Large-scale pharmacogenomic study of sulfonylureas and the QT, JT and QRS intervals: CHARGE Pharmacogenomics Working Group

JS Floyd1,47, CM Sittlani2,47, CL Avery3,47, R Noordam4,5, X Li6, AV Smith7,8, SM Gogarten9, J Li10, L Broer11, DS Evans12, S Trompet13, JA Brody2, JD Stewman3,14, JD Eicher15,16, AA Seyerle17, J Roach18, LA Lange19, HJ Lin20, JA Kors21, TB Harris22, R Li-Gao23, N Sattar24, SR Cummings12, KL Wiggins2, MD Napier3, T Stürmer3,25, JC Bis2, KF Kerr17, AG Uitterlinden11, K Taylor6, DJ Stott16, R de Mutsert13, LJ Launer22, EL Busch27,28, R Mendoza-Giraldez3, N Sotoodehnia1, EZ Soliman29, Y-DI Chen6, SR Heckbert34, RC Kaplan35, KM Rice9, JW Jukema36,37,38, AD Johnson15,16, Y Liu39, SR Cummings12, KL Wiggins2, MD Napier3, T Stürmer3,25, JC Bis2, KF Kerr17, AG Uitterlinden11, K Taylor6, DJ Stott16, R de Mutsert13, LJ Launer22, EL Busch27,28, R Mendoza-Giraldez3, N Sotoodehnia1, EZ Soliman29, Y-DI Chen6, SR Heckbert34, RC Kaplan35, KM Rice9, JW Jukema36,37,38, AD Johnson15,16, Y Liu39, DO Mook-Kanamori23,40, V Gudnason7,8, JL Wilson41, JI Rotter6, CC Laurie9, BM Psaty42,43, EA Whitsell44, LA Cupples16,45 and BH Stricker46

Sulfonylureas, a commonly used class of medication used to treat type 2 diabetes, have been associated with an increased risk of cardiovascular disease. Their effects on QT interval duration and related electrocardiographic phenotypes are potential mechanisms for this adverse effect. In 11 ethnically diverse cohorts that included 71857 European, African-American and Hispanic/Latino ancestry individuals with repeated measures of medication use and electrocardiogram (ECG) measurements, we conducted a pharmacogenomic genome-wide association study of sulfonylurea use and three ECG phenotypes: QT, JT and QRS intervals. In ancestry-specific meta-analyses, eight novel pharmacogenomic loci met the threshold for genome-wide significance (P < 5 × 10−8), and a pharmacokinetic variant in CYP2C9 (rs1057910) that has been associated with sulfonylurea-related treatment effects and other adverse drug reactions in previous studies was replicated. Additional research is needed to replicate the novel findings and to understand their biological basis.

The Pharmacogenomics Journal (2018) 18, 127–135; doi:10.1038/tpj.2016.90; published online 13 December 2016
INTRODUCTION
Sulfonylureas are the oldest class of oral glucose-lowering therapy used to treat type 2 diabetes, and despite the emergence of several new classes of diabetes drugs in recent years, sulfonylureas remain the most widely prescribed oral therapy after metformin. Since the University Group Diabetes Program trial found that the first-generation sulfonylurea chlorpropamide increased the risk of cardiovascular mortality over 40 years ago, there have been concerns about the cardiovascular safety of sulfonylureas. Several studies since then have found that treatment with sulfonylureas is associated with an increased risk of cardiovascular events and mortality compared with other glucose-lowering drugs.

As one potential mechanism of cardiovascular toxicity, sulfonylureas can prolong the QT interval, a marker of cardiac repolarization that is associated with fatal arrhythmias and sudden cardiac death. Indeed, QT prolongation has been one of the most common side issues leading to drug withdrawals from the market. Since 2005, the Food and Drug Administration has required clinical studies to evaluate whether a new drug prolongs the QT interval of 5 ms before regulatory approval. Variation in the QT interval isheritable, and large scale genome-wide association (GWA) studies have identified at least 35 genetic loci associated with this trait that collectively explain ~10% of interindividual variation in the QT interval. Pharmacogenomic studies of sulfonylurea use and the QT interval may help to unravel the biologic mechanisms underlying the cardiovascular toxicity of sulfonylureas. However, previous pharmacogenomic studies of the glucose-lowering or adverse effects of sulfonylureas have been small and focused on candidate genes, and most findings have not replicated. In the Cohorts for Heart and Aging Research in Genomic Epidemiology (CHARGE) Consortium Pharmacogenomics Working Group, a previous GWA study of sulfonylurea–QT interactions that included ~300,000 European ancestry individuals with cross-sectional measures of drug use and the QT interval did not identify any pharmacogenomic loci at genome-wide levels of significance.

To increase our power to identify novel pharmacogenomic loci for sulfonylureas, we extended this effort to include several additional diverse-ancestry cohorts with a high prevalence of sulfonylurea use. In addition, we incorporated repeated measures of drug exposure and phenotype with novel analytic methods. Because genetic variants can have different effects on the two components of the QT interval—the JT interval, which measures primarily repolarization, and the QRS interval, which measures primarily conduction and depolarization—we also extended our analyses to include them.

MATERIALS AND METHODS
Study population and overview
Eleven cohorts participated in this meta-analysis from the CHARGEC. The Pharmacogenomics Working Group: Age, Gene/Environment Susceptibility – Reykjavik Study (AGES); Atherosclerosis Risk in Communities (ARIC) Study; Cardiovascular Health Study (CHS); Health, Aging, and Body Composition (Health ABC); Hispanic Community Health Study/Study of Latinos (HCHS/SOL); Jackson Heart Study (JHS); Multi-Ethnic Study of Atherosclerosis (MESA); Netherlands Epidemiology of Obesity (NEO) Study; Prospective Study of Pravastatin in the Elderly at Risk (PROSPER); Rotterdam Study cohorts 1 and 2; and the Women’s Health Initiative (WHI) (Supplementary Information). Cohorts contributed results from European ancestry (EA), African-American (AA) and/or Hispanic/Latino ancestry (HA) populations. All cohorts had at least one study visit with an assessment of medication use and a resting 12-lead electrocardiogram (ECG); AGES, ARIC, CHS, the Rotterdam Study, MESA and WHI had multiple study visits with these assessments and contributed repeated measures. Each cohort followed a prespecified analysis protocol, and findings from within-cohort analyses were combined in three sets of ancestry-specific meta-analyses (EA, AA and HA) for three ECG phenotypes (QT, JT and QRS intervals), for a total of nine primary analyses. All available cohorts were included in this single discovery effort, rather than a two-stage design with discovery and replication, to improve our power to identify significant pharmacogenomic interactions. This study was approved by the institutional review board of each cohort.

Inclusion and exclusion criteria
Participants with genome-wide genotype data and with ECG measurements and medication assessments at the same study visits were eligible. The following exclusion criteria were applied: poor ECG quality; atrial fibrillation; second- or third-degree atrioventricular heart block; QRS interval > 120 ms; a paced rhythm; history of heart failure; pacemaker implantation; pregnancy; and ancestry other than European, African American or Hispanic/Latino. For studies with repeated measures, exclusion criteria were applied for each visit-specific observation.

Drug exposure assessment
Sulfonylurea drugs are listed in Supplementary Table 1. Sulfonylurea use was assessed through medication inventories conducted at study visits, or using information from a pharmacy database for the Rotterdam Study (Supplementary Table 2). Some cohorts assessed medication use on the day of the study visit, whereas others assessed medication use within an interval of time before the study visit, typically 2 weeks. For cohorts with repeated measures, the number of participants exposed to sulfonylureas (Nexposed) was the sum of the estimated number of independent observations at which each participant was exposed, calculated from the following equation:

\[ N_{\text{exposed}} = \sum \frac{n_i}{1 + (n_i - 1)\hat{\rho}} \]

where the summand is the product of the estimated number of independent observations and the proportion of observations at which a participant was exposed, \( n_i \) being the number of observations for participant \( i \), \( \hat{\rho} \) an estimate of the pairwise visit-to-visit correlation in outcome within participants from a generalized estimating equation-exchangeable model that does not contain genetic data and \( \#\{E_i = 1\} \) the number of observations for which participant \( i \) was exposed.

Phenotype measurement
QT and QRS intervals were recorded from resting, supine or semirecumbent standard 12-lead ECGs (Supplementary Table 2). Across all cohorts, comparable procedures were used for preparing participants, placing electrodes, recording, transmitting, processing and controlling the quality of ECGs. Cohorts used Marquette MAC 5000, MAC 1200 or MAC PC (GE Healthcare, Milwaukee, WI, USA), Burdick Eclipse 850 (Cardiac Science, Manchester, UK) or ACTA (EASOTE, Florence, Italy) machines. Recordings were processed using Marquette 12SL, MEANS or University of Glasgow software (Glasgow, Scotland, UK). The JT interval was calculated by the formula: \( JT = QT – QRS \).

Genotyping and imputation
All cohorts performed genome-wide genotyping with either Affymetrix (Santa Clara, CA, USA) or Illumina (San Diego, CA, USA) arrays, and used similar quality control thresholds for excluding samples and single-nucleotide polymorphisms (SNPs) (Supplementary Table 3). Sex mismatches, duplicate samples and first-degree relatives (except in HCHS/SOL and JHS) were excluded. DNA samples and SNPs with call rates of < 90–98%, depending on the cohort, were excluded. Within each cohort, SNPs with minor allele frequencies (MAFs) of < 1% or that failed Hardy–Weinberg equilibrium were excluded.

Genotypes were imputed using ancestry-specific HapMap2,3–34. HapMap3, 1000 Genomes Phase 1 or 1000 Genomes Phase 3 reference panels (Supplementary Table 3). Genotypes imputed from build 37 of the human genome were lifted over to build 36 (refs 37) to enable comparisons between imputation platforms, and all results were restricted to SNPs present in HapMap2.

Statistical analysis
GWA analyses were performed by each cohort separately, and ancestry-specific results for each ECG phenotype were combined with meta-analysis. Within each cohort, for ∼2.5 million genotyped or imputed
autosomal SNPs, sulfonylurea–SNP interactions were estimated with an additive genetic model using mixed effects models, generalized estimating equation or linear regression with robust standard errors. The analytic model varied based on the study design and the availability of longitudinal data (Supplementary Table 4). All analyses were adjusted for age, sex, study site or region, principal components of genetic ancestry, visit-specific RR interval (inversely related to heart rate) and visit-specific use of QT-prolonging medications. The QT-prolonging effect of medications was categorized as definite, possible or conditional, according to the University of Arizona Center for Education and Research on Therapeutics (UAZ CERT) system of classification, and adjusted for as binary variables for each category (presence of any versus none). HCHS/SOL incorporated estimates of relatedness into all analyses. Cohort-specific results were corrected for genomic inflation.

Previous simulations demonstrated that models using robust standard errors underestimate the variance of coefficient estimates for SNPs with low MAFs. To account for this, corrected standard errors were calculated using a t distribution as the reference distribution. Cohort and SNP-specific degrees of freedom (d.f.) for the t distribution were estimated primarily using the method of Satterthwaite. For cohorts unable to implement the method of Satterthwaite, an approximate d.f. was calculated as two times the cohort- and SNP-specific product of the SNP imputation quality (0–1), MAF (0.00–0.50) and Nexposed. Standard errors were then corrected by assuming a normal reference distribution that yielded the t distribution-based P-values from the coefficient estimates. Furthermore, because simulations demonstrated that corrected standard errors were unstable when minor allele counts among the exposed were low, an approximate d.f. filter of 10 was applied to cohort-specific results across all SNPs.

Table 1. Characteristics of study populations

| Cohort | N | Nexposed (%) | Age, years (s.d.) | Female, N (%) | QT interval, ms (s.d.) | JT interval, ms (s.d.) | QRS interval, ms (s.d.) |
|--------|---|--------------|-------------------|--------------|-----------------------|-----------------------|-----------------------|
| **European Ancestry** | | | | | | | |
| AGES | 2587 | 64 (2.5) | 75 (4.7) | 925 (64) | 406 (34) | 316 (33) | 90 (10) |
| ARIC | 8597 | 379 (4.4) | 54 (5.7) | 4453 (53) | 399 (29) | 308 (29) | 91 (10) |
| CHS | 3055 | 280 (9.2) | 72 (5.3) | 1880 (63) | 414 (32) | 321 (30) | 88 (10) |
| Health ABC | 1441 | 81 (5.6) | 74 (2.8) | 714 (49) | 414 (32) | 324 (32) | 90 (11) |
| MESA | 2256 | 71 (3.1) | 62 (10.1) | 1156 (52) | 412 (29) | 320 (29) | 93 (9) |
| NEO | 5366 | 94 (1.8) | 56 (5.9) | 2521 (47) | 406 (29) | 313 (29) | 93 (10) |
| PROSPER | 4555 | 243 (5.3) | 75 (3.3) | 2445 (47) | 414 (36) | 320 (35) | 94 (11) |
| Rotterdam 1 | 4805 | 216 (4.5) | 69 (8.6) | 2891 (60) | 397 (29) | 300 (28) | 97 (11) |
| Rotterdam 2 | 1889 | 84 (4.4) | 65 (7.6) | 1070 (57) | 403 (28) | 305 (28) | 98 (11) |
| WHI GARNET | 3943 | 304 (7.7) | 66 (6.8) | 3642 (100) | 400 (32) | 314 (31) | 86 (9) |
| WHI MOPMAP | 1324 | 36 (2.7) | 63 (6.6) | 1224 (100) | 402 (30) | 316 (30) | 86 (8) |
| WHIMS | 2194 | 243 (4.7) | 69 (6.0) | 4811 (100) | 401 (30) | 315 (30) | 86 (9) |
| Total | 45002 | 2095 (4.7) | | | | | |
| **African American** | | | | | | | |
| ARIC | 2191 | 213 (9.7) | 53 (5.8) | 1322 (62) | 400 (33) | 310 (32) | 90 (10) |
| CHS | 707 | 141 (20.0) | 73 (5.6) | 447 (65) | 409 (35) | 317 (36) | 88 (11) |
| Health ABC | 1020 | 111 (10.9) | 73 (2.9) | 588 (58) | 411 (35) | 322 (34) | 88 (11) |
| JHS | 2122 | 117 (5.5) | 50 (11.8) | 1244 (61) | 410 (30) | 319 (30) | 92 (11) |
| MESA | 1464 | 135 (9.2) | 62 (10.0) | 796 (54) | 410 (32) | 319 (31) | 91 (10) |
| WHI SHARE | 4227 | 450 (10.6) | 61 (6.8) | 3860 (100) | 401 (34) | 316 (33) | 85 (9) |
| Total | 11731 | 1167 (9.9) | | | | | |
| **Hispanic/Latino** | | | | | | | |
| HCHS/SOL | 12024 | 518 (4.3) | 46 (13.8) | 7155 (60) | 416 (28) | 325 (29) | 91 (10) |
| MESA | 1316 | 134 (10.2) | 61 (10.3) | 681 (52) | 409 (30) | 318 (30) | 91 (10) |
| WHI SHARE | 1784 | 142 (7.9) | 60 (6.4) | 1627 (100) | 402 (30) | 316 (30) | 86 (9) |
| Total | 15124 | 794 (5.2) | | | | | |
| Total, all ancestries | 71857 | 4056 (5.6) | | | | | |

Abbreviations: AGES, Age, Gene/Environment Susceptibility–Reykjavik Study; ARIC, Atherosclerosis Risk in Communities Study; CHS, Cardiovascular Health Study; Health ABC, Health, Aging, and Body Composition Study; HCHS/SOL, Hispanic Community Health Study/Study of Latinos; JHS, Jackson Heart Study; MESA, Multi-Ethnic Study of Atherosclerosis; NEO, Netherlands Epidemiology of Obesity; PROSPER, Prospective Study of Pravastatin in the Elderly at Risk; Rotterdam 1, first cohort of the Rotterdam Study; Rotterdam 2, second cohort of the Rotterdam study; WHI GARNET, Women’s Health Initiative Genome-wide Association Research Network into Effects of Treatment; WHI MOPMAP, Women’s Health Initiative Modification of Particulate Matter-Mediated Arrhythmogenesis in Populations; WHI SHARE, Women’s Health Initiative SNP Health Association Resource; WHIMS, Women’s Health Initiative Memory Study. Nexposed = ∑ i=1 n xi / n

Primary analyses

For each ECG phenotype and for each ancestral population, SNP-by-treatment interaction coefficients and corrected standard errors were combined with inverse-variance-weighted meta-analysis using METAL. SNPs had to meet quality control criteria and pass the d.f. filter in at least two studies to be included. The threshold for statistical significance was P < 5 × 10⁻⁶, and this has been used in other GWAS studies of correlated phenotypes. Each locus with multiple SNPs meeting the threshold for statistical significance, a lead SNP with the lowest P-value was identified. Significant loci and loci at suggestive levels of statistical significance (P < 10⁻⁴) were annotated using information from several genomics and bioinformatics databases. RefSeq genes within 500 kb of lead SNPs were identified from the UCSC (University of California, Santa Cruz) Genome Browser. The NHGRI-EBI GWAS Catalog was queried for other traits associated with lead SNPs in GWA studies. HaploReg (Broad Institute, Cambridge, MA, USA) was queried to identify missense coding variants in linkage disequilibrium (LD) (R² < 0.8) with lead SNPs. Cis-expression quantitative trait loci (cis-eQTLs) in LD with lead SNPs were identified from several gene expression databases, including ScanDB and the Broad Institute GTEx Portal, that include samples from multiple cell lines and tissue sites, including whole blood, leukocytes, subcutaneous adipose, skeletal muscle, lung, skin, fibroblasts, arterial wall and left ventricular and atrial heart tissue.

Secondary analyses

All ancestry-specific summary results were combined in a transethnic inverse-variance weighted meta-analysis using METAL. Because effects

© 2018 Macmillan Publishers Limited, part of Springer Nature. The Pharmacogenomics Journal (2018), 127–135
may be heterogeneous across different racial/ethnic populations,\textsuperscript{47,48} we conducted additional transethnic analyses using the Bayesian MANTRA method, with a genome-wide significance threshold of $\log_{10} \text{Bayes factor} \geq 6.49$

Previous candidate gene pharmacogenetic studies have identified several pharmacokinetic and pharmacodynamic loci for sulfonylurea-associated glucose-lowering effects and hypoglycemia.\textsuperscript{19–23,50–53} In addition, large-scale GWA studies have identified 35 replicated genetic loci for QT interval main effects.\textsuperscript{18} For these candidate SNPs, the $P$-value threshold for statistical significance was 0.05 divided by the total number of tests conducted across all ECG phenotypes and populations: $0.05/158 = 3.2 \times 10^{-4}$.

For the QT interval, we also assessed for enrichment of candidate SNP-by-treatment interactions with a high probability of being functional for cardiac conduction and repolarization phenotypes. SNPs that fell within 50 kb of transcripts that are preferentially expressed in the left ventricle were identified using the GTEx database (839 transcripts). SNPs in these gene regions were filtered to those falling within DNAse I hypersensitivity, H3K4me3 or CTCF chromatin immunoprecipitation sequencing peaks assayed in human cardiomyocytes from the NIH (National Institutes of Health) Roadmap Epigenomics Consortium (http://www.roadmapepigenomics.org). In addition, SNPs that were eQTLs in left ventricle tissue ($P < 1 \times 10^{-10}$) were selected.\textsuperscript{54,55} All variants were pruned using ancestry-matched LD patterns from the 1000 Genomes project at a level of $R^2 > 0.5$,\textsuperscript{56} resulting in 9004, 8424 and 5437 candidate SNPs for EA, AA and HA analyses, respectively.

The $P$-value threshold for statistical significance for these candidate SNP analyses was 0.05 divided by the total number of SNPs selected ($P < 5.6 \times 10^{-6}$ for EA, $P < 5.9 \times 10^{-6}$ for AA and $P < 5.6 \times 10^{-6}$ for HA). The selection of candidate SNPs was validated by evaluating enrichment for low $P$-value variants using main-effect SNP associations from the QT Interval-International GWAS Consortium.\textsuperscript{18}

RESULTS

The characteristics of the 11 cohorts and 21 ancestry-specific analysis populations are listed in Table 1. There were 45 002 EA participants ($N_{\text{exposed}}$ 2095 (4.7%)), 11 731 AA participants ($N_{\text{exposed}}$ 1167 (9.9%)) and 15 124 HA participants ($N_{\text{exposed}}$ 794 (5.2%)), for a total of 71 857 ($N_{\text{exposed}}$ 4056 (5.6%)). Mean durations of ECG intervals ranged from 397 to 414 ms for QT, 300 to 325 ms for JT and 85 to 98 ms for QRS. The correlation between traits was evaluated among EA and AA participants of CHS: QRS and JT were highly correlated ($R^2 \geq 0.5$), whereas QRS was not correlated with either QRS or JT ($R^2 < 0.1$).

Primary analysis results

Sulfonylurea–SNP interaction results from cohort-specific GWA analyses were well calibrated: genomic inflation factors for ancestry-specific meta-analyzed results ranged from 0.97 to 1.04 (Supplementary Table 5). A total of 31 sulfonylurea–SNP interaction associations met the genome-wide threshold for significance, comprising 8 unique loci (Figure 1 and Table 2). Each locus was significant for only one of the three ECG phenotypes (2 QT, 5 JT and 1 QRS) and in only one racial/ethnic population (3 EA and 5 AA). Absolute values for effect sizes ranged from 4 to 16 ms. All
Some of the novel pharmacogenomic loci discovered in our study were near (but not in LD with) loci for related traits, such as lead SNP (rs1799853). For example, a missense variant, 3 were in LD (< 0.8) with lead SNP. The eQTLs indicate transcripts associated with SNPs in linkage disequilibrium (< 0.8) with lead SNP.

**DISCUSSION**

In this study, we identified eight novel loci for sulfonylurea–genetic interactions with the QT, JT and QRS intervals. For seven of these pharmacogenomic associations, the effect size was > 5 ms, the threshold for regulatory concern established by the FDA (Food and Drug Administration). Compared with our previous effort, which included 869 sulfonylurea users among ~30,000 EA participants and failed to identify any genome-wide significant loci, this effort included over 4000 sulfonylurea users among over 70,000 participants from diverse ancestries. Broadening the racial/ethnic composition of the study population and extending our investigation to related ECG phenotypes improved our ability to identify pharmacogenomic loci; most were identified in AA populations and for the JT interval.

Some of the novel pharmacogenomic loci discovered in our study were near (but not in LD with) loci for related traits, such as the NFIA locus on chromosome 1 was ~ 200 kb away from a locus associated with QT interval main effects; NFIA encodes a transcription factor of cardiac tissue development. A locus on chromosome 2 (rs12468579) was 2 kb away from GLS and was also identified as a cis-eQTL for GLS and MFS6 transcripts in blood, lung and prostate. GLS encodes glutaminase that catalyzes the production of glutamine, the most abundant excitatory neurotransmitter in the central nervous system. The chromosome 3 locus (rs1478173) was ~ 115 kb away from a locus for coronary artery disease. The only locus associated with another trait (periodontitis) in other GWASs. Coding indicates lead SNP in linkage disequilibrium (< 0.8).

Secondary analysis results:

Transethnic fixed effects meta-analyses and MANTRA analyses did not identify any additional loci (results not shown). Among the candidate SNPs, only one was significantly associated with an ECG phenotype when multiple comparisons were accounted for (Table 3). This SNP, rs1057910 (Ile359Leu), is a loss-of-function variant that defines the *2 haplotype of CYP2C9, a highly polymorphic cytochrome P450 (CYP) enzyme that metabolizes 15–20% of all known drugs that undergo phase I oxidative metabolism. For the sulfonylurea–SNP interaction, the minor allele of rs1057910 was associated with a 7.6 ms (s.e. 2.1 ms) decrease in the QT interval (P = 2.3 × 10⁻⁴) in HA cohorts (MAF 0.05), but not in EA cohorts (MAF 0.07). This SNP did not meet filtering criteria for meta-analysis in the AA cohorts. The more common functional variant (rs1799853) that defines the *2 haplotype of CYP2C9 (MAF 0.13 in EA and 0.09 in HA) was also evaluated, but it was not significantly associated with any of the ECG phenotypes.

Selecting additional candidate SNPs based on bioinformatic analysis of annotation from cardiac gene expression and regulatory marks active in cardiomyocytes did not identify additional loci. Although these variants were enriched for signals among main-effect QT analyses (Supplementary Figure 1), none met our statistical significance threshold for sulfonylurea–SNP interactions with the QT, JT or QRS intervals (Supplementary Figure 2).
these SNPs, only a well-known functional variant in CYP2C9 was identified as a pharmacogenomic locus in our study, and among HA participants only. Variant rs1057910 (CYP2C9*3) reduces the catalytic activity of CYP2C9, the main CYP isoenzyme involved in the metabolism of sulfonylureas, and this variant has been associated with severe skin reactions from phenytoin use and warfarin-related hemorrhage. Allele frequencies for rs1057910 were similar among HA and EA participants in our study that has associated with severe skin reactions from phenytoin use and warfarin therapy, functional variants in CYP2C9 and VKORC1 were associated with lower warfarin dose requirements and a higher risk of warfarin adverse effects. Other studies, conducted primarily in EA populations, have evaluated the impact of CYP2C9 functional variants on sulfonylurea-related treatment response and adverse effects. In one study, the presence of either the CYP2C9*2 or the CYP2C9*3 haplotype was associated with an increased reduction in hemoglobin A1c and an increased probability of achieving adequate glycemic control, and in another study these variants were associated with an increased risk of hypoglycemia among elderly individuals. In our study, the variant rs1057910 was associated with a shorter QT interval among HA participants. This was a surprising

The Pharmacogenomics Journal (2018), 127 – 135 © 2018 Macmillan Publishers Limited, part of Springer Nature.

### Table 3. Results for pharmacokinetic, pharmacodynamic and QT main-effect candidate SNPs

| SNP              | Chr | Gene     | EA   | AA   | HA   | P-values      |
|------------------|-----|----------|------|------|------|---------------|
| Pharmacokinetic  |     |          |      |      |      |               |
| rs1057910        | 19  | CYP2C9   | 0.42 | 0.06 | 0.55 | 2.3E – 4      |
| rs1799853        | 10  | CYP2C9   | 0.99 | 0.88 | 0.62 | 0.37          |
| Pharmacodynamic  |     |          |      |      |      |               |
| rs1049435        | 5   | NOS1AP   | 0.27 | 0.20 | 0.78 | 0.58          |
| rs7903146        | 51  | TCFL2    | 0.30 | 0.34 | 0.84 | 0.64          |
| rs1225537        | 51  | TCFL2    | 0.39 | 0.16 | 0.50 | 0.35          |
| rs5215          | 3.53 | KCNJ11   | 0.93 | 0.04 | 0.44 | 0.49          |
| rs757110        | 41  | ABCB8    | 1.00 | 0.08 | 0.24 | 0.42          |
| QT main effect   |     |          |      |      |      |               |
| rs2298632       | 1   | TCEA3    | 0.20 | 0.52 | 0.78 | 0.58          |
| rs846111        | 1   | RNK207   | 1.00 | 0.06 | 0.44 | 0.37          |
| rs19091970      | 1   | ATP1B1   | 0.91 | 0.89 | 0.82 | 0.86          |
| rs12143842      | 1   | NOS1AP   | 0.44 | 0.67 | 0.52 | 0.67          |
| rs295140        | 2   | SPAT5L2  | 0.12 | 0.62 | 0.29 | 0.67          |
| rs938291        | 2   | SP3      | 0.79 | 0.41 | 0.83 | 0.75          |
| rs7561149       | 2   | TTN-CCDC141 | 0.85 | 0.84 | 0.44 | 0.43          |
| rs12997023      | 2   | SLC8A1   | 0.29 | 0.62 | 0.65 | 0.77          |
| rs6793245       | 3   | SCN5A-SCN10A | 0.95 | 0.15 | 0.74 | 0.79          |
| rs17784882      | 3   | C3ORF75  | 0.15 | 0.35 | 0.51 | 0.40          |
| rs3857067       | 4   | SMARCAD1 | 0.82 | 0.40 | 0.19 | 0.35          |
| rs2363719       | 4   | SLC4A4   | 0.23 | 0.89 | 0.93 | 0.30          |
| rs10049089      | 5   | GPR3     | 0.43 | 0.12 | 0.35 | 0.30          |
| rs7765828       | 6   | GMPR     | 0.63 | 0.23 | 0.19 | 0.39          |
| rs11153730      | 6   | SLC35F1-PLN | 0.84 | 0.24 | 0.40 | 0.40          |
| rs9920          | 7   | CAV1     | 0.36 | 0.64 | 0.85 | 0.85          |
| rs2072413       | 7   | KCNH2    | 0.30 | 0.77 | 0.44 | 0.77          |
| rs1961102       | 8   | AZIN1    | 0.33 | 0.39 | 0.44 | 0.39          |
| rs11779860      | 8   | LAPTMB   | 0.74 | 0.38 | 0.95 | 0.30          |
| rs16936870      | 8   | NCOA2    | 0.08 | 0.93 | 0.32 | 0.09          |
| rs174583       | 10  | FEN1-FADS2 | 0.87 | 0.89 | 0.74 | 0.74          |
| rs2485376       | 10  | GF1B     | 0.40 | 0.94 | 0.73 | 0.94          |
| rs7129397       | 11  | KCNQ1    | 0.25 | 0.81 | 0.39 | 0.39          |
| rs3026445       | 12  | ATP2A2   | 0.49 | 0.62 | 0.24 | 0.35          |
| rs728926        | 13  | KLF12    | 0.29 | 0.60 | 0.29 | 0.19          |
| rs2273905       | 14  | ANKRQ9D  | 0.38 | 0.71 | 0.24 | 0.35          |
| rs3105593       | 15  | USP50-IPR7M | 0.71 | 0.73 | 0.91 | 0.41          |
| rs735951       | 16  | LITAF     | 0.34 | 0.93 | 0.35 | 0.93          |
| rs1052536       | 17  | LIG3     | 0.58 | 0.67 | 0.39 | 0.72          |
| rs246185       | 16  | MRK2     | 0.11 | 0.39 | 0.39 | 0.72          |
| rs246196       | 16  | CNOT1    | 0.30 | 0.94 | 0.39 | 0.39          |
| rs1296720       | 16  | CREBBP   | 0.33 | 0.72 | 0.24 | 0.35          |
| rs1396515       | 17  | KCNJ2    | 0.76 | 0.64 | 0.94 | 0.49          |
| rs9892651      | 17  | PRKCA    | 0.49 | 0.40 | 0.95 | 0.40          |
| rs1805128       | 21  | KCNE1    | 0.69 | 0.92 | 0.64 | 0.94          |

Abbreviations: AA, African American; Chr, chromosome; EA, European ancestry; HA, Hispanic/Latino ancestry; SNP, single-nucleotide polymorphism. With Bonferroni correction for 158 tests, the threshold for statistical significance was 3.1 × 10⁻⁷. Significant associations are in bold.
Pharmacogenomics of sulfonylureas and ECG phenotypes
JS Floyd et al

Committee, VSN: 00-063. The researchers are indebted to the participants for their willingness to participate in the study.

Atherosclerosis Risk in Communities (ARIC): The Atherosclerosis Risk in Communities Study is carried out as a collaborative study supported by National Heart, Lung and Blood Institute Contracts (HHSN26820100005C, HHSN26820100006C, HHSN268-201100007C, HHSN26820100008C, HHSN26820100009C, HHSN268201100010C, HHSN268201100011C and HHSN268201100012C), R01HL087641, R01HL103636 and R01HL108694; National Human Genome Research Institute Contract U01HG004402; and National Institutes of Health Contract HHSN268200625226C. We thank the staff and participants of the ARIC study for their important contributions. Infrastructure was partly supported by Grant No. UL1RR025005, a component of the National Institutes of Health and NIH Roadmap for Medical Research.

Cardiovascular Health Study (CHS): This CHS research was supported by NHLBI contracts (HHSN268201200036C, HHSN268201200037C, HHSN268201200038C, HHSN268201200039C, HHSN268201200040C, HHSN268201200041C, HHSN268201200042C, HHSN268201200043C, HHSN268201200044C, HHSN268200100011C and HHSN268200100012C), R01HL087641, R01HL103636 and R01HL108694 with additional contribution from the National Institute of Neurological Disorders and Stroke (NINDS). Additional support was provided through R01AG023629 from the National Institute on Aging (NIA). A full list of principal CHS investigators and institutions can be found at CHS-NHLBI.org. The provision of genotyping data was supported in part by the National Center for Advancing Translational Sciences, CTSA Grant UL1TR000124 and the National Institute of Diabetes and Digestive and Kidney Disease Diabetes Research Center (DRC) Grant DK063491 to the Southern California Diabetes Endocrinology Research Center. NS was supported by R01HL116747 and R01HL111089. JFS was supported by K08HL116640.

Health, Aging, and Body Composition (Health ABC): This research was supported by NIA Contracts N01AG26210, N01AG26213 and N01AG62106. The genome-wide association study was funded by NIA Grant 1R01AG032098-01A1 to Wake Forest University Health Sciences and genotyping services were provided by the Center for Inherited Disease Research (CIDR). CIDR is fully funded through a federal contract from the National Institutes of Health to The Johns Hopkins University, Contract No. HHSN26820072896C. This research was supported in part by the Intramural Research Program of the NIH, National Institute on Aging.

Hispanic Community Health Study/Study of Latinos (HCHS/SOL): We thank the participants and staff of the HCHS/SOL study for their contributions to this study. The baseline examination of HCHS/SOL was carried out as a collaborative study supported by contracts from the National Heart, Lung, and Blood Institute (NHLBI) to the University of North Carolina (N01-HC-65234), Albert Einstein College of Medicine (N01-HC-65235), Northwestern University (N01-HC-65236) and San Diego State University (N01-HC-65237). The following Institutes/Centers/Offices contributed to the first phase of HCHS/SOL through a transfer of funds to the NHLBI: National Institute on Minority Health and Health Disparities, National Institute on Deafness and Other Communication Disorders, National Institute of Dental and Craniofacial Research (NIDCR), National Institute of Diabetes and Digestive and Kidney Diseases, National Institute of Neurological Disorders and Stroke and NIH Institution-Office of Dietary Supplements. The Genetic Analysis Center at University of Washington was supported by NHLBI and NIDCR contracts (HSN268201300005C AM03 and MOD03). Genotyping efforts were supported by NHLBI HS0 2620020054C, NCATS CTSA Grant UL1TR000124, and NIDDK Diabetes Research Center (DRC) Grant DK063491.

Jackson Heart Study (JHS): We thank the Jackson Heart Study (JHS) participants and staff for their contributions to this work. The JHS is supported by contracts HHSN268201300046C, HHSN268201300047C, HSN268201300048C, HHSN268201300049C and HHSN268201300050C from the National Heart, Lung, and Blood Institute and the National Institute on Minority Health and Health Disparities.

Multi-Ethnic Study of Atherosclerosis (MESA): MESA and MESA SNP Health Association Resource (SHARe) are conducted and supported by the National Heart, Lung and Blood Institute (NHLBI) in collaboration with MESA investigators. Support is provided by grants and contracts N01-HC-59159, N01-HC-59160, N01-HC-59161, N01-HC-59162, N01-HC-59163, N01-HC-59164, N01-HC-59165, N01-HC-59166, N01-HC-59167, N01-HC-59168, N01-HC-59169 and RR-024156. Additional funding was supported in part by the Clinical Translational Science Institute Grant UL1RR033176 and is now at the National Center for Advancing Translational Sciences (NCATS) Grant UL1TR000124. We also thank the other investigators and the staff and the participants of the MESA study for their valuable contributions. A full list of participating MESA investigators and institutions can be found at http://www.mesa.nlm.nih.gov.

Netherlands Epidemiology of Obesity (NEO): The authors of the NEO study thank all individuals who participated in the Netherlands Epidemiology in Obesity study, all participating general practitioners for inviting eligible participants and all researchers who contributed to the data collected. We thank the NEO steering committee, Petra Noordijk and Ingeborg de Jonge for the coordination, lab and data management of the NEO study. The genotyping in the NEO study was supported by the Centre National de Génotypage (Paris, France), headed by Jean-François
Deleuze. The NED study is supported by the participating departments, the Division and the Board of Directors of the Leiden University Medical Center and by the Leiden University, Research Profile Area Vascular and Regenerative Medicine. Dennis Mook-Kanamori is supported by Dutch Science Organization (ZonMW-VENI Grant 916,14,023).

Prospective Study of Pravastatin in the Elderly at Risk (PROSPER): The PROSPER study was supported by an investigator-initiated grant obtained from Bristol-Myers Squibb. Professor Dr. J.W. Jukema is an Established Clinical Investigator of the Netherlands Heart Foundation (Grant No. 2001 D 032). Support for genotyping was provided by the seventh framework program of the European commission (Grant No. 223004) and by the Netherlands Genomics Initiative (Netherlands Consortium for Healthy Aging Grant 050-060-810).

Rotterdam Study (RS): The RS is supported by the Erasmus Medical Center and Erasmus University Rotterdam; The Netherlands Organization for Scientific Research; The Netherlands Organization for Health Research and Development (ZonMw); The Research Institute for Diseases in the Elderly; The Netherlands Heart Foundation; the Ministry of Education, Culture and Science; the Ministry of Health Welfare and Sports; the European Commission; and the Municipality of Rotterdam. Support for genotyping was provided by The Netherlands Organization for Scientific Research (NWO) (175.010.2005.011, 911.03.012) and Research Institute for Diseases in the Elderly (RIDE). This study was supported by The Netherlands Genomics Initiative (NGI)/D流出kelder for Scientific Research (NWO) Project No. 050-060-810. This collaborative effort was supported by an award from the National Heart, Lung and Blood Institute (RO1-HL 103612, PI: BMP).

Women’s Health Initiative Clinical Trial (WHI CT): The Women’s Health Initiative clinical trials were funded by the National Heart, Lung, and Blood Institute, National Institutes of Health, US Department of Health and Human Services through Contracts HHSN268201100046C, HHSN268201100023C, HHSN268201100004C and HHSN268201100005C. All contributors to WHI science are listed at https://www.whi.org/researchers/Documents%20Write%20a%20Paper/WHI%20Investigator%20Long%20List.pdf. ELB was supported in part by a grant from the National Cancer Institute (5T32CA009001). A Paper/WHI Investigator Long List.pdf. ELB was supported in part by a grant from the National Cancer Institute (5T32CA009001). All contributors to GARNET science are listed at https://www.genome.gov/27541119/ and Human Services through cooperative agreement U01HG005152 (Reiner). All CVD and Metabolic Outcomes in the WHI was funded by the National Human Genetics Research Institute; The Women’s Health Initiative Clinical Trial (WHI CT). This collaborative effort was supported by an award from the National Heart, Lung and Blood Institute (R01-HL-103612, PI BMP).

Pharmacogenomics of sulfonylureas and ECG phenotypes

White HA, Moore BB, Bajpai SK, Ruskin JN. Use of antidiabetic drugs and QT interval prolongation in type 2 diabetes mellitus treated with gliclazide. Diabetes Metab 1994; 20: 565–567.

Najeeb SA, Khan IA, Molnar J, Somberg JC. Differential effect of glyburide (glibenclamide) and metformin on QT dispersion: a potential adenosine triphosphate-sensitive K+ channel effect. Am J Cardiol 2002; 90: 1103–1106.

Schwartz PJ, Wolf S. QT interval prolongation as predictor of sudden death in patients with myocardial infarction. Circulation 1978; 57: 1074–1077.

Zhang Y, Post WS, Blasco-Colmenares E, Dalal D, Tomasselli GF, Guallar E. Electrocardiographic QT interval and mortality: a meta-analysis. Epidemiology 2011; 22: 660–670.
