Sex Differences in the Risk of Coronary Heart Disease Associated With Type 2 Diabetes: A Mendelian Randomization Analysis

Diabetes Care 2021;44:556–562 | https://doi.org/10.2337/dc20-1137

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OBJECTIVE

Observational studies have demonstrated that type 2 diabetes is a stronger risk factor for coronary heart disease (CHD) in women compared with men. However, it is not clear whether this reflects a sex differential in the causal effect of diabetes on CHD risk or results from sex-specific residual confounding.

RESEARCH DESIGN AND METHODS

Using 270 single nucleotide polymorphisms (SNPs) for type 2 diabetes identified in a type 2 diabetes genome-wide association study, we performed a sex-stratified Mendelian randomization (MR) study of type 2 diabetes and CHD using individual participant data in UK Biobank (251,420 women and 212,049 men). Weighted median, MR-Egger, MR-pleiotropy residual sum and outlier, and radial MR from summary-level analyses were used for pleiotropy assessment.

RESULTS

MR analyses showed that genetic risk of type 2 diabetes increased the odds of CHD for women (odds ratio 1.13 [95% CI 1.08–1.18] per 1-log unit increase in odds of type 2 diabetes) and men (1.21 [1.17–1.26] per 1-log unit increase in odds of type 2 diabetes). Sensitivity analyses showed some evidence of directional pleiotropy; however, results were similar after correction for outlier SNPs.

CONCLUSIONS

This MR analysis supports a causal effect of genetic liability to type 2 diabetes on risk of CHD that is not stronger for women than men. Assuming a lack of bias, these findings suggest that the prevention and management of type 2 diabetes for CHD risk reduction is of equal priority in both sexes.

Type 2 diabetes is a major risk factor for coronary heart disease (CHD) (1). A meta-analysis of observational studies demonstrated that type 2 diabetes is associated with a 44% greater relative risk of CHD in women compared with men (2). However, whether this reflects sex differences in the causal effect of type 2 diabetes on CHD or arises from confounding in observational studies is not well understood. Most observational studies adjust for traditional cardiovascular risk factors, yet novel biomarkers, social and behavioral factors, or women-specific risk factors, such as gestational diabetes mellitus, are not generally adjusted for and may explain some of

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the sex difference (3–5). Sex differences in screening for and treatment of type 2 diabetes might also contribute to the greater excess risk of CHD conferred by type 2 diabetes among women relative to men (6).

Mendelian randomization (MR) analysis exploits the natural random allocation of genetic variants at conception and is an increasingly used approach that can limit potential confounding in human research (7). Under the assumption that differences in the risk of disease arising from genotype mimic changes in the risk of disease acquired during life, MR can be used to detect causal effects. MR studies have supported a causal relationship between genetic predisposition to type 2 diabetes and CHD (8,9). However, these studies did not evaluate sex differences in the causal role of type 2 diabetes in CHD risk. If type 2 diabetes has a stronger causal effect on CHD risk in women compared with men, randomly allocated genetic variants that are risk alleles for type 2 diabetes should also be more strongly associated with the risk of CHD in women than in men. Therefore, in this study, we conducted an MR analysis to examine the sex-specific causal effect of the genetic risk of type 2 diabetes on CHD.

RESEARCH DESIGN AND METHODS

Data Sources and Study Participants

Data from the UK Biobank and a consortium of genome-wide association studies (GWAS) for type 2 diabetes were used. The UK Biobank is a large prospective study of >500,000 individuals (10). Baseline data collection in the UK Biobank was conducted between 2006 and 2010 across 22 assessment centers. Participants aged 37–73 years completed touchscreen questionnaires, were interviewed by trained research nurses, and had physical measurements taken and blood samples extracted and frozen. The presence of type 2 diabetes and CHD was self-reported at study baseline and confirmed by a trained nurse. Genotyping was performed using the Affymetrix UK BiLEVE Axiom array or the Affymetrix UK Biobank Axiom array. A combined reference panel including UK10K samples was used for imputation (11). In accordance with the National Research Ethics Service and the governing research ethics committee of UK Biobank, generic Research Tissue Bank approval was obtained, and study participants provided written informed consent (10).

For the current study, we included individual participant data on 463,469 UK Biobank participants who had concordant genetic and self-reported sex, who clustered with the Great Britain population in 1000 Genomes (12), whose genetic data were of sufficient quality (13), and who provided data on type 2 diabetes and CHD at baseline. Individuals with self-reported type 1 diabetes, gestational diabetes mellitus only, or a diabetes diagnosis before the age of 18 years were excluded. CHD was defined as a self-reported history of angina or myocardial infarction, and linkage with hospital admissions data and the national death register was used to also identify incident diagnoses of CHD after the baseline visit through ICD-9 or ICD-10 codes (410–414 and I20–I25, respectively) using follow-up data from recruitment through the end of February 2016 (mean 5.3 [SD 2.4] years), with 3,453 incident cases of CHD for women and 7,420 incident cases for men. Myocardial infarction was also defined using the UK Biobank algorithm (https://biobank.ctsu.ox.ac.uk/crystal/crystal/docs/alg_outcome_mi.pdf).

Sex-specific summary-level data (β-coefficients and SEs) for the genetic contribution of type 2 diabetes risk were obtained from the European Diabetes Meta-Analysis of Trans-Ethnic Association Studies (DIAMANTE) GWAS of individuals with type 2 diabetes (30,053 women and 41,846 men) and control participants (434,336 women and 383,767 men) of European descent (14). The UK Biobank was excluded from GWAS estimates used in our analyses to avoid sample overlap.

MR and Selection of Single Nucleotide Polymorphisms for Analyses

MR studies exploit the random assortment and independent inheritance of genetic variants in the population, which removes bias that is due to reverse causation and, if conducted appropriately, greatly reduces bias from residual or unmeasured confounding (15). However, three key assumptions must be met for genetic variants to serve as instrumental variables of an exposure in MR analyses (16) (Supplementary Fig. 1). First, the variants must be associated with the exposure of interest; second, they must not be associated with confounders of the relationship between the exposure and the outcome; and third, they must be independent of the outcome except for their association through the exposure. This third assumption relates to the issue of horizontal pleiotropy in which one or more variants used in the instrumental variable influences the outcome through a pathway other than the exposure of interest. When horizontal pleiotropy has a net effect to bias the properties of the genetic instrument, the summary MR estimate can be biased either toward or away from the null. In this situation, horizontal pleiotropy leads

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Received 14 May 2020 and accepted 21 October 2020
This article contains supplementary material online at https://doi.org/10.2337/figshare.13135781.

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to bias of the underlying "true" causal effect, and it is termed unbalanced horizontal, or directional, pleiotropy.

In this study, we used data from the UK Biobank for individual participant MR analysis. Single nucleotide polymorphisms (SNPs) with significant associations ($P < 5 \times 10^{-8}$) with type 2 diabetes from the sex-combined European DIAMANTE GWAS were selected (Supplementary Table 1). We assessed linkage disequilibrium ($r^2 > 0.2$) using PLINK (17) on a reference panel consisting of a random selection of 50,000 individuals from UK Biobank. Of 291 genome-wide significant SNPs from the European DIAMANTE GWAS, 270 were found in UK Biobank that were biallelic, were not in linkage disequilibrium, and were not derived from GWAS that adjusted for BMI. The SNPs were aligned to the same effect allele, and effect allele frequencies were checked for concordance. These 270 SNPs were used to generate sex-specific weighted genetic risk scores as the instrumental variable for analyses (18). Individual SNPs were coded as 0, 1, or 2, depending on the number of type 2 diabetes risk alleles. Each SNP was weighted by the corresponding sex-specific β-coefficient obtained from the European DIAMANTE GWAS and then summed for all SNPs. This method reduces the risk of false-positive results and bias toward the confounded observational association that may occur when all data (SNPs, exposure, outcome) are obtained from a single sample (19).

Statistical Analysis

The strength of the genetic risk score as an instrument for type 2 diabetes was assessed using the F-statistic, where an F-statistic >10 provides evidence against the possibility of bias arising because of a weak instrument (20). The association of sex-specific genetic risk scores with potential confounders was evaluated to assess the validity of the second assumption of MR (i.e., the genetic instrument is not associated with potential confounders) and was also compared with the observational association of type 2 diabetes status with potential confounders.

Two-stage residual inclusion (TSRI) estimation using logistic regression at the second stage (21) and Terza SEs (22) were used to evaluate the association of the genetic risk scores for type 2 diabetes with CHD to estimate the odds of CHD per 1-log unit increase in the odds of type 2 diabetes. This method includes first-stage residuals to correct for endogeneity (21) since application of traditional instrumental variable estimation approaches can be problematic for models that include a binary exposure and a binary outcome (23). Models were adjusted for age, genotype array, and the first four principal components of ancestry.

To assess and account for potential directional horizontal pleiotropy, we also performed summary-level MR analyses using SNP to type 2 diabetes estimates from DIAMANTE and SNP to CHD estimates in UK Biobank. For summary-level analyses, we obtained odds ratios (ORs) and 95% CIs for the causal effect of a 1-log unit increase in the odds of genetic liability to type 2 diabetes on the odds of CHD using the weighted median, MR-Egger, MR-pleiotropy residual sum and outlier (PRESSO), and radial MR methods (24–27). The weighted median method calculates a median of the SNP-specific causal estimates from the ratio method for each SNP (25). It has been shown to yield consistent estimates when the weights of up to half the instruments are not valid. The MR-Egger method is equivalent to an inverse variance–weighted method but does not constrain the intercept to zero, and as such, the MR-Egger estimate is the slope of the modified linear regression equation, and the intercept represents the average pleiotropic effect across SNPs (24). A nonzero intercept provides evidence of unbalanced horizontal pleiotropy, and the slope of the regression coefficient should provide an estimate that is free from bias induced by unbalanced horizontal pleiotropy. Analyses were conducted using the MendelianRandomization package in R Studio version 1.2.1206. The MR-PRESSO test detects and corrects for horizontal pleiotropy and was performed using the MRPRESSO package in R (26). The first part of the test (MR-PRESSO global test) identifies the presence of horizontal pleiotropy, the second part corrects the causal estimate for identified pleiotropy through outlier removal, and the third part (MR-PRESSO distortion test) tests whether the causal estimate significantly differs before and after correction. Additional analyses for pleiotropy assessment used radial MR-Egger models to identify outliers in the UK Biobank analysis using the WSpiller/RadialMR package in R with modified second-order weights (27), and analyses were repeated after exclusion of sex-specific outliers. *P* values for the test of interaction for estimates from separate analyses were used to assess interaction by sex for each analysis (28).

RESULTS

Characteristics of the UK Biobank participants are presented in Table 1 and Supplementary Table 3. The mean age was 57 (SD 8) years, and 46% of participants

Table 1—Population characteristics, UK Biobank (*N* = 463,469)

	Women (*n* = 251,420)	Men (*n* = 212,049)
Age (years), mean (SD)	56.6 (7.95)	57.0 (8.12)
Array type, *n* (%)		
BiLEVE	24,920 (9.9)	24,897 (11.7)
Axiom	226,489 (90.1)	187,147 (88.3)
Type 2 diabetes, *n* (%)	9,964 (4.0)	16,917 (8.0)
BMI (kg/m^2), mean (SD)	27.0 (5.1)	27.9 (4.2)
Waist circumference (cm), mean (SD)	84.6 (12.5)	97.1 (11.4)
Smoking history, *n* (%)		
Never	146,521 (58.3)	102,139 (48.2)
Previous	81,252 (32.3)	82,970 (39.1)
Current	22,574 (9.0)	26,011 (12.3)
Dyslipidemia, *n* (%)	25,549 (10.2)	33,843 (16.0)
Hypertension, *n* (%)	57,721 (23.0)	64,668 (30.5)
Systolic BP (mmHg), mean (SD)	135.3 (19.1)	141.1 (17.4)
Diastolic BP (mmHg), mean (SD)	80.5 (9.9)	84.0 (9.9)
CHD, *n* (%)	12,716 (5.1)	26,344 (12.4)
Myocardial infarction, *n* (%)	3,807 (1.5)	12,871 (6.0)
Angina, *n* (%)	4,864 (1.9)	10,219 (4.8)

BP, blood pressure.

were men. The prevalence of type 2 diabetes was 4% in women and 8% in men. CHD was documented among 5% of women (n = 12,716) and 12% of men (n = 26,344), with myocardial infarction diagnosed in 1.5% of women (n = 3,807) and 6% of men (n = 12,871). Both women and men with CHD were more likely to have traditional CHD risk factors (older age, type 2 diabetes, and history of smoking, dyslipidemia, and hypertension) (Supplementary Table 3).

The sex-specific 270-SNP genetic risk score showed a strong association with type 2 diabetes in both sexes (F-statistic 683 for women and 1,005 for men) (Supplementary Table 2), thus satisfying the first assumption of MR that the genetic instrument is associated with the exposure. We evaluated whether the apparent difference in instrument strength by sex was due to sex differences in the prevalence of type 2 diabetes. In a random subset of UK Biobank participants with 750 cases of type 2 diabetes for both women (n = 18,493) and men (n = 9,100), the adjusted F-statistic of 47 (R^2 = 0.02) for women and adjusted F-statistic of 45 (R^2 = 0.03) for men were similar (data not shown). Thus, because the difference in instrument strength by sex is a product of greater prevalence of type 2 diabetes in men, it is not likely to appreciably affect the comparative validity of estimates derived from MR analyses.

Potential confounders were similarly distributed across quartiles of the genetic risk score for both women and men (Table 2). Conversely, conventional observational analyses showed that type 2 diabetes status was strongly associated with all potential confounders assessed (Table 2), highlighting the need for instrumental variables in this setting.

Individual participant results from TSRI analyses in UK Biobank showed similar effects of genetic risk of type 2 diabetes on CHD for each sex (OR 1.13 [95% CI 1.08–1.18] for women, 1.21 [1.17–1.26] for men) (Table 3). Sensitivity analyses using the weighted median method showed attenuated results (1.04 [1.00–1.08] for women, 1.06 [1.03–1.09] for men) (Table 3). Using MR Egger, evidence of directional pleiotropy was observed in women (OR 1.01 [95% CI 0.96–1.06], intercept 0.004 [95% CI 0.000–0.008]) (Table 3) and men (OR 1.00 [0.96–1.04], intercept 0.008 [0.004–0.011]) (Table 3). Results from MR-PRESSO after outlier correction were slightly attenuated compared with those from TSRI analyses for both women (three outliers removed, OR 1.08 [1.05–1.13]) and men (five outliers removed, 1.13 [1.10–1.17]) (Table 3). Analyses excluding SNPs from the genetic instrument that were identified as outliers by radial MR showed similar effect estimates as the TSRI results (OR 1.09 [1.05–1.14] for women, 1.24 [1.20–1.29] for men) (Table 3). We used additional measures to assess for heterogeneity on the basis of MR-Egger regression, including the Cochran Q test and I^2 statistic. The Q test showed evidence of heterogeneity in the effect of type 2 diabetes SNPs on CHD for both women (Q-statistic 395.8) and men (666.0). The I^2 statistic measures heterogeneity in the genetic associations with the exposure, and results (I^2 = 84.7% for women and 87.1% men) showed some evidence of heterogeneity in the associations of SNPs with type 2 diabetes. Such heterogeneity could be reflective of multiple causal pathways between type 2 diabetes and risk of CHD.

Table 2—Association of sex-specific genetic risk scores (270 SNPs) for type 2 diabetes, by quartile, with potential confounders, and association of observational type 2 diabetes with potential confounders in the UK Biobank

	Genetic type 2 diabetes risk, quartiles of genetic risk score				Type 2 diabetes diagnosis, observational association	
	Q1	Q2	Q3	Q4	No diabetes	Diabetes
Women						
Quartile range	13.82 to <15.94	15.94 to <16.31	16.31 to <16.68	16.68 to <18.71		
Participants, *n*	62,856	62,854	62,855	62,855	241,456	9,964
Height (cm), mean (SD)	162.8 (6.2)	162.6 (6.3)	162.5 (6.2)	162.5 (6.3)	162.7 (6.2)	161.4 (6.3)
Weight (kg), mean (SD)	70.8 (13.8)	71.3 (13.9)	71.6 (14.0)	72.0 (14.1)	70.9 (13.5)	84.6 (18.2)
BMI (kg/m^2), mean (SD)	26.7 (5.1)	27.0 (5.1)	27.1 (5.2)	27.3 (5.2)	26.8 (5.0)	32.5 (6.6)
Waist (cm), mean (SD)	83.7 (12.3)	84.3 (12.4)	84.8 (12.5)	85.5 (12.7)	84.0 (12.0)	99.1 (14.8)
Current smoking, *n* (%)	5,543 (8.8)	5,564 (8.9)	5,755 (9.2)	5,781 (9.2)	21,585 (8.9)	1,058 (10.6)
Dyslipidemia, *n* (%)	5,796 (9.2)	6,108 (9.7)	6,497 (10.3)	7,148 (11.4)	51,833 (21.5)	5,888 (59.1)
Hypertension, *n* (%)	13,190 (21.0)	14,032 (22.3)	14,769 (23.5)	15,730 (25.0)	22,024 (9.1)	3,525 (35.4)
Type 2 diabetes, *n* (%)	1,204 (1.9)	1,898 (3.0)	2,560 (4.1)	4,302 (6.8)		
CHD, *n* (%)	3,008 (4.8)	3,077 (5.0)	3,247 (5.2)	3,348 (5.3)	10,823 (4.5)	1,893 (19.0)
Men						
Quartile range	14.58 to <16.67	16.67 to <17.05	17.05 to <17.43	17.43–19.54		
Participants, *n*	53,014	53,011	53,012	53,012	195,132	16,917
Height (cm), mean (SD)	176.0 (6.8)	175.8 (6.8)	175.8 (6.8)	175.7 (6.8)	175.9 (6.8)	174.7 (6.8)
Weight (kg), mean (SD)	85.9 (14.4)	86.0 (14.3)	86.3 (14.3)	86.5 (14.2)	85.4 (13.7)	95.4 (17.5)
BMI (kg/m^2), mean (SD)	27.7 (4.3)	27.8 (4.3)	27.9 (4.3)	28.0 (4.2)	27.6 (4.0)	31.2 (5.2)
Waist (cm), mean (SD)	96.8 (11.5)	96.9 (11.4)	97.2 (11.3)	97.4 (11.2)	96.2 (10.8)	106.3 (13.2)
Current smoking, *n* (%)	6,417 (12.1)	6,492 (12.2)	6,670 (12.6)	6,555 (12.4)	23,931 (12.3)	2,203 (13.0)
Dyslipidemia, *n* (%)	7,925 (14.9)	8,349 (15.7)	8,473 (16.0)	9,096 (17.2)	27,627 (14.2)	10,749 (63.5)
Hypertension, *n* (%)	15,205 (28.7)	15,784 (29.8)	16,386 (30.9)	17,293 (32.6)	53,919 (27.6)	6,216 (36.7)
Type 2 diabetes, *n* (%)	2,157 (4.1)	3,248 (6.1)	4,495 (8.5)	7,017 (13.2)		
CHD, *n* (%)	6,136 (11.6)	6,512 (12.3)	6,663 (12.6)	7,033 (13.3)	21,132 (10.8)	5,212 (30.8)

Q, quartile.

Table 3—MR analysis of type 2 diabetes and risk of CHD, by sex, in UK Biobank

	Women		Men	
	OR (95% CI)	*P* value	OR (95% CI)	*P* value
Two-stage residual inclusion estimation†	1.13 (1.08–1.18)	5.84×10^{-8}	1.21 (1.17–1.26)	2.31×10^{-24}
Weighted median‡	1.04 (1.00–1.08)	0.067	1.06 (1.03–1.09)	<0.001
MR-Egger‡	1.01 (0.96–1.06)	0.81	1.00 (0.96–1.04)	0.99
MR-PRESSO (outlier corrected)‡	1.08 (1.05–1.13)	3.11×10^{-5}	1.13 (1.10–1.17)	1.57×10^{-12}
Sex-specific outliers removed†‡§	1.09 (1.05–1.14)	6.76×10^{-5}	1.24 (1.20–1.29)	2.78×10^{-27}
	Intercept (95% CI)	*P* value	Intercept (95% CI)	*P* value
MR-Egger (intercept)‡	0.002 (0.000–0.008)	0.027	0.008 (0.004–0.011)	<0.001
Q-test‡	395.8		666.0	
I^2‡, %	84.7		87.1	

Results indicate the increased risk of CHD per 1-log unit increase in genetic risk of type 2 diabetes (OR and 95% CI). Genetic instrument comprised of 270 SNPs for type 2 diabetes identified in the European DIAMANTE GWAS. †Results from two-stage residual inclusion estimation using individual participant data and weighted genetic risk score in UK Biobank. Adjusted for age, genotype array, and principal components of ancestry. *P* value for interaction = 0.02. ‡Results from summary-level analyses using SNP type 2 diabetes estimates from DIAMANTE GWAS (excluding UK Biobank) and SNP CHD estimates from UK Biobank. *P* values for interaction: weighted median = 0.43; MR-Egger = 0.76; and MR-PRESSO = 0.07. §Analysis with type 2 diabetes genetic instrument comprising 258 SNPs for women and 245 SNPs for men, after SNPs identified as sex-specific outliers using radial MR excluded from genetic instrument. *P* value for interaction < 0.001.

CONCLUSIONS

In this MR study of the sex-specific effect of type 2 diabetes on CHD, we found that genetic predisposition to type 2 diabetes does not confer a greater excess risk of CHD for women than for men. While our results are consistent with previous sex-combined MR studies providing support for a causal role of type 2 diabetes in CHD risk (8,9), the finding that the causal effect of genetic liability to type 2 diabetes on CHD risk is not stronger for women than for men is novel and differs from sex-specific estimates from the accumulated observational evidence (2). This includes a recent analysis in the UK Biobank, which showed a stronger association of type 2 diabetes with CHD for women than for men (29).

There are several potential explanations for the differences between the findings of our MR study and the observational evidence. As with any observational study, studies of sex differences in the association of type 2 diabetes with CHD may not have controlled for all relevant confounders or may have controlled for confounders that were poorly measured, leading to residual confounding. If this residual confounding differs between the sexes, a sex difference in the observational association of type 2 diabetes with CHD could arise. For example, men are typically at higher absolute risk of CHD, and the prevalence of many cardiovascular risk factors is higher for men than for women (1). However, cardiovascular risk factors, including type 2 diabetes, appear to confer a greater relative CHD risk for women than for men in observational analyses (29). Furthermore, among individuals with type 2 diabetes compared with those without type 2 diabetes, several studies have shown that the differences in cardiovascular risk factors, including blood pressure, dyslipidemia, and particularly anthropometric variables, are greater among women than among men (3,6). Although women generally display a more favorable cardiometabolic risk profile than men, this favorable risk profile declines and ultimately reverses as glycemic control deteriorates (30).

Yet observational evidence of sex differences in the association of other major risk factors with CHD is not universally observed, suggesting that mechanisms other than confounding alone may be involved. An alternative explanation is that sex differences in the effect of diabetes on CHD risk seen in observational studies reflect the more adverse deterioration in cardiovascular risk profile along the glucose intolerance spectrum in women compared with men. A recent MR study showed that the association of BMI with the risk of diabetes was stronger for women than for men (31). Accordingly, a pathway of type 2 diabetes progression and glycemic dysregulation that leads to more adverse complications of diabetes for women than for men may underpin the observational findings rather than a direct sex difference in the effect of diabetes on CHD risk.

Furthermore, women may be perceived as having lower cardiovascular risk, and consequently, type 2 diabetes and comorbid cardiovascular risk factors may be treated less aggressively (32,33). Guidelines for the diagnosis and treatment of type 2 diabetes and CHD are not sex specific; our results of a similar causal association of type 2 diabetes with CHD by sex would support the notion that for a given state of glycemic dysregulation and burden of cardiovascular risk factors, prevention and management of type 2 diabetes for the reduction of CHD risk should be of equal priority for both women and men. In addition, sex-specific confounders, such as reproductive factors including gestational diabetes mellitus, are rarely adjusted for in observational studies that include both sexes; this could inflate the association of type 2 diabetes with CHD in women if the cumulative duration of the exposure to diabetes is greater, on average, among women than among men. Sex-specific residual confounding may therefore explain some of the discrepancy between the MR and observational evidence. Alternatively, the discrepancy might arise if the MR analysis does not account for genetic variation in the risk of type 2 diabetes that derives from sex chromosomes, as the GWAS data includes only autosomal SNPs. For example, a recent MR study observed a causal association of genetically determined testosterone (X chromosome) with increased type 2 diabetes risk for women but not for men (34). Multiple other mechanisms could also play a role in conferring higher CHD risks for women with type 2 diabetes

compared with men independent of glucose dysregulation or diabetes, including sex differences in microvasculature such as vascular responsivity to aldosterone (35).

The diagnosis of type 2 diabetes is defined by a cut point along a continuum of glycemia that is based on the risk of associated complications, such as retinopathy (36). Accordingly, an individual with borderline glycemia who is not yet diagnosed with type 2 diabetes may display phenotypic and genetic similarity compared with an individual with diagnosed diabetes. Exposure misclassification of this type would tend to bias individual participant MR estimates toward the null, leading to an underestimation of the MR results. In our individual participant MR, this scenario would only affect our conclusion when prediabetes affected a differential proportion of women and men in the study population. Of note, this should not influence summary-level MR results because the exposure is fully defined by genotype.

There are several strengths of our study, including the use of MR, which under specific assumptions can be used to test the hypothesis that a particular risk factor is causal for an outcome (16). In accordance with the first assumption of MR, the sex-specific genetic risk scores were very strong instruments for type 2 diabetes for both women and men. Meeting the second and third assumptions of MR, the genetic risk scores were shown to be broadly independent of measured potential confounding factors. Furthermore, for both women and men, results of sensitivity analyses after correction for outliers were similar to initial results. However, there are also limitations of our study. Although the genetic risk scores were strong instruments for type 2 diabetes, our instruments may have been underpowered to detect modest differences in sex-specific causal effects. Furthermore, our analysis used genetic risk scores derived from 270 genome-wide significant type 2 diabetes SNPs in the sex-combined European DIAMANTE GWAS (14). Genetic instruments obtained from the SNPs that are associated with type 2 diabetes in sex-specific GWASs could also have been constructed. However, the European DIAMANTE GWAS observed only one significant sex-differentiated SNP (14), and thus, the impact of the use of a sex-combined instrument is unlikely to have changed our results substantially. Moreover, such an instrument would not permit direct comparison of sex differences in the overall genetic predisposition to type 2 diabetes but, instead, compares the causal effect of two distinct sex-specific instruments on CHD risk.

SNPs included in the genetic instruments for type 2 diabetes may affect CHD risk through pathways separate from their effect on type 2 diabetes risk, and these pathways could differ by sex. For example, there was some evidence of directional pleiotropy using MR-Egger. However, the intercept for both men and women neared zero, and MR-Egger generally lacks power. Moreover, results from outlier-robust sensitivity analyses were more similar to the overall results. This suggests that our primary results are in fact robust and that MR-Egger results may have been influenced by sensitivity of this method to extreme outliers (37).

These results might reflect multiple different scenarios (38), some of which may have downstream effects on type 2 diabetes risk and may differentially affect CHD risk by sex. Taken together, we cannot exclude a sex-specific causal effect through other pathways not captured in our genetic instrument. Of note, our instrumental variables for type 2 diabetes were derived from the DIAMANTE GWAS effect estimates without adjustment for BMI since the influence of BMI on type 2 diabetes risk may be sex differential (31). Considering the important role of BMI in type 2 diabetes risk, adjusting for measures of adiposity in the type 2 diabetes genetic risk score could bias a true differential effect of type 2 diabetes on CHD to the null. In addition, the UK Biobank and the European DIAMANTE GWAS used for our analyses included primarily European populations, and therefore, we cannot assess sex differences in the causal effect of type 2 diabetes with CHD across ethnicities. Furthermore, despite the large sample size of the UK Biobank, a low overall response rate of ~5.5% limits the generalizability of our results. Considering that the participating population is unlikely representative of the general U.K. population, as recently demonstrated (39), it is possible that our findings might be biased if there is a sex-specific selection bias that is associated with both the exposure and the outcome. Finally, a recent study demonstrated an association of autosomal loci with sex, which may introduce bias as a result of sex differences in study participation (40). If risk alleles for type 2 diabetes were associated with study participation in a sex-specific manner, this may have resulted in an inability to consistently detect a sex difference in the causal effect of type 2 diabetes with CHD in our MR analyses.

In conclusion, the present MR analysis supports a causal effect of type 2 diabetes on the risk of CHD, with similar effects seen between women and men. In the absence of bias, these findings suggest that the prevention and management of type 2 diabetes for the reduction of CHD risk should be of equal priority for both women and men.

Acknowledgments. The authors thank the European DIAMANTE investigators for making data available. This research has been conducted using the UK Biobank resource under application number 24281.

Funding. This research is supported by the Lady Davis Institute for Medical Research and the Department of Medicine, Jewish General Hospital (T.M.P.); the UK Medical Research Council; British Heart Foundation Intermediate Clinical Research Fellowship (FS/18/23/33512); and the National Institute for Health Research (NIHR) Oxford Biomedical Research Centre (M.V.H.). J.B.R. is supported by the Canadian Institutes of Health Research (CIHR), the Lady Davis Institute of the Jewish General Hospital, the Canadian Foundation for Innovation, the NIH Foundation, Cancer Research UK, the Fonds de Recherche Québec Santé (FRQS), and an FRQS Clinical Research Scholarship. C.M.L. is supported by the Li Ka Shing Foundation, the NIHR Oxford Biomedical Research Centre, National Institutes of Health grants CRR00070 CR00.01 and 5P50-HD-028138-27, WT-SSI/John Fell funds, and Widenlife. F.W.A. is supported by University College London Hospitals NIHR Biomedical Research Centre. C.P.N. and N.J.S. are supported by the British Heart Foundation. M.I.M. is supported by Wellcome Trust grants 090532, 098381, 203141, and 212259 and National Institute of Diabetes and Digestive and Kidney Diseases grant U01-DK-105535 and was a Wellcome investigator and an NIHR senior investigator. L.M.O. is supported by a UK Medical Research Council Population Health Scientist fellowship (MR/M014509/1). S.A.E.P. is supported by a UK Medical Research Council Skills Development Fellowship (MR/P014550/1).

The views expressed are those of the authors and not necessarily those of the NHS, the NIHR, or the Department of Health.

Duality of Interest. M.V.H. has collaborated with Boehringer Ingelheim in research and in accordance with the policy of The Clinical Trial Service Unit and Epidemiological Studies Unit

(University of Oxford), did not accept any personal payment. J.B.R. has served as an advisor to GlaxoSmithKline. M.I.M. has served on advisory panels for Pfizer, Novo Nordisk, and Zoe Global; has received honoraria from Merck, Pfizer, Novo Nordisk, and Eli Lilly; and has received research funding from AbbVie, AstraZeneca, Boehringer Ingelheim, Eli Lilly, Janssen, Merck, Novo Nordisk, Pfizer, Roche, Sanofi Aventis, SERVIER, and Takeda. As of June 2019, M.I.M. is an employee of Genentech and a holder of Roche stock. As of January 2020, A.M. is an employee of Genentech and a holder of Roche stock. No other potential conflicts of interest relevant to this article were reported.

Author Contributions. T.M.P. analyzed the data, wrote the manuscript, and contributed to the study design and conception. M.V.H., J.B.R., M.W., and L.M.O. contributed to the study design and reviewed/edited the manuscript. T.P. contributed to the study design and data analysis and reviewed the manuscript. V.F. contributed to the data analysis and reviewed the manuscript. C.M.L., F.W.A., C.P.N., N.J.S., and G.D.S reviewed/edited the manuscript. M.I.M. and A.M. contributed data and reviewed/edited the manuscript. S.A.E.P. contributed to the study design and data analysis and wrote the manuscript. T.M.P. is the guarantor of this work and, as such, had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

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