The causal role of circulating vitamin D concentrations in human complex traits and diseases: a large-scale Mendelian randomization study

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Abstract

Background: Vitamin D has been linked to a variety of diseases in observational studies. However, the causal role of vitamin D in human complex traits and diseases remains unclear and is under much debate. In this study, we conducted Mendelian randomization (MR) analyses using genetic variants associated with circulating 25-hydroxyvitamin D [25(OH)D] as instrumental variables to examine the causal role of vitamin D in human complex traits and diseases.

Methods: We performed MR analyses using genome-wide significant 25(OH)D-associated SNPs (N=58) from a vitamin D GWAS conducted in 329,247 unrelated UK Biobank (UKBB) participants of European ancestry, as well as 78 large-scale meta-GWASs of various outcomes to determine the causal role of 25(OH)D. In addition to inverse-variance weighted meta-analysis, we applied MR-Egger regression, MR-PRESSO and the weighted median approach to estimate the causal effect of 25(OH)D while examining and controlling for potential biases from horizontal pleiotropy. We conducted a sensitivity analysis by including additional genetic instruments (at a relaxed association threshold of $P<1\times10^{-6}$) from the UKBB GWAS (N=100). Since UKBB has sample overlap with some of the outcome GWASs, we complemented the main analyses by replicating our results using index SNPs (N=5) from an independent 25(OH)D GWAS conducted by the SUNLIGHT consortium.

Results: Consistent with previous findings, genetically predicted increased 25(OH)D levels significantly (after multiple testing correction) decreased the risk of multiple sclerosis (OR=0.963; 95%CI=0.945-0.981). We found limited evidence in support of a causal effect of 25(OH)D on anthropometric traits, obesity, cognitive function, sleep behavior, breast and prostate cancer, and autoimmune, cardiovascular, metabolic, neurological and psychiatric traits and diseases. Sensitivity analyses using additional instruments and complementary analyses using instruments from the SUNLIGHT consortium produced similar results to the main analyses.

Conclusions: Despite the largely augmented sample size and substantially improved statistical power of the largest-to-date vitamin D GWAS, our MR analyses did not convincingly support a causal effect of circulating 25(OH)D on complex traits and diseases examined here, except for multiple sclerosis. Our results may inform ongoing and future randomized clinical trials of

vitamin D supplementation. Future studies are warranted to prioritize the most promising target diseases for vitamin D intervention.

Keywords: Vitamin D; Two-sample Mendelian randomization; Causal effect; GWAS; Horizontal pleiotropy

Introduction

Vitamin D and its major circulating form, 25-hydroxyvitamin D [25(OH)D], is a fat soluble, steroid pre-hormone that plays an essential role in human health. Vitamin D deficiency has long been observed to be associated with rickets and osteomalacia.[1] In addition, a variety of common diseases such as cancer, autoimmune diseases, cardiovascular conditions and diabetes, have been reported to be linked with vitamin D in observational studies.[2–6] However, measurement error, confounding and reverse causality hinder a causal interpretation of these results from observational studies. Although the effect of circulating vitamin D on disease risk can be demonstrated by traditional randomized controlled trials (RCTs), large-scale RCTs of vitamin D supplementation are not readily available due to their high cost and long duration.[7, 8] A recently completed RCT VITAL (ClinicalTrials.gov: NCT01169259) involving a total of 25,871 participants with a median follow-up period of 5.3 years did not find lowered incidence of invasive cancer or cardiovascular events for those who took vitamin D_3 at a dose of 2,000 IU per day compared with the placebo group.[9] RCTs aiming at other diseases and conditions, such as depression (DepFuD; ClinicalTrials.gov: NCT02521012) and cognitive change (ClinicalTrials.gov: NCT03613116), are ongoing and have yet to reach conclusions. To better inform the field on the chance of success for such vitamin D trials, an improved understanding of the causal role of vitamin D in human complex diseases and traits, preferably through less costly observational studies, is necessary.

The genetic basis of circulating 25(OH)D has been investigated by several large-scale genome-wide association studies (GWASs), which have identified associations of *GC*, *NADSYN1/DHCR7*, *CYP2R1*, *CYP24A1*, *SEC23A*, *AMDHD1* with serum levels of vitamin D.[10, 11] The hitherto largest GWAS of circulating 25(OH)D is conducted using 329,247 unrelated UK Biobank (UKBB) participants of European ancestry and has revealed 59 independent loci (http://www.nealelab.is/uk-biobank). These genetic discoveries in 25(OH)D may help disentangle the causal relationships between vitamin D and other traits through Mendelian randomization (MR), an approach that uses genetic variants as instrumental variables (IVs) to assess the causal effect of an exposure on an outcome.[12] Under certain assumptions – that the IVs are strongly associated with the exposure, but not associated with any confounders of the exposure-outcome relationship, nor affect the outcome via other pathways than through

the exposure – an unbiased estimate of the causal effect can be estimated using the observed IV-exposure and IV-outcome associations.[13]

To date, a large number of MR studies examining the effect of circulating vitamin D on human complex diseases have been conducted. However, the causal role of vitamin D beyond its importance for bone health remains unclear and is under much debate. The only replicated causal effect of vitamin D from MR studies is that a higher level of vitamin D decreases the risk of adult/pediatric multiple sclerosis[14–16] and Alzheimer's disease;[17, 18] whereas findings for blood pressure, ovarian cancer, and all-cause mortality are yet to be replicated.[19–22] Overall, results from MR studies of vitamin D are largely null and do not support conventional observational studies, which have associated vitamin D with a variety of human diseases and traits. It is also worth noting that almost all hitherto MR studies for vitamin D were based on a small number of genetic instruments from 6 independent loci identified by the SUNLIGHT meta-GWAS published in 2010.[10] The recent vitamin D GWAS in the UKBB has identified 59 25(OH)D-associated genomic loci with more accurate effect size estimates, and provided an unprecedented opportunity to re-evaluate the causal effect of 25(OH)D with much improved genetic instruments. Moreover, the rapidly accumulating publicly available GWAS summary statistics for human complex diseases and traits has allowed us to assess the effect of vitamin D on a wide range of outcomes using MR.

Here, we conducted two-sample MR analyses (where IV-exposure and IV-outcome associations are estimated in two samples) to examine the effect of 25(OH)D on human complex traits and diseases using 78 GWAS outcomes. A total of 58 independent genetic variants associated with plasma 25(OH)D concentration were used as IVs. Summary statistics for the IV-exposure associations were extracted from the largest-to-date UKBB vitamin D GWAS. Summary statistics for the IV-outcome associations were extracted from 78 publicly available GWASs involving a total of 11,698,789 individuals (149,984 individuals on average per GWAS).

Methods

Data for IV-exposure

We extracted summary data for SNPs associated with circulating 25(OH)D concentration from a GWAS based on 329,247 unrelated individuals of European ancestry in the UK Biobank. The GWAS was conducted by the Neale lab and is publicly accessible (http://www.nealelab.is/uk-biobank). Specifically, a linear regression on circulating 25(OH)D (untransformed) was fitted to each SNP using an additive genetic model, controlling for age, sex, age^2, age by sex, age^2 by sex, and top 20 genetic principal components (Manhattan plot and QQ-plot are shown in **Supplementary Figures 1 and 2**). The LD score regression (LDSC)[23] estimated SNP-based heritability for vitamin D was 0.09 ($P=2.62\times10^{-9}$) and the LDSC intercept was 1.04 (SE=0.011), suggesting no population stratification in the GWAS. SNPs with minor allele frequency (MAF)>0.01, imputation INFO score>0.8 and Hardy-Weinberg equilibrium (HWE) $P>1\times10^{-10}$ were included. We performed LD-clumping to identify independent genome-wide significant SNPs as genetic instruments, using an r^2 threshold of 0.0001 with a clumping window size of 1Mb. We also excluded the HLA region (Chromosome 6, 25 to 35 Mb) due to the complex LD structure in the region. We excluded strand ambiguous (A/T and G/C) SNPs from the analysis to harmonize between exposure and outcome GWASs. We identified 58 genetic instruments with details presented in **Supplementary Table 1**. In addition to the 58 independent, genome-wide significant SNPs, we further performed sensitivity analysis using all LD-clumped SNPs associated with 25(OH)D concentration at $P<1\times10^{-6}$, and excluded strand ambiguous SNPs from this extended SNP set. This resulted in an additional 42 independent genetic instruments. Details of the 100 IVs are presented in **Supplementary Table 2**. We used these 2 sets of IVs (N=58 and N=100) to perform our *primary analysis*.

The UKBB Vitamin D GWAS is known to have sample overlap with some of our outcome GWASs (**Supplementary Table 3**). To overcome this limitation and for completeness, we performed additional MR analyses using an independent 25(OH)D GWAS conducted by the SUNLIGHT consortium in 2018 which involves 79,366 discovery samples and 42,757 replication samples of European ancestry. This is the second largest 25(OH)D GWAS to date, and has no sample overlap with neither the UKBB nor the outcome GWASs. Genome-wide analyses were performed within each cohort following a uniform plan. Specifically, linear

regression on natural-log transformed 25(OH)D was fitted to each SNP using an additive genetic model, and a fixed-effects inverse variance weighted meta-analysis across contributing cohorts was performed, with control for age, sex, body mass index, season of sample collection and population structure within each cohort, and consistent quality control thresholds across cohorts.[11] The SUNLIGHT consortium GWAS identified 6 genome-wide significant index SNPs. Since the SUNLIGHT and UKBB GWAS used slightly different 25(OH)D phenotypes (natural log transformed *vs.* untransformed level), instead of meta-analyzing the two cohorts, we performed a *complementary analysis* using 5 IVs (1 strand ambiguous SNP was excluded from MR analyses) from the SUNLIGHT consortium vitamin D GWAS (**Supplementary Table 4**).

Data for IV-outcome

We retrieved summary data for the associations between index SNPs and 78 complex traits and diseases from publicly available GWASs conducted mainly in European ancestry populations (3 GWASs were conducted in a mixed population including both European and Asian samples; **Supplementary Table 3**). These traits span a wide range of phenotypes including anthropometric traits, autoimmune inflammatory diseases, cancers, cardiovascular events, metabolomic traits, autoimmune diseases, and neurological and psychiatric disorders. For each trait, we retrieved the appropriate variant annotations (SNP rsID, chromosome, position, reference and alternate allele) and summary statistics (effect size, standard error, P-value, allele frequency [if available] and sample size of the study [if available]) for each index SNP. The sample sizes of these outcome GWASs ranged from 9,954 to 1,012,240.

Statistical analysis

MR uses SNPs as IVs to estimate the effects of risk factors on outcomes while controlling for potential confounding, by leveraging the random allocation of SNP alleles at conception. Three assumptions need to be satisfied to ensure a valid IV. The first is the relevance assumption, that is, IVs should be strongly associated with the exposure; the second assumption requires no association between IVs and confounders of the exposure-outcome relationship; and the third is the exclusion restriction assumption, requiring that genetic variants affect the outcome only through the exposure. If all MR assumptions are met, a causal effect can be estimated based on the observed IV-exposure and IV-outcome associations.

We conducted two-sample MR analyses to test for the causal relationship between circulating 25(OH)D and various traits. We applied a number of MR methods to estimate causal effects, including inverse-variance weighted (IVW) meta-analysis,[24] MR-PRESSO,[25] MR-Egger,[26] and a weighted median estimator approach.[27] While IVW meta-analysis does not account for potential horizontal pleiotropy, i.e., a genetic variant affects the outcome via a separate biological pathway from the exposure under investigation (violation of exclusion restriction assumption), MR-PRESSO, MR-Egger and weighted median approaches control potential horizontal pleiotropy under different model assumptions. In addition, we performed MR-Egger regression, modified Q test, modified Q' tests, and MR-PRESSO to detect estimation bias due to horizontal pleiotropy. MR-PRESSO implements a global test to evaluate the presence of horizontal pleiotropy, and an outlier test to detect specific SNP outliers.[25] In MR-Egger regression, significant difference of the intercept from zero suggests the existence of average directional horizontal pleiotropy.[26] Modified Q and Q' tests are traditionally used to identify over-dispersion and have been applied in the context of MR to detect outliers caused by horizontal pleiotropy.[28]

Results

As shown in **Table 1, Supplementary Table 5** and **Figure 1**, in the primary analysis where 58 index SNPs were used as IVs, vitamin D did not show causal effects on most of the outcomes examined here. The only significant association that survived Bonferroni correction for multiple testing was observed for multiple sclerosis (Immunochip scan conducted in 2013), where increased 25(OH)D levels decreased the risk of disease (IVW odds ratio [OR]=0.963, 95% confidence interval [CI]: 0.945-0.981; $P=1.04\times10^{-4}$). This result was consistent across different MR approaches (MR-Egger: 0.943 [0.920-0.966]; adjusted MR-PRESSO: 0.963 [0.945-0.981]; weighted median: 0.953 [0.935-0.973]) and was in line with previous findings. Using an earlier multiple sclerosis GWAS with a smaller sample size (conducted in 2011, 2/3 of the sample size of the 2013 MS Immunochip study), we observed a similar protected effect although with greater statistical uncertainties. We visualized the effect sizes of IV-exposure *vs.* IV-outcome for multiple sclerosis in a scatter plot with fitted slopes from the IVW meta-analysis, MR-Egger regression and weighted median estimator **(Figure 2).**

We further identified several other traits as potentially causally affected by 25(OH)D (at a nominal P-value threshold of 0.05) using MR-Egger or the adjusted MR-PRESSO approach, and without apparent signs of horizontal pleiotropy. These traits included triglycerides (β=−0.017, 95%CI: −0.030- −0.004), fasting insulin (β=0.003, 95%CI: 0.0001-0.005), estimated glomerular filtration rate (creatinine) (β=−0.001, 95%CI: −0.002- −0.0001), lupus (OR=1.028, 95%CI: 1.003-1.055), depressive symptoms (β=0.004, 95%CI: 0.001-0.008) and extraversion (β=0.005, 95%CI: 0.001-0.009). However, none of these suggestive causal pairs survived multiple testing corrections, and the results were often inconsistent across different MR approaches (**Table 1** and **Supplementary Table 5**). Therefore, these results should be interpreted with caution given the large number of statistical tests we performed. Further investigations are warranted to elucidate these suggestive findings.

Notably, we identified horizontal pleiotropy (as reflected by the MR-Egger intercept, MR-PRESSO global test and modified Q-test) for 65% of the trait pairs analyzed here (2 from MR-Egger regression, 51 from MR-PRESSO, 49 from the Modified Q test, and 46 from the Modified Q' test). Results for horizontal pleiotropic bias tests are presented in **Table 2**.

In addition, we performed a sensitivity analysis using an expanded set of UKBB IVs (100 independent SNPs associated with 25(OH)D levels at the P-value threshold of 1×10^{-6}, **Supplementary Table 2**), to increase the number of IVs in the MR analyses.[29, 30] As shown in **Supplementary Table 6** and **Supplementary Table 7**, the results did not alter substantially when incorporating additional IVs. Most of the outcomes were not influenced by genetically predicted 25(OH)D; and only multiple sclerosis showed a significant association with 25(OH)D which was consistent across MR approaches (IVW: OR=0.968, P=0.015; MR-Egger: OR=0.941, $P=8.04\times10^{-4}$; MR-PRESSO: OR=0.965, $P_{MR\text{-}PRESSO}=5.20\times10^{-5}$; weighted median: OR=0.953, $P=4.16\times10^{-7}$).

Results from our complementary analysis, where 5 25(OH)D-associated IVs from the SUNGLIGHT consortium were used, were consistent with the primary analyses. Specifically, increasing 25(OH)D levels also showed a strong effect on decreasing the risk of multiple sclerosis (Immunochip 2013). However, only 3 genetic instruments were available for the vitamin D-multiple sclerosis analysis, and the result was subject to potential pleiotropic bias. Several outcomes were identified to be causally associated with 25(OH)D at a nominally significant level ($P<0.05$) using MR-Egger (triglycerides, modified Stumvoll insulin sensitivity index, HbA1c, Crohn’s disease, anorexia) or the adjusted MR-PRESSO approach (oversleeper, age of menopause, Alzheimer's disease, HbA1c, height). However, none of these suggestive results survived multiple testing correction (**Supplementary Table 8** and **Supplementary Table 9**).

Discussion

In this study, we performed comprehensive two-sample MR analyses with 25(OH)D-associated SNPs and 78 large-scale meta-GWASs covering a wide spectrum of human complex traits and diseases to determine the causal role of 25(OH)D. In line with previously established findings, we have successfully replicated a significant causal relationship between 25(OH)D and multiple sclerosis. However, in general, our results, which were based on a markedly increased number of IVs (58 IVs from UKBB *vs.* 4 IVs used by most existing MR studies of 25(OH)D) and largely augmented samples of outcome GWASs, showed limited evidence for a causal effect of 25(OH)D on majority of the traits and diseases investigated here.

The causal role of 25(OH)D has been considered solid with multiple sclerosis, where three independent MR studies have consistently identified an association between genetically predicated 25(OH)D and a decreased risk of multiple sclerosis.[14–16] The successful replication of this causal relationship by our study acts as a positive control and has demonstrated the validity of our results. As a larger number of vitamin D IVs increases the power of MR analyses, we had the opportunity to obtain a more accurate estimate with narrower confidence intervals for the protective effect of 25(OH)D on multiple sclerosis, which appeared to be less strong in magnitude than previously estimated (5% decreased risk in our study *vs.* 15-20% decreased risk in previous studies).

Our null findings are in line with previous MR studies. An earlier MR which aggregated information from 21 adult cohorts with up to 42,024 participants to explore the causal relationship between vitamin D status and obesity, did not identify any effect of genetically instrumented 25(OH)D on BMI (P=0.57).[31] In our study, we expanded this outcome category by incorporating several adipose-related phenotypes in addition to BMI, such as body fat percentage, hip circumference, waist circumference, and waist-to-hip ratio (both BMI adjusted and unadjusted). Despite that we used the latest GWAS summary statistics of these outcomes with substantially augmented sample sizes, ranging from 76,137 in body fat percentage to 322,154 in BMI, we didn't find any evidence in support for a causal role of 25(OH)D in obesity-related traits (all P>0.50). Likewise, earlier MR studies have reported null findings for most of the autoimmune diseases including inflammatory bowel disease (Crohn's disease (P=0.67), ulcerative colitis (P=0.42)),[32] eczema (P=0.27),[33] lupus (P=0.79) and rheumatoid arthritis

(P=0.66).[34] Our results using summary statistics from greatly enlarged GWASs (for both exposure and outcomes) confirmed previous findings on a null relationship between 25(OH)D and autoimmune diseases. As for cardiovascular and metabolic traits, previous MR studies did not identify any causal role of 25(OH)D with coronary artery disease,[35] myocardial infarction,[36] total cholesterol levels including triglycerides and low-density lipoprotein (LDL) cholesterol,[37] type 2 diabetes,[38, 39] fasting insulin and adiponectin,[40] all of which, were confirmed by our analyses. There are, however, some discrepancies. For example, an earlier study that incorporated 31,435 individuals and used 4 index SNPs from two 25(OH)D-associated genes found no association between 25(OH)D-lowering alleles and either non-fasting remnant cholesterol or LDL-cholesterol, but identified a 50% decrease in 25(OH)D levels to be genetically associated with 6.0% (P=0.001) lower high-density lipoprotein (HDL) cholesterol levels.[37] The effect of 25(OH)D on HDL-cholesterol was no longer significant in our analysis by using the summary statistics of a more recent HDL GWAS involving 187,167 individuals (a 5-fold increase in sample size) and a greatly increased number of IVs. Similar null associations were observed in cancers where our MR did not convincingly support a causal effect of circulating 25(OH)D concentrations on the risk of breast or prostate cancer (P=0.58 and 0.46), consistent with previous findings (P=0.47 and 0.99). Finally, putative causal relationships between 25(OH)D and neurological or psychiatric traits have rarely been investigated. So far, only a few MR studies explored the role of 25(OH)D in cognitive function,[41] major depression,[42] and Alzheimer's disease;[17, 18] and nominally significant findings have only been reported in Alzheimer's disease but not the other two traits. It might worth noting that previous MR studies for Alzheimer's disease used either IV-exposure association estimates from a smaller 25(OH)D GWAS (SUNLIGHT consortium 2010)[10] or correlated IVs, which affected the validity of results. In our updated analysis, the causal relationship between 25(OH)D and Alzheimer's disease attenuated to null (P=0.63 using 2019 Alzheimer's GWAS and P=0.29 using 2013 Alzheimer's GWAS). Expanding the outcome list by incorporating additional neurological or psychiatric traits also did not identify any significant causal effects. These results are important in informing the ongoing RCT of vitamin D on depression (ClinicalTrials.gov: NCT02521012) and cognitive change (ClinicalTrials.gov: NCT03613116).

To ensure the validity of MR results, several important assumptions need to be satisfied. First of all, the relevance assumption, that IVs should be strongly associated with the exposure, is

naturally guaranteed by selecting independent 25(OH)D-associated SNPs with genome-wide significance as IVs. We also confirmed that there is no inflation of GWAS associations due to population stratification by LDSC (intercept = 1.04 which indicates minimal confounding). This assumption could also be reassured by the negligible associations between 25(OH)D and various traits and diseases from the current and past MR studies, many of which act as potential confounders for other outcomes (e.g., obesity-related traits). Sample overlap is a potential concern for our MR analyses using genetic instruments derived from the UK Biobank, given that many recent large-scale outcome meta-GWASs include UK Biobank samples (as shown in **Supplementary Table 3**). Two-sample MR analysis assumes no sample overlap between exposure and outcome GWASs, and the expected covariance of sampling error between the genetic effects on the exposure and the outcome is assumed to be zero. Violating these assumptions (i.e., sample overlap) in two-sample MR analyses will lead to inflated type I error for hypothesis testing on the causal effect due to ignoring non-zero covariance of sampling error.[43] However, as majority of our MR results were null, the potentially inflated type I error (false positives) does not change the interpretation of our results. Finally, the exclusion restriction assumption requires IVs to affect the outcome only through the exposure (no horizontal pleiotropy). We employed a number of methods to test and control for horizontal pleiotropy. While MR-Egger regression is designed to detect average pleiotropic bias, the MR-PRESSO global test and modified Q and Q' tests can detect bias caused by individual outlier SNPs with horizontal pleiotropy. While 65% of the trait pairs analyzed here show significant pleiotropy, we presented MR causal estimates adjusted for pleiotropic bias using 3 different methods. Notably, the significant causal effect of vitamin D on multiple sclerosis is also subject to potential pleiotropic bias; however, the causal effect estimates are consistently significant using different bias adjusted MR methods.

In summary, our MR analysis showed limited evidence of causal effects of vitamin D on a wide range of human complex traits and diseases, except for multiple sclerosis. Our results may inform future RCTs and help to prioritize the most promising target diseases for vitamin D intervention.

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Author contributions

X.J. and C-Y.C. contributed to study conception, data analysis, interpretation of the results and drafting of the manuscript. T.G. contributed to interpretation of the results and critical revision of the manuscript.

Competing interests

The authors declare no competing interests.

Supplementary information

Supplementary Figure 1. Genome-wide association of circulating 25-hydroxyvitamin D graphed by chromosome positions and –log10 P-value (Manhattan plot).

Supplementary Figure 2. Quantile-quantile plot of all SNPs from the genome-wide association analysis of circulating 25-hydroxyvitamin D (QQ-plot).

Supplementary Table 1. List of genetic instrumental variables from UK Biobank vitamin D GWAS (genome-wide significant SNPs; N=58).

Supplementary Table 2. List of genetic instrumental variables from UK Biobank vitamin D GWAS (association $P<1\times10^{-6}$; N =100).

Supplementary Table 3. References for publicly available GWAS summary statistics used in this study.

Supplementary Table 4. List of genetic instrumental variables from the SUNLIGHT consortium vitamin D GWAS.

Supplementary Table 5. Causal effect estimates converted to odds ratio for binary traits (genome-wide significant SNPs; N=58).

Supplementary Table 6. Causal effect estimates of 25(OH)D with complex traits and diseases using SNPs associated with 25(OH)D at $P<1\times10^{-6}$ from UK Biobank (N=100).

Supplementary Table 7. Test for potential biases in MR analysis using SNPs associated with 25(OH)D at $P<1\times10^{-6}$ from UK Biobank (N=100).

Supplementary Table 8. Causal effect estimates of 25(OH)D with complex traits and diseases using genome-wide significant SNPs of 25(OH)D from the SUNLIGHT GWAS (N=5).

Supplementary Table 9. Test for potential biases in MR analyses using genome-wide significant SNPs for 25(OH)D from the SUNLIGHT GWAS (N=5).

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Figure 1. Estimated causal effects of 25(OH)D on various traits and chronic diseases based on MR-PRESSO approach using genome-wide significant SNPs of 25(OH)D.

Figure 2. Scatter plots of effect sizes of IV-exposure *vs.* IV-outcome for multiple sclerosis.

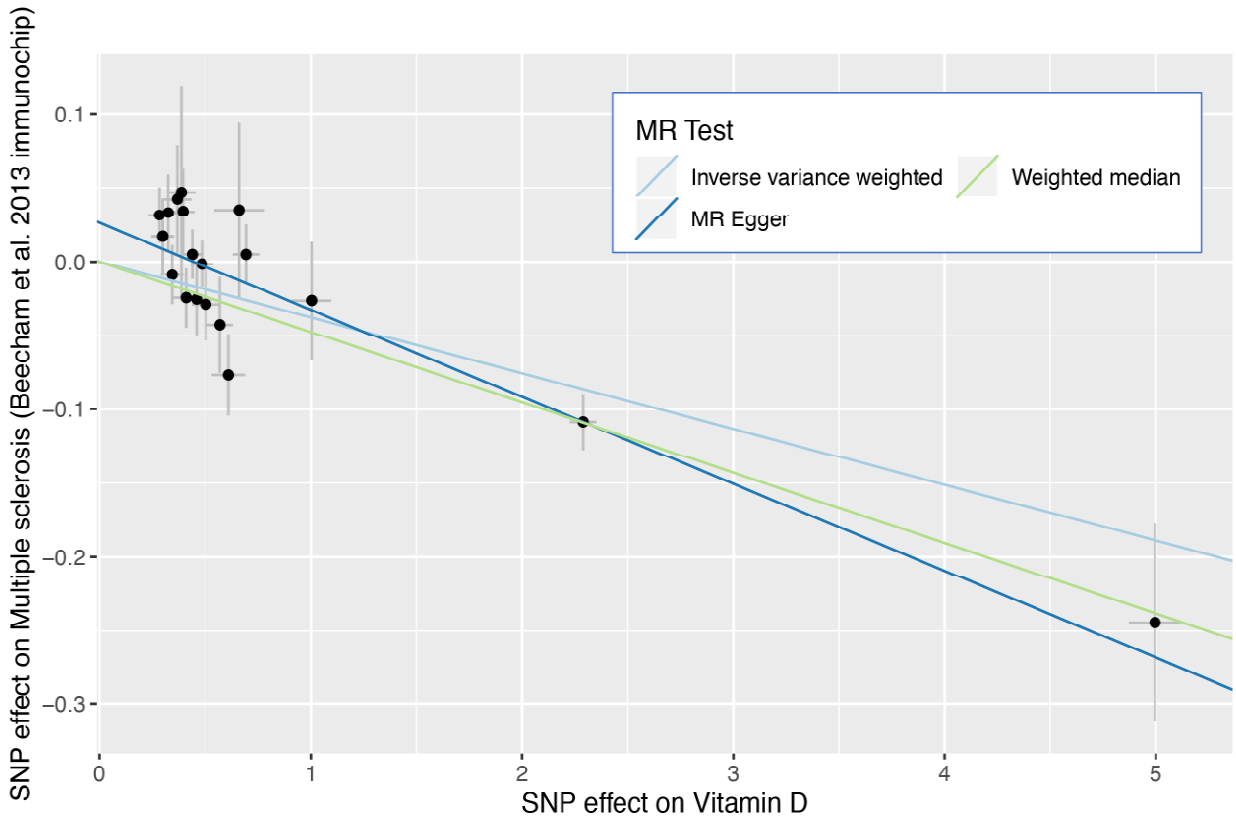

Table 1. Causal effect estimates of 25(OH)D with complex traits and diseases using genome-wide significant SNPs of 25(OH)D (genome-wide significant SNPs; N=58).

Outcome GWAS	Group	Random effect IVW meta-analysis			MR-Egger			MR-PRESSO adjusted			Median estimate		
		BETA	95% CI	P	BETA	95% CI	P	BETA	95% CI	P	BETA	95% CI	P
Extreme BMI	Anthropometric	0	(-0.033 - 0.034)	1	-0.005	(-0.057 - 0.046)	0.85	0.0001	(-0.033 - 0.034)	1	-0.0004	(-0.025 - 0.024)	0.98
Extreme waist-to-hip ratio	Anthropometric	-0.001	(-0.028 - 0.027)	0.95	0.003	(-0.040 - 0.045)	0.9	-0.001	(-0.028 - 0.027)	0.95	0.001	(-0.025 - 0.027)	0.92
Obesity class 1	Anthropometric	0.002	(-0.015 - 0.018)	0.83	0.003	(-0.022 - 0.029)	0.79	0.006	(-0.006 - 0.019)	0.33	0.002	(-0.010 - 0.014)	0.75
Obesity class 2	Anthropometric	-0.009	(-0.035 - 0.017)	0.49	-0.011	(-0.051 - 0.028)	0.58	-0.002	(-0.019 - 0.016)	0.85	-0.009	(-0.027 - 0.008)	0.3
Obesity class 3	Anthropometric	0.008	(-0.019 - 0.035)	0.56	-0.009	(-0.048 - 0.030)	0.65	0.008	(-0.019 - 0.035)	0.56	0.005	(-0.029 - 0.039)	0.77
Overweight	Anthropometric	-0.003	(-0.015 - 0.009)	0.61	-0.001	(-0.019 - 0.017)	0.89	0.0001	(-0.008 - 0.008)	0.98	-0.003	(-0.011 - 0.006)	0.54
Heart rate	Anthropometric	0.003	(-0.040 - 0.047)	0.87	0.007	(-0.059 - 0.073)	0.83	0.015	(-0.014 - 0.044)	0.3	0.004	(-0.043 - 0.052)	0.86
BMI	Anthropometric	0.001	(-0.006 - 0.008)	0.83	-0.001	(-0.011 - 0.010)	0.92	0.002	(-0.002 - 0.006)	0.35	-0.0003	(-0.004 - 0.003)	0.86
Body fat percentage	Anthropometric	0.004	(-0.002 - 0.009)	0.19	-0.0004	(-0.009 - 0.008)	0.93	0.003	(-0.002 - 0.008)	0.23	0.001	(-0.004 - 0.006)	0.58
Hip circumference	Anthropometric	0.001	(-0.006 - 0.008)	0.7	0.0002	(-0.011 - 0.011)	0.97	0.001	(-0.004 - 0.005)	0.76	-0.00001	(-0.004 - 0.004)	1
Hip circumference adjusted for BMI	Anthropometric	0.002	(-0.002 - 0.006)	0.39	0.002	(-0.004 - 0.008)	0.52	0.002	(-0.002 - 0.006)	0.39	0.002	(-0.002 - 0.006)	0.35
Waist circumference	Anthropometric	0.001	(-0.006 - 0.008)	0.72	0.0003	(-0.011 - 0.011)	0.96	0.003	(-0.002 - 0.008)	0.21	0.001	(-0.003 - 0.005)	0.62
Waist circumference adjusted for BMI	Anthropometric	0.001	(-0.003 - 0.004)	0.61	0.001	(-0.005 - 0.006)	0.78	0.001	(-0.003 - 0.004)	0.61	0.0004	(-0.003 - 0.004)	0.84
Waist-to-hip ratio	Anthropometric	0	(-0.005 - 0.005)	0.96	-0.0001	(-0.007 - 0.007)	0.98	0.0001	(-0.005 - 0.005)	0.96	-0.001	(-0.004 - 0.003)	0.68
Waist-to-hip ratio adjusted for BMI	Anthropometric	-0.001	(-0.005 - 0.004)	0.77	-0.0001	(-0.007 - 0.006)	0.97	-0.001	(-0.005 - 0.004)	0.77	-0.001	(-0.005 - 0.003)	0.53
Life span*	Anthropometric	0	(-0.003 - 0.004)	0.92	-0.001	(-0.006 - 0.005)	0.84	-0.001	(-0.003 - 0.002)	0.62	-0.002	(-0.005 - 0.0004)	0.1
Height	Anthropometric	0.003	(-0.004 - 0.009)	0.41	0.002	(-0.008 - 0.012)	0.67	0.002	(-0.002 - 0.006)	0.28	0.002	(-0.001 - 0.005)	0.14
Breast cancer	Cancer	0.001	(-0.003 - 0.005)	0.58	0.002	(-0.004 - 0.007)	0.61	0.001	(-0.002 - 0.004)	0.62	-0.00003	(-0.004 - 0.004)	0.99
Prostate cancer	Cancer	-0.002	(-0.007 - 0.003)	0.46	-0.004	(-0.011 - 0.004)	0.35	-0.003	(-0.007 - 0.002)	0.22	-0.003	(-0.009 - 0.002)	0.23
SOFT CAD (Nelson CP et al. 2017)*	Cardiovascular	-0.006	(-0.016 - 0.004)	0.25	0.002	(-0.013 - 0.017)	0.77	-0.002	(-0.007 - 0.003)	0.45	-0.001	(-0.006 - 0.005)	0.78
Coronary artery disease (cross-population)	Cardiovascular	-0.009	(-0.021 - 0.004)	0.17	-0.002	(-0.022 - 0.017)	0.82	-0.004	(-0.011 - 0.002)	0.16	-0.001	(-0.008 - 0.006)	0.82
Coronary artery disease (European only)	Cardiovascular	-0.0001	(-0.025 - 0.025)	1	0.005	(-0.033 - 0.043)	0.8	0.006	(-0.008 - 0.020)	0.37	0.004	(-0.008 - 0.017)	0.53
Myocardial infarction	Cardiovascular	-0.008	(-0.019 - 0.003)	0.17	-0.004	(-0.022 - 0.014)	0.67	-0.004	(-0.011 - 0.003)	0.25	-0.003	(-0.011 - 0.004)	0.4
Lipid - High-density lipoprotein cholesterol	Cardiovascular	-0.012	(-0.056 - 0.032)	0.59	0.001	(-0.071 - 0.073)	0.98	-0.007	(-0.022 - 0.008)	0.34	0.006	(0.001 - 0.011)	0.02
Lipid - Low-density lipoprotein cholesterol	Cardiovascular	-0.037	(-0.103 - 0.029)	0.27	0.009	(-0.096 - 0.113)	0.87	-0.023	(-0.048 - 0.001)	0.06	0.004	(-0.001 - 0.010)	0.12
Lipid - Total cholesterol	Cardiovascular	-0.046	(-0.098 - 0.005)	0.08	0.01	(-0.066 - 0.086)	0.79	-0.02	(-0.044 - 0.005)	0.11	0.004	(-0.004 - 0.012)	0.34
Lipid - Triglycerides	Cardiovascular	-0.026	(-0.062 - 0.009)	0.15	0.002	(-0.053 - 0.057)	0.94	-0.017	(-0.030 - -0.004)	0.01	-0.004	(-0.009 - 0.001)	0.11
Adiponectin	Metabolic	0.003	(-0.002 - 0.008)	0.21	0.003	(-0.004 - 0.010)	0.43	0.003	(-0.002 - 0.008)	0.21	0.003	(-0.001 - 0.008)	0.13
Fasting insulin	Metabolic	0.004	(-0.001 - 0.010)	0.12	0.001	(-0.007 - 0.008)	0.85	0.003	(0.0001 - 0.005)	0.04	0.003	(-0.001 - 0.006)	0.16
Estimated glomerular filtration rate (creatinine)	Metabolic	-0.002	(-0.004 - 0.0005)	0.14	-0.001	(-0.004 - 0.002)	0.71	-0.001	(-0.002 - -0.0001)	0.02	-0.001	(-0.002 - -0.0002)	0.01
Type-2 diabetes	Metabolic	-0.001	(-0.009 - 0.007)	0.87	0.006	(-0.006 - 0.018)	0.32	-0.001	(-0.008 - 0.006)	0.77	0.004	(-0.004 - 0.011)	0.38
Modified Stumvoll Insulin Sensitivity Index (ISI)	Metabolic	-0.004	(-0.012 - 0.004)	0.3	0.004	(-0.007 - 0.015)	0.48	-0.004	(-0.012 - 0.004)	0.3	0.001	(-0.008 - 0.010)	0.83
HbA1c	Metabolic	0.001	(-0.0001 - 0.003)	0.08	0.001	(-0.002 - 0.003)	0.52	0.001	(-0.0001 - 0.002)	0.08	0.001	(-0.001 - 0.003)	0.3
Lupus	Autoimmune	0.013	(-0.004 - 0.030)	0.14	0.028	(0.003 - 0.053)	0.03	0.013	(-0.004 - 0.030)	0.14	0.026	(0.003 - 0.050)	0.03
Primary biliary cirrhosis	Autoimmune	0.033	(-0.026 - 0.092)	0.28	0.02	(-0.067 - 0.108)	0.65	0.03	(-0.009 - 0.069)	0.13	0.024	(-0.010 - 0.059)	0.16
C-reactive protein	Autoimmune	-0.002	(-0.008 - 0.004)	0.53	0.001	(-0.007 - 0.010)	0.76	-0.001	(-0.004 - 0.002)	0.67	-0.001	(-0.004 - 0.002)	0.38
Crohn's disease	Autoimmune	-0.008	(-0.025 - 0.009)	0.35	-0.002	(-0.028 - 0.023)	0.87	-0.008	(-0.020 - 0.003)	0.16	-0.001	(-0.016 - 0.014)	0.88
Inflammatory bowel disease	Autoimmune	-0.006	(-0.018 - 0.005)	0.28	-0.0004	(-0.017 - 0.017)	0.96	-0.003	(-0.012 - 0.005)	0.43	-0.003	(-0.014 - 0.009)	0.65
Ulcerative colitis	Autoimmune	-0.006	(-0.017 - 0.005)	0.27	0.001	(-0.014 - 0.017)	0.86	-0.003	(-0.011 - 0.004)	0.42	-0.001	(-0.015 - 0.013)	0.89
Rheumatoid arthritis	Autoimmune	-0.002	(-0.012 - 0.008)	0.69	-0.004	(-0.019 - 0.012)	0.63	-0.004	(-0.012 - 0.005)	0.39	-0.003	(-0.017 - 0.011)	0.65
Eczema	Autoimmune	0.006	(-0.007 - 0.019)	0.39	0.0003	(-0.019 - 0.020)	0.98	0.0002	(-0.008 - 0.008)	0.96	-0.000003	(-0.012 - 0.012)	1
Cognitive performance (SSGAC 2018)*	Cognition	-0.001	(-0.004 - 0.001)	0.29	0.001	(-0.003 - 0.004)	0.74	-0.001	(-0.003 - 0.001)	0.18	-0.001	(-0.003 - 0.001)	0.55
Educational attainment (SSGAC 2018)*	Cognition	-0.001	(-0.003 - 0.001)	0.52	0.001	(-0.002 - 0.004)	0.69	-0.001	(-0.002 - 0.001)	0.34	-0.001	(-0.002 - 0.001)	0.37
Years of schooling (SSGAC 2016)*	Cognition	-0.0004	(-0.002 - 0.002)	0.7	0.001	(-0.002 - 0.004)	0.66	-0.0003	(-0.002 - 0.001)	0.74	0	(-0.002 - 0.002)	1
Fluid intelligence*	Cognition	-0.001	(-0.005 - 0.002)	0.51	0.001	(-0.004 - 0.006)	0.59	-0.0004	(-0.003 - 0.003)	0.78	-0.001	(-0.005 - 0.003)	0.69
Age-related hearing impairment	Neurological	-0.0001	(-0.010 - 0.009)	0.98	-0.005	(-0.019 - 0.009)	0.49	-0.0001	(-0.010 - 0.009)	0.98	-0.009	(-0.022 - 0.004)	0.19
Alzheimer's disease (IGAP 2019)	Neurological	-0.004	(-0.018 - 0.011)	0.63	-0.004	(-0.027 - 0.018)	0.69	-0.0003	(-0.007 - 0.007)	0.94	-0.005	(-0.014 - 0.005)	0.33
Alzheimer's disease (IGAP 2013)	Neurological	-0.007	(-0.021 - 0.006)	0.29	-0.012	(-0.032 - 0.009)	0.28	-0.005	(-0.014 - 0.004)	0.26	-0.01	(-0.021 - 0.001)	0.07
Any ischemic stroke	Neurological	0.002	(-0.003 - 0.008)	0.4	0.005	(-0.003 - 0.014)	0.24	0.003	(-0.002 - 0.009)	0.21	0.004	(-0.003 - 0.010)	0.3
Any stroke	Neurological	0.002	(-0.003 - 0.008)	0.42	0.003	(-0.005 - 0.011)	0.41	0.003	(-0.002 - 0.008)	0.22	0.001	(-0.005 - 0.008)	0.73
Cardioembolic stroke	Neurological	0.002	(-0.007 - 0.011)	0.67	0.008	(-0.005 - 0.021)	0.25	0.002	(-0.007 - 0.011)	0.67	0.005	(-0.008 - 0.017)	0.48
Large artery stroke	Neurological	0.00001	(-0.013 - 0.013)	1	0.003	(-0.016 - 0.023)	0.74	0.00001	(-0.013 - 0.013)	1	0.001	(-0.015 - 0.018)	0.87
Small vessel stroke	Neurological	0.0003	(-0.012 - 0.013)	0.97	0.008	(-0.011 - 0.026)	0.41	0.0003	(-0.012 - 0.013)	0.97	0.003	(-0.013 - 0.020)	0.71
Parkinson's disease	Neurological	-0.003	(-0.016 - 0.010)	0.64	-0.0003	(-0.021 - 0.021)	0.97	-0.003	(-0.016 - 0.010)	0.64	-0.006	(-0.022 - 0.011)	0.48
Amyotrophic lateral sclerosis	Neurological	-0.003	(-0.009 - 0.002)	0.25	-0.003	(-0.011 - 0.005)	0.45	-0.003	(-0.009 - 0.002)	0.25	0.002	(-0.008 - 0.012)	0.72
Multiple sclerosis (Beecham et al. 2013 immunochip)	**Neurological**	**-0.038**	**(-0.057 - -0.019)**	**1.04E-04**	**-0.059**	**(-0.083 - -0.034)**	**2.29E-06**	**-0.038**	**(-0.057 - -0.019)**	**1.04E-04**	**-0.048**	**(-0.068 - -0.028)**	**2.59E-06**
Multiple sclerosis (Sawcer et al. 2011 GWAS)	Neurological	-0.006	(-0.017 - 0.005)	0.26	-0.02	(-0.035 - -0.004)	0.01	-0.006	(-0.017 - 0.005)	0.26	-0.014	(-0.026 - -0.001)	0.03
Bipolar disorder (PGC 2018)	Psychiatric	-0.001	(-0.010 - 0.009)	0.91	0.008	(-0.005 - 0.022)	0.24	0.003	(-0.004 - 0.010)	0.47	0.003	(-0.006 - 0.013)	0.46
Bipolar disorder (PGC+ICCBD 2017)	Psychiatric	0.002	(-0.008 - 0.012)	0.68	0.01	(-0.004 - 0.025)	0.16	0.002	(-0.005 - 0.010)	0.55	0.006	(-0.004 - 0.017)	0.25
Anorexia	Psychiatric	-0.008	(-0.022 - 0.006)	0.27	-0.005	(-0.026 - 0.015)	0.61	-0.008	(-0.022 - 0.006)	0.27	-0.008	(-0.030 - 0.014)	0.47

Posttraumatic stress disorder (PGC 2017)	Psychiatric	-0.009	(-0.029 - 0.010)	0.36	0.005	(-0.024 - 0.033)	0.74	-0.009	(-0.029 - 0.010)	0.36	-0.008	(-0.036 - 0.021)	0.59
Depressive symptoms*	Psychiatric	0.002	(-0.0002 - 0.004)	0.07	0.004	(0.001 - 0.008)	0.01	0.0004	(-0.002 - 0.003)	0.73	0.001	(-0.002 - 0.004)	0.37
Subjective well-being	Psychiatric	0	(-0.004 - 0.003)	0.81	0.001	(-0.005 - 0.007)	0.77	-0.001	(-0.005 - 0.002)	0.44	0.0004	(-0.003 - 0.004)	0.82
Neuroticism	Psychiatric	0.001	(-0.001 - 0.003)	0.36	0.0002	(-0.003 - 0.003)	0.89	0.001	(-0.001 - 0.003)	0.36	0.001	(-0.001 - 0.003)	0.45
Extraversion	Psychiatric	0.002	(-0.0005 - 0.005)	0.1	0.005	(0.001 - 0.009)	0.02	0.002	(-0.0005 - 0.005)	0.1	0.004	(-0.0003 - 0.008)	0.07
Loneliness	Psychiatric	0.005	(-0.011 - 0.020)	0.55	0.001	(-0.022 - 0.024)	0.95	0.005	(-0.011 - 0.020)	0.55	0.008	(-0.014 - 0.029)	0.5
Generalized Anxiety disorder (case-control)	Psychiatric	-0.000002	(-0.015 - 0.015)	1	0.003	(-0.021 - 0.027)	0.81	-0.000002	(-0.015 - 0.015)	1	-0.00004	(-0.022 - 0.021)	1
Generalized Anxiety disorder (factor score)	Psychiatric	-0.001	(-0.003 - 0.002)	0.64	-0.001	(-0.005 - 0.003)	0.61	-0.001	(-0.003 - 0.002)	0.64	-0.0002	(-0.004 - 0.004)	0.91
Schizophrenia (PGC 2014)	Psychiatric	-0.001	(-0.009 - 0.007)	0.72	0.003	(-0.008 - 0.015)	0.58	-0.001	(-0.006 - 0.005)	0.82	0.001	(-0.007 - 0.008)	0.86
Major depressive disorder (PGC 2013)	Psychiatric	0.016	(-0.008 - 0.040)	0.19	-0.005	(-0.034 - 0.024)	0.75	0.012	(-0.006 - 0.030)	0.18	0.008	(-0.013 - 0.028)	0.47
Major depressive disorder (PGC 2018)*	Psychiatric	-0.001	(-0.005 - 0.003)	0.76	0.001	(-0.005 - 0.007)	0.72	-0.001	(-0.005 - 0.003)	0.61	0.002	(-0.003 - 0.007)	0.35
Age of menopause	Reproductive	-0.017	(-0.046 - 0.011)	0.22	-0.01	(-0.051 - 0.032)	0.65	-0.01	(-0.026 - 0.006)	0.23	-0.013	(-0.030 - 0.005)	0.15
Age of menarche	Reproductive	-0.003	(-0.008 - 0.003)	0.39	0.001	(-0.007 - 0.010)	0.77	-0.003	(-0.008 - 0.002)	0.19	0.00002	(-0.005 - 0.005)	0.99
Insomnia complaints*	Sleep	-0.0001	(-0.004 - 0.004)	0.97	0.0003	(-0.006 - 0.007)	0.93	-0.0001	(-0.004 - 0.004)	0.97	-0.001	(-0.007 - 0.005)	0.75
Chronotype*	Sleep	-0.0002	(-0.002 - 0.001)	0.82	-0.001	(-0.004 - 0.001)	0.31	-0.0002	(-0.002 - 0.001)	0.82	-0.002	(-0.004 - 0.001)	0.19
Oversleeper*	Sleep	0.0002	(-0.0003 - 0.001)	0.49	0.0001	(-0.001 - 0.001)	0.68	0.0002	(-0.0003 - 0.001)	0.49	0.0001	(-0.001 - 0.001)	0.89
Sleep duration*	Sleep	0.001	(-0.0003 - 0.003)	0.11	-0.0003	(-0.003 - 0.002)	0.82	0.001	(-0.0003 - 0.003)	0.11	0.001	(-0.002 - 0.003)	0.49
Undersleeper*	Sleep	-0.001	(-0.002 - 0.00004)	0.06	0.0001	(-0.001 - 0.001)	0.91	-0.001	(-0.002 - 0.00004)	0.06	-0.001	(-0.002 - 0.0003)	0.18

Significant result after Bonferroni correction (0.05/78=0.00064) is bolded. *Outcome GWAS has sample overlapped with the vitamin D GWAS in UK Biobank, which may lead to inflated type 1 error and causal effect estimates (ref. Burgess, S, Davies NM, and Thompson SG. 2016. Genet Epidemiol).

Table 2. Test for potential biases in MR analysis using genome-wide significant SNPs for 25(OH)D (genome-wide significant SNPs; N=58).

Outcome GWAS	Group	MR-Egger intercept test p-value	MR-PRESSO global test p-value	# outlier	Modified Q test p-value	Modified Q' test p-value
Extreme BMI	Anthropometric	0.79	0.01	0	5.18E-04	5.56E-04
Extreme waist-to-hip ratio	Anthropometric	0.82	0.20	0	0.10	0.10
Obesity class 1	Anthropometric	0.87	0.00	2	3.72E-05	3.84E-05
Obesity class 2	Anthropometric	0.88	0.01	2	1.10E-04	1.14E-04
Obesity class 3	Anthropometric	0.25	0.50	0	0.58	0.67
Overweight	Anthropometric	0.79	0.00	2	3.21E-05	3.47E-05
Heart rate	Anthropometric	0.88	0.35	1	0.24	0.24
BMI	Anthropometric	0.76	<0.001	3	5.51E-16	7.23E-16
Body fat percentage	Anthropometric	0.21	0.01	1	0.01	0.02
Hip circumference	Anthropometric	0.78	<0.001	3	7.23E-11	8.41E-11
Hip circumference adjusted for BMI	Anthropometric	0.90	0.19	0	0.11	0.11
Waist circumference	Anthropometric	0.81	<0.001	2	3.84E-11	4.30E-11
Waist circumference adjusted for BMI	Anthropometric	0.95	0.27	0	0.27	0.27
Waist-to-hip ratio	Anthropometric	0.95	0.01	0	1.39E-03	1.40E-03
Waist-to-hip ratio adjusted for BMI	Anthropometric	0.85	0.06	0	0.02	0.02
life span	Anthropometric	0.71	<0.001	5	1.10E-26	1.43E-26
Height	Anthropometric	0.88	<0.001	5	7.38E-20	7.67E-20
Breast cancer	Cancer	0.86	<0.001	3	7.05E-07	7.22E-07
Prostate cancer	Cancer	0.54	0.00	2	8.80E-04	1.06E-03
SOFT CAD (Nelson CP et al. 2017)	Cardiovascular	0.15	<0.001	7	1.83E-54	1.42E-51
Coronary artery disease (cross-population)	Cardiovascular	0.42	<0.001	6	3.77E-38	2.13E-37
Coronary artery disease (European only)	Cardiovascular	0.72	<0.001	4	1.40E-10	1.88E-10
Myocardial infarction	Cardiovascular	0.58	<0.001	5	1.09E-21	1.77E-21
Lipid - High-density lipoprotein cholesterol	Cardiovascular	0.65	<0.001	9	<1.00E-100	<1.00E-100
Lipid - Low-density lipoprotein cholesterol	Cardiovascular	0.29	<0.001	13	<1.00E-100	<1.00E-100
Lipid - Total cholesterol	Cardiovascular	0.08	<0.001	14	<1.00E-100	<1.00E-100
Lipid - Triglycerides	Cardiovascular	0.21	<0.001	9	<1.00E-100	<1.00E-100
Adiponectin	Metabolic	0.97	0.06	0	0.02	0.02
Fasting insulin	Metabolic	0.23	<0.001	1	1.78E-06	1.15E-05
Estimated glomerular filtration rate (creatinine)	Metabolic	0.40	<0.001	5	7.90E-22	9.12E-21
Type-2 diabetes	Metabolic	0.13	<0.001	2	2.06E-07	1.09E-06
Modified Stumvoll Insulin Sensitivity Index (ISI)	Metabolic	0.06	0.29	0	0.36	0.60
HbA1c	Metabolic	0.46	0.42	1	0.42	0.46
Lupus	Autoimmune	0.10	<0.001	0	4.86E-05	2.03E-04
Primary biliary cirrhosis	Autoimmune	0.71	0.01	2	4.37E-05	5.42E-05
C-reactive protein	Autoimmune	0.32	<0.001	5	2.66E-78	2.13E-76
Crohn's disease	Autoimmune	0.52	<0.001	4	3.11E-09	4.44E-09
Inflammatory bowel disease	Autoimmune	0.34	<0.001	2	8.37E-07	1.54E-06
Ulcerative colitis	Autoimmune	0.20	0.16	2	0.12	0.17
Rheumatoid arthritis	Autoimmune	0.76	0.22	1	0.21	0.21
Eczema	Autoimmune	0.46	<0.001	3	3.69E-12	6.32E-12
Cognitive performance (SSGAC 2018)	Cognition	0.15	<0.001	4	6.44E-30	3.98E-28
Educational attainment (SSGAC 2018)	Cognition	0.25	<0.001	3	5.96E-63	8.00E-61
Years of schooling (SSGAC 2016)	Cognition	0.34	<0.001	3	4.04E-16	1.30E-15
Fluid intelligence	Cognition	0.18	<0.001	2	2.25E-05	5.66E-05
Age-related hearing impairment	Neurological	0.36	0.19	0	0.18	0.20
Alzheimer's disease (IGAP 2019)	Neurological	0.92	<0.001	1	8.88E-35	9.19E-35
Alzheimer's disease (IGAP 2013)	Neurological	0.61	<0.001	1	3.02E-13	4.24E-13
Any ischemic stroke	Neurological	0.41	0.00	1	1.29E-03	1.70E-03
Any stroke	Neurological	0.71	0.00	1	7.32E-04	7.79E-04
Cardioembolic stroke	Neurological	0.24	0.43	0	0.39	0.44
Large artery stroke	Neurological	0.65	0.04	0	0.03	0.03
Small vessel stroke	Neurological	0.29	0.04	0	0.03	0.04
Parkinson's disease	Neurological	0.73	0.14	0	0.12	0.13
Amyotrophic lateral sclerosis	Neurological	0.97	0.81	0	0.83	0.83
Multiple sclerosis (Beecham et al. 2013 immunochip)	Neurological	0.003	0.04	0	0.04	0.51
Multiple sclerosis (Sawcer et al. 2011 GWAS)	Neurological	0.02	0.03	0	0.03	0.10
Bipolar disorder (PGC 2018)	Psychiatric	0.09	<0.001	3	5.06E-05	2.51E-04
Bipolar disorder (PGC+ICCBD 2017)	Psychiatric	0.12	0.00	3	1.27E-04	3.89E-04
Anorexia	Psychiatric	0.73	0.16	0	0.18	0.18
Posttraumatic stress disorder (PGC 2017)	Psychiatric	0.19	0.07	0	0.08	0.11
Depressive symptoms	Psychiatric	0.05	0.01	1	0.01	0.03
Subjective well-being	Psychiatric	0.57	0.03	1	0.01	0.01
Neuroticism	Psychiatric	0.51	<0.001	0	1.39E-03	1.66E-03
Extraversion	Psychiatric	0.09	0.35	0	0.36	0.48

Loneliness	Psychiatric	0.65	0.47	0	0.45	0.46
Generalized Anxiety disorder (case-control)	Psychiatric	0.76	0.45	0	0.44	0.45
Generalized Anxiety disorder (factor score)	Psychiatric	0.78	0.53	0	0.48	0.48
Schizophrenia (PGC 2014)	Psychiatric	0.28	<0.001	3	2.76E-09	7.91E-09
Major depressive disorder (PGC 2013)	Psychiatric	0.08	0.05	1	0.13	0.41
Major depressive disorder (PGC 2018)	Psychiatric	0.44	0.08	1	0.07	0.08
Age of menopause	Reproductive	0.60	0.00	1	4.91E-06	6.99E-06
Age of menarche	Reproductive	0.25	0.01	1	0.04	0.07
Insomnia complaints	Sleep	0.87	0.20	0	0.15	0.15
Chronotype	Sleep	0.24	0.51	0	0.52	0.57
Oversleeper	Sleep	0.94	0.97	0	0.96	0.96
Sleep duration	Sleep	0.07	0.48	0	0.45	0.57
Undersleeper	Sleep	0.05	0.02	0	0.02	0.05