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Sex Hormone-Binding Globulin and Risk of Coronary Heart Disease in Men and Women

Jie Li[a,b,c,d,*,†], **Lingling Zheng**[a,†], **Kei Hang Katie Chan**[d,e,†], **Xia Zou**[a,†], **Jihui Zhang**[f], **Jundong Liu**[e], **Qingwei Zhong**[a], **Tracy E. Madsen**[d,g], **Wen-Chih Wu**[d,h], **JoAnn E. Manson**[i], **Xueqing Yu**[a,j,*], **Simin Liu**[a,d,k,*]

[a]Global Health Research Center, Guangdong Provincial People's Hospital, Guangdong Academy of Medical Sciences, Guangzhou, China

[b]The Second School of Clinical Medicine, Southern Medical University, Guangzhou 510280, China

[c]School of Medicine, South China University of Technology, Guangzhou 510006, China

[d]Department of Epidemiology and Center for Global Cardiometabolic Health, School of Public Health, Brown University, Providence, RI, USA

[e]Departments of Biomedical Sciences and Electrical Engineering, City University of Hong Kong, Hong Kong SAR, China

[*]Address correspondence to: J.L. at Global Health Research Center, Guangdong Provincial People's Hospital, Guangdong Academy of Medical Sciences, Guangzhou 510080, China. jie_li@brown.edu. S.L. at Department of Epidemiology and Center for Global Cardiometabolic Health, School of Public Health, Brown University, Providence, RI 02906. simin_liu@brown.edu. X.Y. at Department of Nephrology, Guangdong Provincial People's Hospital, Guangdong Academy of Medical Sciences, Guangzhou, Guangdong 510080, China. yuxueqing@gdph.org.cn.

[†]JL, LZ, KC, and XZ contributed equally to this work.

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Supplementary Material

Supplementary material is available at *Clinical Chemistry* online.

Human Genes: *GKRP*, glucokinase regulatory protein; *GCK*, glucokinase; *ChREBP*, carbohydrate response element-binding protein; *HNF4α*, hepatocyte nuclear factor 4 alpha; *PNPLA3*, patatin-like phospholipase domain-coding protein 3.

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[f]Guangdong Mental Health Center, Guangdong Provincial People's Hospital, Guangdong Academy of Medical Sciences, Guangzhou, China

[g]Department of Emergency Medicine, The Warren Alpert Medical School, Brown University, Providence, RI, USA

[h]Department of Medicine, Division of Cardiology, Providence VA Medical Center and Warren Alpert Medical School of Brown University, Providence, RI, USA

[i]Department of Medicine, Brigham and Women's Hospital, Harvard Medical School, and Department of Epidemiology, Harvard T.H. Chan School of Public Health, Boston, MA, USA

[j]Department of Nephrology, Guangdong-Hong Kong Joint Laboratory on Immunological and Genetic Kidney Diseases, Guangdong Provincial People's Hospital, Guangdong Academy of Medical Sciences, Guangzhou, China

[k]Departments of Medicine and Surgery, The Warren Alpert Medical School, Brown University, Providence, RI, USA.

Abstract

BACKGROUND: The role of sex hormone-binding globulin (SHBG) levels in clinical risk stratification and intervention for coronary heart disease (CHD) remains uncertain. We aimed to examine whether circulating levels of SHBG are predictive of CHD risk in men and women.

METHODS: We investigated the association between SHBG and the risk of incident CHD in 128 322 men and 135 103 women free of CHD at baseline in the prospective United Kingdom Biobank (UKB) cohort. The unconfounded associations were estimated using Mendelian randomization (MR) analysis. We further conducted a meta-analysis to integrate currently available prospective evidence. CHD events included nonfatal and fatal myocardial infarction and coronary revascularization.

RESULTS: In the UKB, during a median of 11.7 follow-up years, 10 405 men and 4512 women developed CHD. Serum levels of SHBG were monotonically associated with a decreased risk of CHD in both men (adjusted hazard ratio [HR] per log nmol/L increase in SHBG: 0.88 [0.83–0.94]) and women (HR: 0.89 [0.83–0.96]). MR-based analyses suggested causality and a dose-response relationship of SHBG with CHD risk. A cumulative meta-analysis including 216 417 men and 138 282 women from 11 studies showed that higher levels of SHBG were prospectively associated with decreased CHD risk in men comparing the highest with the lowest quartile: pooled relative risk (RR) 0.81 (0.74–0.89) and women (pooled RR: 0.86 [0.78–0.94]).

CONCLUSIONS: Higher circulating SHBG levels were directly and independently predictive of lower CHD risk in both men and women. The utility of SHBG for CHD risk stratification and prediction warrants further study.

Introduction

Sex hormone binding globulin (SHBG) was first identified as the principal protein regulating the bioavailability of sex hormones by binding circulating testosterone and estradiol with high affinity (1, 2). Our previous work in several large prospective cohorts has shown low

circulating levels of SHBG as a strong predictor for risks of type 2 diabetes (T2D) and metabolic syndrome in men and women, naturally leading to the expectation that low SHBG would be associated with high risk of cardiovascular diseases (3–6). A recent study also confirmed that low SHBG could be a biomarker of metabolic dysfunction, for example de novo lipogenesis, in the liver and that SHBG is an hepatokine, playing a direct and causal role in the pathogenesis of T2D (7, 8). T2D, dyslipidemia, and metabolic syndrome are well-defined risk factors for coronary heart disease (CHD), and they share similar metabolic pathology mechanisms (9, 10). However, previous studies have shown either positive (11–14), negative (15–17), or null (18–20) associations of the levels of SHBG with CHD risk, with limited data in women.

Given the observational nature of previous study designs, these findings on the SHBG-CHD association may have been biased by sample selection, confounding factors, or reverse causality. Thanks to the increasing number of genome-wide association studies, Mendelian randomization (MR) based analysis can now be readily employed to estimate the strength of “unconfounded” associations using single nucleotide polymorphisms (SNPs) as randomization instruments for specific exposures of interest (e.g., SHBG). Given that alleles of SNPs are randomly assigned and fixed at conception, MR is considered analogous to a randomized controlled trial, supporting causal estimates of exposure on outcome (21).

To clarify the associations of SHBG with CHD risk, we comprehensively assessed all observational data available to date relating circulating levels of SHBG to CHD risk in men and women and then performed MR-based analyses with SHBG-related genetic variants as instruments to aid in causal inference.

Methods

STUDY DESIGN

This study consisted of 3 sequential parts, with the aim to elucidate the sex-specific association of SHBG with the risk of CHD. First, we investigated the prospective association between serum levels of SHBG and the risk of incident CHD in men and women in the United Kingdom Biobank (UKB) cohort. Second, we performed an MR-based analysis to further determine the unconfounded association between levels of SHBG and CHD risk by utilizing recently identified SHBG-related sex-specific genetic variants as instrumental variables. Third, we conducted a meta-analysis to comprehensively evaluate sources of heterogeneity and calculate summary statistics integrating available studies linking SHBG to CHD risk.

STUDY POPULATION

The UKB is a large cohort study with >500 000 participants ages 40 to 69 years recruited from 22 assessment centers across the United Kingdom in 2006–2010. Detailed information on UKB is provided elsewhere (22). We excluded participants based on the following criteria: (*a*) with diagnosed cardiovascular diseases or cancers at baseline; (*b*) with prior pituitary disease, infertility, orchidectomy, or congenital adrenal hyperplasia or receiving androgen, anti-androgen, or other hormone therapy; (*c*) without measured

serum levels of SHBG; (*d*) without genetic data or genotyping quality control data; (*e*) with sex discordance between self-reported and genetically predicted sex; (*f*) with extreme heterozygosity indicating poor DNA sample quality; (*g*) with sex chromosome aneuploidy; (*h*) with genetic kinship to other participants (10 or more third-degree relatives identified); and (*i*) with >2% of instrumental SNPs with missing data (23). A total of 128 322 men and 135 103 women were eligible for subsequent analyses (Supplemental Fig. 1). This study was performed under ethical approval obtained by UKB from the National Health Service National Research Ethics Service.

MEASUREMENT OF SERUM LEVELS OF SHBG, TESTOSTERONE, AND ESTRADIOL IN THE UKB

Blood samples were collected at baseline (2006–2010) and stored at −80°C. Serum levels of SHBG, testosterone, and estradiol were measured in the UKB central laboratory between June 2019 and April 2020 by chemiluminescent immunoassay (DXI 800; Beckman Coulter). For the SHBG assay, the CVs were 5.7%, 5.3%, and 5.2% for concentration ranges of 15.0 to 27.7, 31.9 to 55.5, and 56.3 to 87.8 nmol/L (1.43–2.63, 3.03–5.27, 5.35–8.34 mg/L), respectively, and the detection range was 0.33 to 242 nmol/L (0.03–22.99 mg/L). For the testosterone assay, the CVs were 8.3%, 3.7%, and 4.2% for concentrations ranges of 1.0 to 2.2, 13.4 to 22.8, and 29.3 to 49.4 nmol/L (0.29–0.63, 3.86–6.57, 8.44–14.23 μg/L), respectively, and the detection range was 0.35 to 55.52 nmol/L (0.10–15.99 μg/L). For the estradiol assay, the CVs were 15.3%, 8.7%, and 6.5% for concentrations ranges of 175 to 417, 446 to 990, and 1305 to 2503 pmol/L (48–113, 121–270, 355–682 ng/L), respectively, and the detection range was 73 to 17 621 pmol/L (20–4800 ng/L).

DETERMINATION OF INCIDENT CHD CASES IN THE UKB

The incidence of CHD, including nonfatal and fatal myocardial infarction and coronary revascularization (coronary artery bypass surgery or percutaneous transluminal coronary intervention), was identified based on the linked hospital admissions and mortality data according to the International Classification of Diseases 10 code (I21–I25) and the Office of Population Censuses and Surveys Classification of Interventions and Procedures (OPCS-4) code (K40–K50, K75) (Supplemental Table 1). Each participant's follow-up time was defined as the duration between entry to the cohort and the date of occurrence of CHD or censor (death, lost to follow-up, or the end of follow-up [January 28, 2021]), whichever occurred first.

GENETIC INSTRUMENTS

Given that sex hormones may exert different effects on CHD risk in men vs women and that genetic determinants of sex hormones and SHBG have sex differences (7), we selected sex-specific instrumental variables for SHBG in MR analyses. A recent sex-specific genome-wide association studies in 425 097 UKB participants identified 357 genome-wide significant SNPs for SHBG, independent of body mass index (BMI), in men and 359 SNPs in women (Supplemental Tables 11 and 12) (7). We calculated the genetic risk scores (GRS) for SHBG by summing up the number of effect alleles (i.e., higher GRSs predict higher SHBG levels), which were used as instrumental variates in the current MR analysis. Before the calculation, the expected dosage for each imputed SNP was rounded to an integer value.

COVARIATES

Demographic characteristics, lifestyle factors, and other confounders collected at baseline for each participant were included in our models as covariates, including age, Townsend deprivation index, physical activity, smoking status, alcohol intake, BMI, family history of cardiovascular diseases, and presence of medical conditions at baseline, such as diabetes, hypertension, and dyslipidemia. The rates of missing data for most covariates were less than 1%, and mean and mode imputation were used to replace the missing data for continuous and categorical variables, respectively.

STATISTICAL ANALYSIS

We used Cox proportional hazard models to estimate sex-specific associations of serum levels of SHBG with risk of incident CHD with adjustment for the aforementioned covariates and serum levels of total testosterone. We also included a restricted cubic spline term for SHBG with 3 knots at the 10th, 50th, and 90th centiles into the model to explore the nonlinear relationship between SHBG and CHD risk. The nonlinearity *P* value was estimated using a likelihood ratio test. In a series of sensitivity analyses, we further excluded participants with diabetes, hypertension, or dyslipidemia at baseline, and CHD cases that occurred during the first 2 years of follow-up from the analysis. In the UKB, serum levels of estradiol were measured in only a very small portion of participants, so we were unable to take estradiol into consideration in the main analysis. To evaluate the impact of estradiol on the SHBG-CHD association, we conducted a sensitivity analysis with further adjustment for serum levels of estradiol in 10 630 men and 29 062 women with measured levels of SHBG and estradiol in the UKB.

In linear MR analyses, we used a two-stage least square regression with adjustment for age, BMI, assessment center, genotyping batch, and the top 10 genetic principal components to estimate the causal effect of genetically predicted SHBG on CHD risk in men and women (24). Results of MR analysis were presented as predictive odds ratios of CHD for a 1-unit increase in genetically predicted SHBG levels. We conducted a series of sensitivity analyses to test if the assumptions for MR-based assessment were violated using different strategies. Due to the correlation between total testosterone (TT) and SHBG, we performed MR analysis using restricted GRS, in which the overlapped SNPs between hormones were removed (Supplemental Fig. 2) and multivariable MR analyses with further adjustment for genetically predicted TT. We also calculated the HR by excluding prevalent cases at baseline to estimate the association between genetically predicted SHBG levels and CHD risk. For nonlinear MR analysis, we used a fractional polynomial method to examine the shape of the relationship between genetically predicted levels of sex hormones and SHBG and CHD risk (25). Nonlinearity was tested using the quadratic test (25). Detailed MR methods are described in the Supplemental Methods section.

In the meta-analysis (Registration ID in PROSPERO: CRD42021241706), we systematically searched for prospective cohort and nested case-control studies relating circulating levels of SHBG to CHD published in Medline and Embase from database inception to June 2022. We calculated the sex-specific relative risks (RRs) of CHD by comparing the highest with the lowest quartiles of SHBG levels via an inverse variance pooling method. We pooled RRs

of CHD with fixed effects models and evaluated heterogeneity across studies by examining the funnel plots of RR estimates as well as using Cochran Q test and I^2 statistics (with I^2 of 0%–25% representing minimal, 26%–75% moderate, and >75% substantial heterogeneity). As sensitivity analyses, we performed a cumulative meta-analysis with studies ranked by published year to test the stability and sufficiency of the evidence as it accumulated over time. Detailed methods for meta-analysis are summarized in Supplemental Methods.

All analyses were conducted using R (v3.5.1) and packages, including *meta, MendelianRandom,* and *nlmr,* with 95% CIs and 2-sided *P* values calculated for statistical inference.

Results

ASSOCIATION BETWEEN SERUM LEVELS OF SHBG AND RISK OF INCIDENT CHD IN MEN AND WOMEN IN THE UKB

During a median follow-up of 11.7 years, 10 405 participants developed CHD among the 128 322 men (mean age of 56.1 at baseline) and 4512 developed CHD among the 135 103 women (mean age 56.4 years). Men had lower serum levels of SHBG than women (median interquartile range [IQR]: 37.0 [28.1–48.1] nmol/L in men vs 56.1 [40.4–75.4] nmol/L in women). Men with higher levels of SHBG were older and had higher levels of TT. In contrast, women with higher levels of SHBG tended to be younger and have higher levels of estradiol. Both men and women with higher levels of SHBG were more likely to be smokers and physically active and had lower BMI and prevalence of diabetes, hypertension, and hyperlipidemia (Table 1).

In the fully adjusted model, higher levels of SHBG were associated with 14% lower incident CHD in men (adjusted HR comparing the highest with the lowest quintile: 0.86 [95% CI, 0.79–0.93], *P* for trend <0.001) and women (HR: 0.84 [0.75–0.94], *P* for trend<0.001) (Table 2). Nonlinear analysis showed a linear shape relationship between serum levels of SHBG and CHD risk in men (*P* for nonlinear=0.62) and women (*P* for nonlinear=0.54) (Fig. 1, A and C). Each log nmol/L unit increase in serum levels of SHBG was associated with a decreased risk of incident CHD in men (adjusted HR: 0.88 [0.83–0.94]) and women (adjusted HR: 0.89 [0.83–0.96]) (Table 3). These findings were robust in all sensitivity analyses (Supplemental Table 3).

MR-BASED ASSESSMENT OF SHBG LEVELS WITH CHD RISK IN MEN AND WOMEN IN THE UKB

The established sex-specific GRSs explained 7.6% and 5.3% variance of measured SHBG in men and women, respectively, with all F statistics far greater than 10, supporting the reliability of the genetic instrumental variables (Supplemental Fig. 2). We described the characteristics of men and women according to quintiles of GRSs in Supplemental Table 4 and found that most of the covariates were balanced across quintiles, which means that the MR process was reliable. The unbalanced covariates, such as alcohol intake, BMI, and TT, were adjusted in the regression models. Simple analysis showed that the GRS for SHBG was

positively associated with levels of SHBG but inversely associated with CHD risk both in men and women (Supplementary Tables 3 and 5).

Nonlinear MR analyses showed that an increase in genetically predicted SHBG was monotonically related to a lower CHD risk in men (*P* for nonlinear=0.73) and women (*P* for nonlinear=0.93) (Fig. 1, B and D). Linear MR analysis showed that each unit increase in the genetically predicted SHBG was associated with a decreased CHD risk both in men (predicted odds ratio: 0.75 [0.63–0.89]) and women (predicted odds ratio: 0.69 [0.54–0.89]) (Table 3).

We further conducted a series of sensitivity analyses to test the robustness of the MR estimates. Given the strong intercorrelation between TT and SHBG, we removed the overlapped instrumental SNPs to construct a restricted GRS for SHBG (Supplemental Fig. 2) and obtained consistent findings (Supplemental Table 6). Two-sample MR strategies, including inverse-variance weighted, weighted median, weighted mode, and MR-Egger, also provided consistent results, indicating that our findings were robust and not likely to be affected by pleiotropy or outliers (Supplemental Table 6).

META-ANALYSIS OF ASSOCIATION OF CIRCULATING LEVELS OF SHBG WITH CHD RISK IN MEN AND WOMEN

Among the 10 unique observational studies included in the meta-analysis, a total of 5104 CHD cases were confirmed in 216 417 men from 6 studies and 4936 CHD cases in 138 282 women from 5 studies (Supplemental Tables 8 and 9). The funnel plots and Egger test showed no evidence of publication bias (Supplemental Fig. 4).

Circulating levels of SHBG were associated with a lower risk of CHD in men (pooled RR: 0.81 [0.74–0.89], I^2=0%) and women (pooled RR: 0.86 [0.78–0.94], I^2=19%) with no substantial heterogeneity in estimates across studies (Fig. 2, A and C). In the meta-analyses with fixed-effect model, the UKB results accounted for the majority of the weight (80.9% in men and 88.2% in women). Cumulative meta-analysis showed that, if the UKB data were not included, levels of SHBG were associated with a lower risk of CHD in men (pooled RR: 0.74 [0.59–0.92]) but not in women (pooled RR: 1.00 [0.77–1.29]) (Fig. 2, B and D).

Discussion

Higher serum levels of SHBG were monotonically associated with a decreased risk of incident CHD in both men and women in the UKB cohort. The causality and linear shape of these observational associations were confirmed by MR-based analyses in the cohort. A systematic review and cumulative meta-analysis of 10 studies that enrolled 216 417 men and 138 282 women from diverse populations further confirmed these findings.

Previously, we have shown that SHBG levels were directly predictive of T2D risk and metabolic syndrome in several prospective cohorts of diverse populations (3, 4, 6, 26), indicating that SHGB may be a reliable measure that is predictive of lower cardiometabolic risk in men and women. However, most of the previous studies were conducted in men and have not examined the association with CHD outcomes. A recent analysis of the UKB data

has reported an inverse association of SHBG levels with risk of myocardial infarction in men but did not examine the association in women (27). In the current study, our analysis in the UKB not only confirmed that SHBG levels were associated with a lower risk of CHD in men but also found a similar association in women. Furthermore, our cumulative meta-analysis of all available data to date showed that the relationship between SHBG levels and CHD risk in both men and women is both consistent and reliable.

The results of meta-analysis were mainly driven by the large UKB cohort, which accounted for more than 97% of total participants, especially in the analysis for women. All studies included in the meta-analysis recruited adults from Europe and America. It is highly likely that any discrepancy of findings between the UKB and other studies is attributable to the overwhelmingly large sample size in the UKB but not different characteristics between studies.

Although these observational findings may be subject to residual confounders or reverse causation, we have evaluated and adjusted for known confounders in the models. Most importantly, our MR-based analysis not only confirmed the “unconfounded” association but also indicated a dose-response causal relationship between SHBG and risk of CHD in both men and women. These findings are consistent with a previous MR study that used the same set of genetic variants as sex-specific instrumental variables for SHBG and reported a protective effect of SHBG on T2D in both men and women (7).

The SHBG molecule has 2 types of binding site: one for steroids and the other for the SHBG receptor. The steroid binding site is capable of binding testosterone or estradiol; therefore, SHBG has been considered the principal protein regulating the bioavailability of sex hormones through binding with them (8). In addition to regulating the bioavailable fraction of sex hormones, SHBG can directly bind to its membrane receptor and permit certain steroid hormones to act without entering the target cell (4, 28). SHBG receptors primarily exist in reproductive tissues and hepatocytes but have low expression levels in skeletal muscle (29–31). Unoccupied SHBG (not bound to sex steroids) has the ability to bind to the SHBG receptor. However, the binding of SHBG to its receptor cannot initiate a downstream signal until the subsequent binding of a steroid to the SHBG-receptor complex. The sex steroid-SHBG-receptor complex can have either an agonist or antagonist effect via the cAMP-mediated second messenger system, depending on the specific sex steroid and target tissue (28).

Consistent with the biological function of SHBG, men with higher levels of SHBG have higher levels of total testosterone and lower levels of bioavailable testosterone. The associations of endogenous testosterone and estradiol with CHD risk remain uncertain. While several studies found no clear association (32–35), some reported inverse associations between testosterone and CVD risk (15, 27, 36). However, a recent MR-based analysis concluded that endogenous testosterone was associated with increased risk of myocardial infarction in men (37). Similarly, some studies reported that higher levels of estradiol were associated with a lower CHD risk in women (18), while others reported no association in either men or women (38, 39). In the current study, additional adjustment for TT and estradiol in analyses did not materially change the inverse association between SHBG

and CHD risk. Therefore, the observed SHBG-CHD association may be independent of endogenous testosterone and estradiol, despite the close relationship between SHBG and sex steroids. Further studies are needed to elucidate the mechanism underlying the SHBG-CHD association.

Previously, we have demonstrated that germline mutations of *SHBG* and the level of SHBG directly predict risk of clinical T2D (3, 5). Additionally, consistent with other reports (40, 41), we also found that low SHBG levels were associated with low levels of high-density lipoprotein and high levels of C-reactive protein (data not shown). T2D, low high-density lipoprotein, and high C-reactive protein are well-established risk factors for CHD and may serve as mediators linking low SHBG to high CHD risk.

SHBG is mainly produced by hepatocytes, which can be influenced by genetic variants and metabolic factors. A recent large-scale genome-wide association study identified several genetic variants that are significantly associated with serum SHBG, which encode genes involved in the de novo lipogenesis, such as glucokinase regulatory protein (*GKRP*), glucokinase (*GCK*), carbohydrate response element-binding protein (*ChREBP*), hepatocyte nuclear factor 4 alpha (*HNF4α*), and patatin-like phospholipase domain-coding protein 3 (*PNPLA3*) (7, 8). A previous study indicates that monosaccharide-induced lipogenesis reduces hepatic HNF4α levels and then inhibits SHBG expression (42). These findings suggest that SHBG is a sensitive biomarker of metabolic processes in the liver, including de novo lipogenesis. Therefore, the observed inverse association between SHBG and CHD may be interpreted as indicating that SHBG provides a mechanism linking acute-phase reactants due to hepatic metabolic dysfunction to CHD.

Our findings benefit from several strengths of the study design. The current study includes the first comprehensive and systematic analysis of prospective studies available to date assessing the roles of SHBG in relation to CHD risk in approximately 263 439 men and women from diverse populations. Second, the causality of findings from our observational analysis was further confirmed by both linear and nonlinear MR-based analyses, which directly addressed and corrected potential biases due to confounders or reverse causation in previous studies. Third, given the strong correlation between SHBG and sex hormones, we conducted sex-specific analyses and found consistent findings in both sexes.

Several important issues should be kept in mind when interpreting these findings. First, in the meta-analysis, results of the UKB accounted for the vast majority of the weight. After the removal of the UKB study, SHBG levels were associated with a lower risk of CHD only in men but not in women. The neutral association in women may be due to small sample size resulting in limited statistical power. Second, MR-based analyses estimated genetically predicted CHD risk due to SHBG levels that reflect a lifelong cumulative effect. Third, the estimates of 1-sample MR analysis may be biased by weak instruments and potential confounding. A simulation study supports using 2-sample MR methods to validate the findings of 1-sample MR performed within large biobanks (43). In the current study, both 1-sample and 2-sample MR methods gave consistent results. Additionally, the large F statistics of the genetic instruments in the current study indicate that our findings may not be biased by weak instruments.

Conclusions

In summary, our comprehensive and systematic analysis integrating all available observational data from diverse populations indicates that higher levels of SHBG measured in adulthood or predicted by genetic variants were prospectively associated with lower CHD risk in men and women, independent of known and unknown CHD risk factors. These findings suggest an important causal role of SHBG in the development of CHD. The utility of SHBG for CHD risk stratification and prediction and monitoring of treatment response warrants further study. Further mechanistic work to understand downstream targets of SHBG may also represent a promising direction to mitigate risks of cardiometabolic outcomes.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Data Availability

All data used in prospective cohort analysis and Mendelian randomization analysis are available from UKB upon request (https://www.ukbiobank.ac.uk).

Nonstandard Abbreviations:

SHBG	sex hormone binding globulin
T2D	type 2 diabetes
CHD	coronary heart disease
MR	Mendelian randomization
SNP	single nucleotide polymorphism
UKB	United Kingdom Biobank
BMI	body mass index
GRS	genetic risk scores

TT	total testosterone
RR	relative risk
IQR	interquartile range

References

1. Dunn JF, Nisula BC, Rodbard D. Transport of steroid hormones: binding of 21 endogenous steroids to both testosterone-binding globulin and corticosteroid-binding globulin in human plasma. J Clin Endocrinol Metab 1981;53:58–68. [PubMed: 7195404]
2. Joseph DR. Structure, function, and regulation of androgen-binding protein/sex hormone-binding globulin. Vitam Horm 1994;49:197–280. [PubMed: 7810071]
3. Ding EL, Song Y, Manson JE, Hunter DJ, Lee CC, Rifai N, et al. Sex hormone-binding globulin and risk of type 2 diabetes in women and men. N Engl J Med 2009;361:1152–63. [PubMed: 19657112]
4. Chen BH, Brennan K, Goto A, Song Y, Aziz N, You NC, et al. Sex hormone-binding globulin and risk of clinical diabetes in American Black, Hispanic, and Asian/Pacific Islander postmenopausal women. Clin Chem 2012;58:1457–66. [PubMed: 22908136]
5. Perry JR, Weedon MN, Langenberg C, Jackson AU, Lyssenko V, Sparsø T, et al. Genetic evidence that raised sex hormone binding globulin (SHBG) levels reduce the risk of type 2 diabetes. Hum Mol Genet 2010;19:535–44. [PubMed: 19933169]
6. Li C, Ford ES, Li B, Giles WH, Liu S. Association of testosterone and sex hormone-binding globulin with metabolic syndrome and insulin resistance in men. Diabetes Care 2010;33:1618–24. [PubMed: 20368409]
7. Ruth KS, Day FR, Tyrrell J, Thompson DJ, Wood AR, Mahajan A, et al. Using human genetics to understand the disease impacts of testosterone in men and women. Nat Med 2020;26:252–8. [PubMed: 32042192]
8. Simons P, Valkenburg O, Stehouwer CDA, Brouwers M. Sex hormone-binding globulin: biomarker and hepatokine? Trends Endocrinol Metab 2021;32:544–53. [PubMed: 34052096]
9. Chan KH, Huang YT, Meng Q, Wu C, Reiner A, Sobel EM, et al. Shared molecular pathways and gene networks for cardiovascular disease and type 2 diabetes mellitus in women across diverse ethnicities. Circ Cardiovasc Genet 2014;7:911–9. [PubMed: 25371518]
10. Shu L, Chan KHK, Zhang G, Huan T, Kurt Z, Zhao Y, et al. Shared genetic regulatory networks for cardiovascular disease and type 2 diabetes in multiple populations of diverse ethnicities in the United States. PLoS Genet 2017;13:e1007040. [PubMed: 28957322]
11. Schederecker F, Cecil A, Prehn C, Nano J, Koenig W, Adamski J, et al. Sex hormone-binding globulin, androgens and mortality: the KORA-F4 cohort study. Endocr Connect 2020;9:326–36. [PubMed: 32168474]
12. Gyawali P, Martin SA, Heilbronn LK, Vincent AD, Jenkins AJ, Januszewski AS, et al. Higher serum sex hormone-binding globulin levels are associated with incident cardiovascular disease in men. J Clin Endocrinol Metab 2019;104:6301–15. [PubMed: 31390027]
13. Wang A, Gerstein HC, Lee SF, Hess S, Paré G, Rydén L, Mellbin LG. Testosterone and sex hormone-binding globulin in dysglycemic women at high cardiovascular risk: a report from the outcome reduction with an initial glargine intervention trial. Diab Vasc Dis Res 2021;18:14791641211002475. [PubMed: 33752449]
14. Yeap BB, Marriott RJ, Antonio L, Chan YX, Raj S, Dwivedi G, et al. Serum testosterone is inversely and sex hormone-binding globulin is directly associated with all-cause mortality in men. J Clin Endocrinol Metab 2021;106:e625–e37. [PubMed: 33059368]
15. Ohlsson C, Barrett-Connor E, Bhasin S, Orwoll E, Labrie F, Karlsson MK, et al. High serum testosterone is associated with reduced risk of cardiovascular events in elderly men. The MrOS (Osteoporotic Fractures in Men) study in Sweden. J Am Coll Cardiol 2011;58:1674–81. [PubMed: 21982312]

16. Schaffrath G, Kische H, Gross S, Wallaschofski H, Völzke H, Dörr M, et al. Association of sex hormones with incident 10-year cardiovascular disease and mortality in women. Maturitas 2015;82:424–30. [PubMed: 26385535]
17. Malipatil NS, Yadegarfar G, Lunt M, Keevil B, Siddals K, Livingston M, et al. Male hypogonadism: 14-year prospective outcome in 550 men with type 2 diabetes. Endocrinol Diabetes Metab 2019;2:e00064. [PubMed: 31294081]
18. Zhao D, Guallar E, Ouyang P, Subramanya V, Vaidya D, Ndumele CE, et al. Endogenous sex hormones and incident cardiovascular disease in postmenopausal women. J Am Coll Cardiol 2018;71:2555–66. [PubMed: 29852978]
19. Holmboe SA, Skakkebaek NE, Juul A, Scheike T, Jensen TK, Linneberg A, et al. Individual testosterone decline and future mortality risk in men. Eur J Endocrinol 2018;178:123–30. [PubMed: 29066571]
20. Wehr E, Pilz S, Boehm BO, März W, Grammer TB, Obermayer-Pietsch B. Sex steroids and mortality in men referred for coronary angiography. Clin Endocrinol (Oxf) 2010;73:613–21. [PubMed: 20718764]
21. Lawlor DA, Harbord RM, Sterne JA, Timpson N, Davey Smith G. Mendelian randomization: using genes as instruments for making causal inferences in epidemiology. Stat Med 2008;27:1133–63. [PubMed: 17886233]
22. Sudlow C, Gallacher J, Allen N, Beral V, Burton P, Danesh J, et al. UK Biobank: an open access resource for identifying the causes of a wide range of complex diseases of middle and old age. PLoS Med 2015;12:e1001779. [PubMed: 25826379]
23. Dashti HS, Daghlas I, Lane JM, Huang Y, Udler MS, Wang H, et al. Genetic determinants of daytime napping and effects on cardiometabolic health. Nat Commun 2021;12:900. [PubMed: 33568662]
24. Boef AG. Dekkers OM, le Cessie S. Mendelian randomization studies: a review of the approaches used and the quality of reporting. Int J Epidemiol 2015;44:496–511. [PubMed: 25953784]
25. Staley JR, Burgess S. Semiparametric methods for estimation of a nonlinear exposure-outcome relationship using instrumental variables with application to Mendelian randomization. Genet Epidemiol 2017;41:341–52. [PubMed: 28317167]
26. Ding EL, Song Y, Malik VS, Liu S. Sex differences of endogenous sex hormones and risk of type 2 diabetes: a systematic review and meta-analysis. JAMA 2006;295:1288–99. [PubMed: 16537739]
27. Yeap BB, Marriott RJ, Antonio L, Raj S, Dwivedi G, Reid CM, et al. Associations of serum testosterone and sex hormone-binding globulin with incident cardiovascular events in middle-aged to older men. Ann Intern Med 2022;175:159–70. [PubMed: 34958606]
28. Rosner W, Hryb DJ, Khan MS, Nakhla AM, Romas NA. Sex hormone-binding globulin mediates steroid hormone signal transduction at the plasma membrane. J Steroid Biochem Mol Biol 1999;69:481–5. [PubMed: 10419028]
29. Nakhla AM, Khan MS, Rosner W. Biologically active steroids activate receptor-bound human sex hormone-binding globulin to cause LNCaP cells to accumulate adenosine 3’,5’-monophosphate. J Clin Endocrinol Metab 1990;71:398–404. [PubMed: 2166070]
30. Frairia R, Fortunati N, Berta L, Fazzari A, Fissore F, Gaidano G. Sex steroid binding protein (SBP) receptors in estrogen sensitive tissues. J Steroid Biochem Mol Biol 1991;40:805–12. [PubMed: 1659893]
31. Krupenko SA, Krupenko NI, Danzo BJ. Interaction of sex hormone-binding globulin with plasma membranes from the rat epididymis and other tissues. J Steroid Biochem Mol Biol 1994;51:115–24. [PubMed: 7947346]
32. Smith GD, Ben-Shlomo Y, Beswick A, Yarnell J, Lightman S, Elwood P. Cortisol, testosterone, and coronary heart disease: prospective evidence from the Caerphilly study. Circulation 2005;112:332–40. [PubMed: 16009799]
33. Arnlov J, Pencina MJ, Amin S, Nam BH, Benjamin EJ, Murabito JM, et al. Endogenous sex hormones and cardiovascular disease incidence in men. Ann Intern Med 2006;145:176–84. [PubMed: 16880459]

34. Vikan T, Schirmer H, Njolstad I, Svartberg J. Endogenous sex hormones and the prospective association with cardiovascular disease and mortality in men: the Tromsø study. Eur J Endocrinol 2009;161:435–42. [PubMed: 19542243]

35. Chan YX, Knuiman MW, Hung J, Divitini ML, Beilby JP, Handelsman DJ, et al. Neutral associations of testosterone, dihydrotestosterone and estradiol with fatal and non-fatal cardiovascular events, and mortality in men aged 17–97 years. Clin Endocrinol (Oxf) 2016;85:575–82. [PubMed: 27106765]

36. Yeap BB, Alfonso H, Chubb SA, Hankey GJ, Handelsman DJ, Golledge J, et al. In older men, higher plasma testosterone or dihydrotestosterone is an independent predictor for reduced incidence of stroke but not myocardial infarction. J Clin Endocrinol Metab 2014;99:4565–73. [PubMed: 25268392]

37. Luo S, Au Yeung SL, Zhao JV, Burgess S, Schooling CM. Association of genetically predicted testosterone with thromboembolism, heart failure, and myocardial infarction: Mendelian randomisation study in UK Biobank. BMJ 2019;364:l476. [PubMed: 30842065]

38. Peters SAE, Woodward M. Oestradiol and the risk of myocardial infarction in women: a cohort study of UK Biobank participants. Int J Epidemiol 2021;50:1241–9. [PubMed: 33734370]

39. Menke A, Guallar E, Rohrmann S, Nelson WG, Rifai N, Kanarek N, et al. Sex steroid hormone concentrations and risk of death in US men. Am J Epidemiol 2010;171:583–92. [PubMed: 20083549]

40. Pugeat M, Moulin P, Cousin P, Fimbel S, Nicolas MH, Crave JC, Lejeune H. Interrelations between sex hormone-binding globulin (SHBG), plasma lipoproteins and cardiovascular risk. J Steroid Biochem Mol Biol 1995;53:567–72. [PubMed: 7626511]

41. Joffe HV, Ridker PM, Manson JE, Cook NR, Buring JE, Rexrode KM. Sex hormone-binding globulin and serum testosterone are inversely associated with C-reactive protein levels in postmenopausal women at high risk for cardiovascular disease. Ann Epidemiol 2006;16:105–12. [PubMed: 16216530]

42. Selva DM, Hogeveen KN, Innis SM, Hammond GL. Monosaccharide-induced lipogenesis regulates the human hepatic sex hormone-binding globulin gene. J Clin Invest 2007;117:3979–87. [PubMed: 17992261]

43. Minelli C, Del Greco MF, van der Plaat DA, Bowden J, Sheehan NA, Thompson J. The use of two-sample methods for Mendelian randomization analyses on single large datasets. Int J Epidemiol 2021;50:1651–9. [PubMed: 33899104]

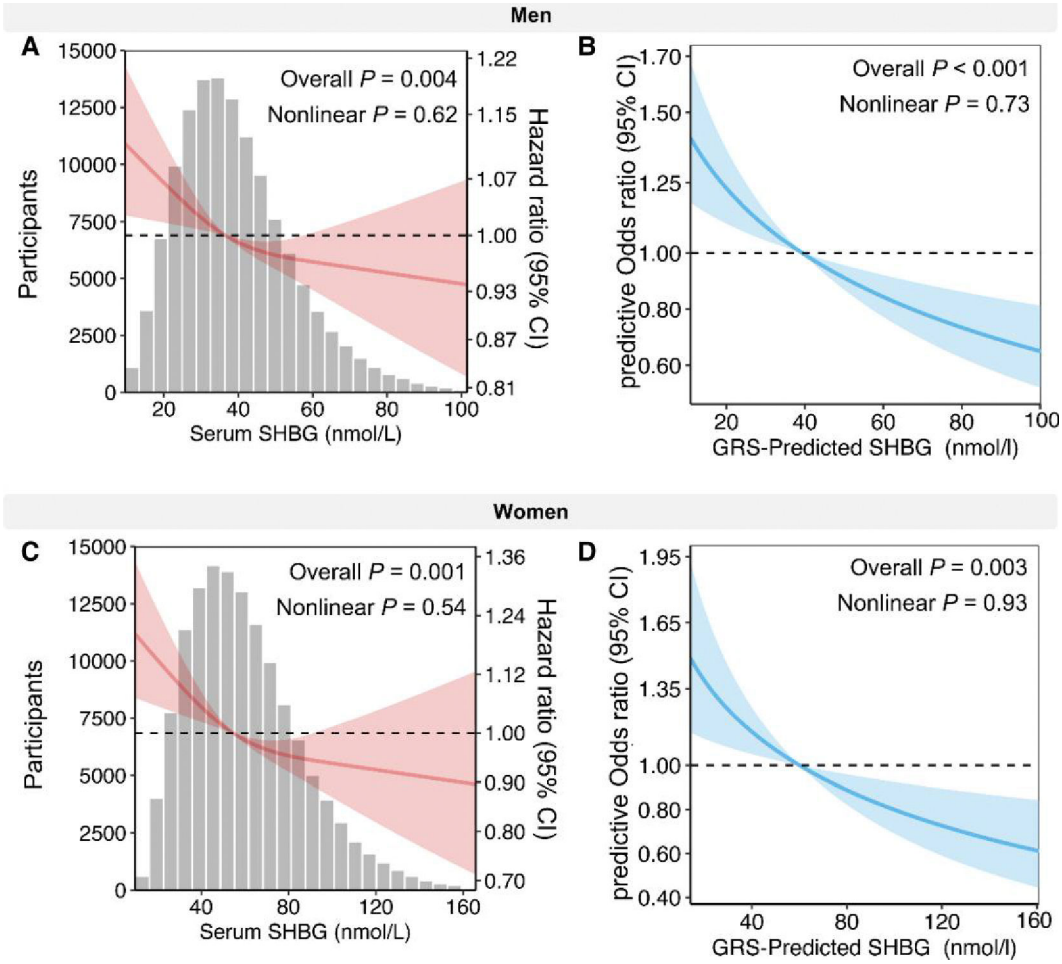

Fig. 1.

Conventional (A and C) and MR-based (B and D) nonlinear analysis of the associations of SHBG with risk of CHD among men and women in the UKB. In the conventional nonlinear analysis, we used Cox proportional hazard models and included a restricted cubic spline term for SHBG with 3 knots at the 10th, 50th, and 90th centiles, with adjustment for age, assessment centers, Townsend Deprivation Index, smoking, drinking, physical activity, and BMI. In nonlinear MR analysis, we divided men and women into deciles of the residual SHBG, which was calculated as the residual from a regression of SHBG on genetic risk score with adjustment for age, assessment centers, genotyping arrays, top 10 genetic principal components, and BMI. Then we generated a linear MR estimate in each decile, referred to as a localized average causal effect (LACE). Lastly, we fitted a fractional polynomial model to meta-regress these LACE estimates against the mean of SHBG in each decile. Nonlinearity was tested using the quadratic test.

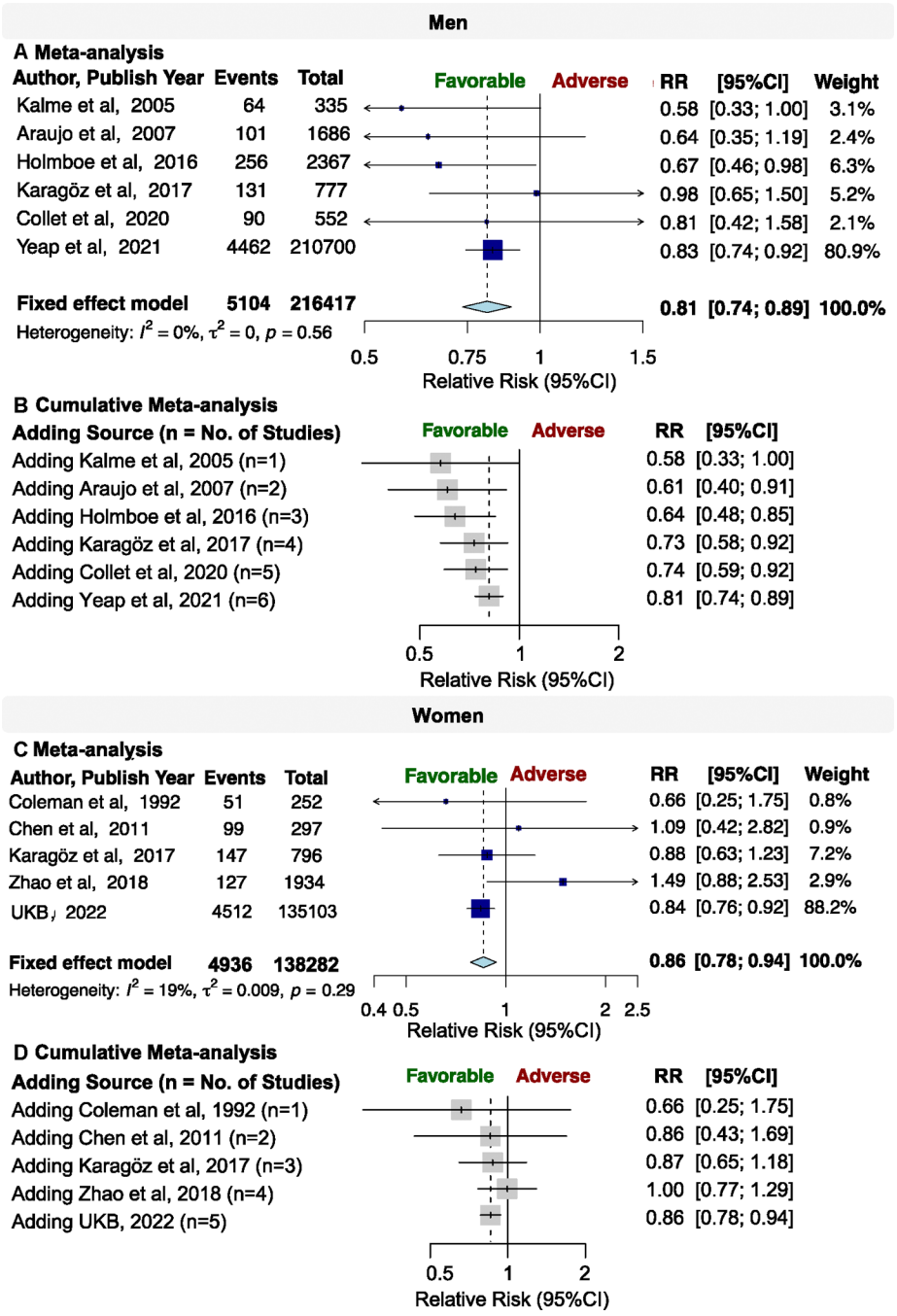

Fig. 2.

Pooled estimates of associations between SHBG and CHD risk in men and women.

Table 1.

Baseline characteristics of men and women according to serum levels of SHBG among UKB participants.

	Quintiles of serum levels of SHBG				
Characteristics	**Q1**	**Q2**	**Q3**	**Q4**	**Q5**
Men					
n	25683	25681	25 664	25650	25644
Follow-up year, y	11.8 ± 1.4	11.7 ± 1.5	11.7 ± 1.6	11.6 ± 1.6	11.5 ± 1.8
Age, y	52.5 ± 7.9	54.8±8.0	56.4±7.9	57.6±7.8	59.2±7.3
Townsend Deprivation Index	−1.51 ± 2.96	−1.65 ± 2.90	−1.68 ± 2.89	−1.63 ± 2.93	−1.46 ± 3.03
Family history of CVD, n (%)	9804 (38.2)	9809 (38.2)	9800 (38.2)	10023 (39.1)	9920 (38.7)
College degree, n (%)	8717 (33.9)	8801 (34.3)	8741 (34.1)	8713 (34.0)	8368 (32.6)
Smoking status, n (%)					
Never	13 784 (53.7)	13540 (52.7)	13179 (51.4)	12 828 (50.0)	12592 (49.1)
Previous	9511 (37.0)	9480 (36.9)	9650 (37.6)	9529 (37.2)	9020 (35.2)
Current	2388 (9.3)	2661 (10.4)	2835 (11.0)	3293 (12.8)	4032 (15.7)
Alcohol intake frequency, n (%)					
Never	946 (3.7)	918 (3.6)	1008 (3.9)	1116 (4.4)	1578 (6.2)
Special occasions only	1588 (6.2)	1437 (5.6)	1467 (5.7)	1498 (5.8)	1909 (7.5)
1 to 3 times a month	2464 (9.6)	2131 (8.3)	2098 (8.2)	2197 (8.6)	2215 (8.6)
Once or twice a week	7173 (27.9)	6871 (26.8)	6778 (26.4)	6606 (25.8)	6621 (25.8)
3 or 4 times a week	7048 (27.5)	7430 (28.9)	7352 (28.7)	7244 (28.3)	6682 (26.1)
Daily or almost daily	6450 (25.1)	6885 (26.8)	6946 (27.1)	6966 (27.2)	6615 (25.8)
Physical activity level, n (%)					
Insufficient	4920 (19.2)	4297 (16.7)	3848 (15.0)	3265 (12.7)	3036 (11.8)
Sufficient	14581 (56.8)	14660 (57.1)	14564 (56.7)	14569 (56.8)	14 246 (55.6)
Additional	6182 (24.1)	6724 (26.2)	7252 (28.3)	7816 (30.5)	8362 (32.6)
BMI, kg/m^2	29.5 ± 4.19	28.4 ± 3.99	27.7 ± 3.89	26.9 ± 3.78	25.9 ± 3.81
TT, nmol/L	9.10 (7.57, 10.8)	10.6 (9.01,12.4)	11.7 (9.94, 13.6)	12.9 (11.0, 14.9)	14.9 (12.8, 17.4)
Estradiol, pmol/L[a]	203 (188, 231)	204 (189, 234)	204 (189, 229)	204 (189, 230)	204 (190, 232)
SHBG, nmol/L	21.4 (18.0, 23.9)	29.9 (28.1, 31.6)	37.0 (35.2, 38.9)	45.4 (43.0, 48.1)	60.7 (55.3, 69.5)
Prevalence of diabetes, n (%)	1949 (7.6)	1342 (5.2)	1185 (4.6)	1050 (4.1)	1069 (4.2)

Characteristics	Quintiles of serum levels of SHBG				
	Q1	Q2	Q3	Q4	Q5
Prevalence of hypertension, n (%)	7729 (30.1)	7315 (28.5)	7246 (28.2)	6836 (26.7)	6248 (24.4)
Prevalence of hyperlipidemia, n (%)	3832 (14.9)	3377 (13.1)	3346 (13.0)	3008 (11.7)	2638 (10.3)
Women					
n	27 031	27 034	27 018	27 008	27 012
Follow-up year, y	11.8±1.4	11.8±1.3	11.8±1.3	11.8±1.3	11.8±1.3
Age, y	56.9 ± 7.32	57.0 ± 7.70	56.6 ± 7.89	56.0 ± 8.10	55.2 ± 8.34
Townsend Deprivation Index	−1.47 ± 2.93	−1.65 ± 2.84	−1.74±2.79	−1.76±2.79	−1.67 ± 2.84
Family history of CVD, n (%)	13147 (48.6)	12567 (46.5)	12106 (44.8)	11 653 (43.1)	11 306 (41.9)
College degree, n (%)	6815 (25.2)	7491 (27.7)	8350 (30.9)	8810 (32.6)	9368 (34.7)
Smoking status, n (%)					
Never	16 076 (59.5)	16 225 (60.0)	16410 (60.7)	16 726 (61.9)	17 005 (63.0)
Previous	9019 (33.4)	8624 (31.9)	8341 (30.9)	7948 (29.4)	7592 (28.1)
Current	1936 (7.2)	2185 (8.1)	2267 (8.4)	2334 (8.6)	2415 (8.9)
Alcohol intake frequency, n (%)					
Never	2239 (8.3)	1844 (6.8)	1744 (6.5)	1809 (6.7)	2283 (8.5)
Special occasions only	4387 (16.2)	3683 (13.6)	3407 (12.6)	3282 (12.2)	3620 (13.4)
1 to 3 times a month	3963 (14.7)	3523 (13.0)	3423 (12.7)	3321 (12.3)	3483 (12.9)
Once or twice a week	7089 (26.2)	7231 (26.8)	7300 (27.0)	7438 (27.6)	7383 (27.4)
3 or 4 times a week	5257 (19.5)	5953 (22.0)	6269 (23.2)	6355 (23.5)	6172 (22.9)
Daily or almost daily	4080 (15.1)	4785 (17.7)	4863 (18.0)	4784 (17.7)	4052 (15.0)
Physical activity level, n (%)					
Insufficient	4792 (17.7)	4098 (15.2)	3611 (13.4)	3313 (12.3)	3160 (11.7)
Sufficient	17 044 (63.1)	17 018 (63.0)	17146 (63.5)	16 831 (62.3)	16650 (61.6)
Additional	5195 (19.2)	5918 (21.9)	6261 (23.2)	6864 (25.4)	7202 (26.7)
BMI, kg/m^2	30.7 ± 5.52	28.2 ± 4.85	26.6 ± 4.34	25.3 ± 3.96	24.0 ± 3.57
TT, nmol/L	1.04 (0.74, 1.41)	1.04 (0.74, 1.41)	1.03 (0.74, 1.38)	1.02 (0.73, 1.37)	1.02 (0.73, 1.38)
Estradiol, pmol/L[a]	332 (231, 508)	368 (251, 568)	399 (270, 624)	430 (288, 673)	494 (322, 819)
SHBG, nmol/L	29.6 (24.2, 33.6)	43.6 (40.4, 46.6)	56.1 (52.9, 59.4)	70.8 (66.7, 75.5)	96.6 (87.5, 111)
Prevalence of diabetes, n (%)	2000 (7.4)	773 (2.9)	466 (1.7)	357 (1.3)	381 (1.4)
Prevalence of hypertension, n (%)	9474 (35.0)	7121 (26.3)	5701 (21.1)	4700 (17.4)	3743 (13.9)

Characteristics	Quintiles of serum levels of SHBG				
	Q1	Q2	Q3	Q4	Q5
Prevalence of hyperlipidemia, n (%)	4073 (15.1)	2724 (10.1)	2113 (7.8)	1642 (6.1)	1168 (4.3)

Continuous variables are described as mean±SD for those with normal distribution and median (IQR) for those with nonnormal distribution.

[a] In the UKB, only 10630 men and 29062 women had the levels of both SHBG and estradiol.

Abbreviations: Q, quintile.

Table 2.

Associations of serum SHBG levels with incident CHD among men and women followed for 12 years in the UKB.

	Quintiles of serum levels of SHBG					
	Q1	**Q2**	**Q3**	**Q4**	**Q5**	***P* for trend**
Men						
SHBG, nmol/L	21.4 (18.0, 23.9)	29.9 (28.1, 31.6)	37.0 (35.2, 38.9)	45.4 (43.0, 48.1)	60.7 (55.3, 69.5)	
Events/total (%)	1953/25683 (7.60)	2033/25681 (7.92)	2143/25664 (8.35)	2094/25650 (8.16)	2182/25644 (8.51)	
HR (95% Cl)						
Model 1	Ref.	0.91 (0.85, 0.97)	0.87 (0.82, 0.93)	0.78 (0.73, 0.83)	0.73 (0.69, 0.78)	<0.001
Model 2	Ref.	0.94 (0.88, 1.00)	0.92 (0.86, 0.98)	0.84 (0.79, 0.90)	0.81 (0.76, 0.87)	<0.001
Model 3	Ref.	0.97 (0.91, 1.04)	0.97 (0.91, 1.03)	0.91 (0.85, 0.97)	0.91 (0.85, 0.97)	0.001
Model 4	Ref.	0.96 (0.90, 1.02)	0.94 (0.88, 1.00)	0.88 (0.82, 0.94)	0.86 (0.79, 0.93)	<0.001
Women						
SHBG, nmol/L	29.6 (24.2, 33.6)	43.6 (40.4, 46.6)	56.1 (52.9, 59.4)	70.8 (66.7, 75.5)	96.6 (87.5, 111.3)	
Events/total (%)	1235/27 031 (4.57)	1028/27 034 (3.80)	845/27 018 (3.13)	741/27 008 (2.74)	663/27 012 (2.45)	
HR (95% Cl)						
Model 1	Ref.	0.83 (0.76, 0.90)	0.70 (0.65, 0.77)	0.64 (0.59, 0.70)	0.59 (0.54, 0.65)	<0.001
Model 2	Ref.	0.91 (0.83, 0.98)	0.81 (0.74, 0.88)	0.76 (0.69, 0.83)	0.72 (0.65, 0.79)	<0.001
Model 3	Ref.	0.95 (0.87, 1.03)	0.87 (0.80, 0.96)	0.84 (0.76, 0.93)	0.83 (0.75, 0.92)	<0.001
Model 4	Ref.	0.95 (0.86, 1.04)	0.89 (0.80, 0.98)	0.85 (0.76, 0.95)	0.84 (0.75, 0.94)	<0.001

Serum levels of SHBG were described as medium (IQR).

Model 1: Adjusted for age, assessment center, Townsend Deprivation Index, family history of cardiovascular diseases, smoking status, alcohol intake frequency, and physical activity level; Model 2: Model 1+prevalent diabetes, hypertension, or hyperlipidemia at baseline; Model 3: Model 2+BMI; Model 4: Model 3+TT.

Abbreviation: Q, quintile.

Table 3.

Conventional and MR-based analysis of associations between per unit of increase in SHBG level and CHD risk among men and women in the UKB.

	Men	Women
Event/total (%)	**10405/128 322 (8.11)**	**4512/135103 (3.34)**
Conventional analysis: HR (95% CI)		
Model 1	0.77 (0.73, 0.81)	0.68 (0.64, 0.72)
Model 2	0.84 (0.80, 0.88)	0.79 (0.74, 0.84)
Model 3	0.93 (0.88, 0.97)	0.88 (0.82, 0.94)
Model 4	0.88 (0.83, 0.94)	0.89 (0.83, 0.96)
MR-based analysis: pOR[a] (95% CI)		
Model 5	0.74 (0.62, 0.88)	0.73 (0.58, 0.92)
Model 6	0.75 (0.63, 0.89)	0.69 (0.54, 0.89)

In conventional analysis, we used Cox proportional hazard model to calculate HR for CHD risk per unit (log nmol/L) of increase in serum SHBG levels.

Model 1: Adjusted for age, assessment center, Townsend Deprivation Index, family history of cardiovascular diseases, smoking status, alcohol intake frequency, and physical activity level; Model 2: Model 1+prevalent diabetes, hypertension, or hyperlipidemia at baseline; Model 3: Model 2+BMI; Model 4: Model 3+TT [in linear MR analysis, predictive odds ratios for CHD risk per unit (log nmol/L) increase in genetically predicted SHBG were calculated by using a 2-stage least squares logistic regression model (24)]; Model 5: adjusted for age, BMI, assessment centers, genotyping arrays, and top 10 genetic principal components; Model 6: Model 5+TT.

[a]predicted odds ratio.