provided pathogenic mechanisms to explain the association between fire suppression activity and acute myocardial infarction in fire fighters.

The implications of these findings for prevention are discussed in an accompanying editorial from Dr. Kales, of Harvard school of Public Health and Dr. Smith from Skidmore College and University of Illinois fire service institute.

Well, those were your summaries. Let's welcome our guests for our feature discussion.

Today's feature paper describes a pilot study that shows that a common genetic variant risk score, is associated with drug induced QT prolongation and torsades de pointes. This paper is so interesting to me because I found that the learning points, at least for me, really extended well beyond the trial itself. I'm so delighted to have with me the co corresponding authors, Dr. David Strauss from the US FDA, as well as Dr. Christopher Newton-Cheh from Massachusetts General Hospital. Welcome, gentlemen.

David: Thanks very much, glad to be here.

Christopher: Thank you, Carolyn.

Carolyn: So, I've always thought that common genetic variants identified via GWAS, for example, are individually very weak effects on medical traits. For example, systolic blood pressure or in this case, QT interval. But what I'm so impressed with this study is that you show, I think for the first time, that even these small effects can add up to clinically meaningful results that are testable or

demonstrable in a trial. David, could you begin by telling us a little bit about this trial and what the primary results were.

David: In the study, we tested the hypothesis that a weighted combination of common genetic variants, contributing to the QT interval at base line, identified through prior GWAS studies, can predict individual response to multiple QT prolonging drugs. We performed a genetic analysis of 22 subjects and a secondary analysis of a randomized, double blind, placebo controlled cross over trial, that included three QT prolonging drugs, with 15 tie matched QT and plasma drug concentration measurements. This allowed us to carefully control for the inter individual differences in pharmacokinetics and just focus on the pharmacodynamics so the direct effect of the drug on the heart.

What we found was, there was a significant correlation between the weighted combination of common genetic variants, which we call the genetic QT score, and drug induced QT prolongation. More specifically, we found that the genetic QT score explained 30 percent of the variability in response to dofetilide, 23 percent in response to quinidine, and 27 in response to ranolazine.

We also investigated how response to one QT prolonging drug predicted the response to other QT prolonging drugs. There were significant correlations between all the drug/drug relationships with response to each drug explaining 24 to 29 percent of the variability in response to each of the other drugs. It's important to note that QT prolongation, by itself, is not harmful. The real concern is torsades de pointes, which can degenerate into ventricular fibrillation and cause sudden death. So, the test, irrelevant to the common genetic variants in predicting drug induced torsades, we then went on to examine a previously published, genome wide association study that included 215 patients with drug induced torsades, compared to 771 ancestry match controls and that prior study that was previously published had found that each individual common genetic variant did not reach genome wide significance, as you suggested, Carolyn. However, when we applied the weighted combination of common genetic variants, we found that the genetic QT risk score was associated with significantly increased risk of drug induced torsade, explaining 12 percent of the variation in risk.

Carolyn: So, my simplistic understanding was more or less there. That these genetic risks of these common variants kind of add up. I'm just curious ... Chris, do you think that this has implications for even other diseases? That's one question. And then secondly, I really appreciated your comment about using an intermediate trait, if you may, of QT interval versus looking at the disease itself of torsade de pointes. Could you give me comments on both these things?

Christopher: The study of intermediate traits, such as, quantitative traits like QT variability on the EKG are, I think very tractable for the study of genetic bases of underlying physiologic processes because we can study so many people. So the original genome wide association study that detected these individually weak genetic

effects could only find them because we studied about 75,000 people who had had genome wide genome typing and QT intervals measured. It requires such large sample sizes to reach p values that are able to distinguish true positive associations from false positive associations, due to the multiple testing burden.

I think a challenge of what to do with these genetic effects once they've been reliably detected is that they do have weak effects and they influence intermediate traits. Nobody really cares whether their QT interval is three milliseconds longer, or three milliseconds shorter. What they care about is hard outcomes, or the likelihood that they'll have a toxic drug response. So, it was a natural follow on to that work to try to test these variants, and we knew that based on their weak effects individually on QT interval in the general population, that it was unlikely that they would individually explain a significant portion of either drug response or torsade. Which is why we aggregated the facts into the weighted score.

I think we tried to examine what we thought were the most proximal, clinically relevant outcomes. Specifically, drug response. QT drug response to drugs that are established to cause QT prolongation and arrhythmias. Whether the QT score will have meaningful or detectable impact on drugs that have much weaker effects on re polarization and risk of torsade, I think, would remain to be seen.

Carolyn: That's really remarkable.

David, how about your perspective of the implications of this? It's so unique that you're actually from the FDA so, why is this important to the FDA?

David: As Chris mentioned, the specific application we studied here, a drug induced QT prolongation and torsade have resulted in the withdrawal of several drugs from the market both in the US and worldwide. Many critical drugs remain on the market that are associated with QT prolongation and torsade…over 100 drugs, likely. What some people may not be familiar with is that at FDA we perform research to move new science into the drug review process and close the gap between scientific innovation and drug review. Like practicing clinicians, we seek to understand inter patient variabilities and we conduct research to better evaluate, benefit, and risk of medications. This is in line with the broader initiative ... the precision medicine initiative, which seeks to move away from the traditional “one size fits all” approach for medical therapy and instead, take into account specific characteristics of individual patients.

People are most familiar with this being applied in oncology and advances in pharmacogenomics have been more limited in other areas with the exception of the genetic bases of metabolism and pharmacokinetics where the traits are often controlled by one or a few genetic mechanisms, rather than the many mechanisms responsible for complex traits and diseases, as Chris discussed. As I mentioned earlier, what was relatively unique about this study is that we were

able to control for the difference in pharmacokinetics and investigate the inter individual differences in the direct effect of drugs on the heart, the pharmacodynamics. We think it's very exciting that a combination of common genetic variants and aggregate can explain a significant portion of the inter individual variability and, as Chris mentioned, this is also important because the incidence of torsade is quite low. Only a small number of patients will develop drug induced torsade. It's possible that in the future analysis of a large number of common genetic variants that can be identified through genome wide association studies as in this case, may help to better define the personalized benefit risk profiles for individual patients.

Carolyn: You've really articulated that remarkably. That's exactly the excitement I think the entire editorial team shared when we read your paper. Thank you so much for it. Maybe just one last question thrown out to both of you, what's the next step? What's in the future.

Christopher: I think one next step, based on this proof of principle study, will be to try to test the impact of these genetic risk scores in real world clinical settings where individual patients with the diversity of different comorbidities and different drug exposures are also receiving QT prolonging drugs. Because that will have the biggest relevance for our patients who faced increased risk of drug toxicity.

David: The issue of cardiac safety of drugs is something that is very important to us at the FDA and we have some parallel initiatives that, in collaboration with other global drugs ... regulatory agencies and industry and academic collaborators ... we are working to develop new cardiac safety evaluation paradigms for new drugs, or existing drugs, that could even be applied in the preclinical setting and really focus on the mechanistic base, pro arrhythmic risk. So, we should have more exciting work coming forward in the near future for better prediction and individualized prediction of benefit and risk of medication.

Carolyn: Thank you, listeners, for joining us. You've been listening to Circulation on the Run. Join us next week.