Integrative analysis of Mendelian randomization and Bayesian colocalization highlights four genes with putative BMI-mediated causal pathways to diabetes

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Genome-wide association studies have identified hundreds of single nucleotide polymorphisms (SNPs) that are associated with BMI and diabetes. However, lack of adequate data has for long time prevented investigations on the pathogenesis of diabetes where BMI was a mediator of the genetic causal effects on this disease. Of our particular interest is the underlying causal mechanisms of diabetes. We leveraged the summary statistics reported in two studies: UK Biobank (N = 336,473) and Genetic Investigation of ANthropometric Traits (GIANT, N = 339,224) to investigate BMI-mediated genetic causal pathways to diabetes. We first estimated the causal effect of BMI on diabetes by using four Mendelian randomization methods, where a total of 76 independent BMI-associated SNPs (R² ≤ 0.001, P < 5 × 10⁻⁸) were used as instrumental variables. It was consistently shown that higher level of BMI (kg/m²) led to increased risk of diabetes. We then applied two Bayesian colocalization methods and identified shared causal SNPs of BMI and diabetes in genes TFAP2B, TCF7L2, FTO and ZC3H4. This study utilized integrative analysis of Mendelian randomization and colocalization to uncover causal relationships between genetic variants, BMI and diabetes. It highlighted putative causal pathways to diabetes mediated by BMI for four genes.

Diabetes is a long term health condition that affects approximately 1 in 11 adults with rapid increase in prevalence worldwide. Elevated BMI in both children and adults has been consistently found causally associated with the risk of diabetes. Genome-wide association studies (GWASs) have identified hundreds of genetic variants, in particular, single nucleotide polymorphisms (SNPs) that are associated with both BMI and diabetes, which have induced investigations on the role of BMI-associated SNPs in the development of diabetes. However, there was limited data on the pathogenesis of diabetes where BMI was a mediator of the genetic causal effects on this disease.

Publicly accessible large-scale GWAS summary results provide great resources of integrative analyses of disease pathogeneses, e.g., Mendelian randomization (MR) and colocalization. MR is designed for estimating causal effect of an exposure on a disease, where exposure associated SNPs are selected as instruments. These instruments are not necessarily causal SNPs due to linkage disequilibrium (LD). Colocalization explores shared causal SNPs of a pair of traits, whether they are exposures, diseases, or exposure and disease. It was not developed for identifying causal relationship between the traits. Thus, the causal questions addressed by the two approaches are different. Each of the approaches alone is insufficient to investigate exposure-mediated causal pathways to a disease. Very recently, frameworks of integrative analysis by combining MR with colocalization have been developed to identify biological mediators in the causal pathways to various clinical outcomes.

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Of our particular interest is the underlying causal mechanisms of diabetes. In this study, we aim to (i) couple MR with Bayesian colocalization to explore whether BMI is a mediator in the genetic causal pathways to diabetes; (ii) investigate the performance of two Bayesian colocalization methods, COLOC and eCAVIAR. COLOC estimates how likely there is a shared causal SNP in a genetic test region for a pair of traits, by assuming there exists at most one causal SNP in the region for either trait, while eCAVIAR allows for multiple causal SNPs. In particular, we exploit the summary results of two independent large-scale GWASs: BMI from the Genetic Investigation of ANthropometric Traits (GIANT) consortium and diabetes from the UK Biobank (round 1), by using BMI-associated SNPs as instruments in MR analysis to estimate causal effect of BMI on diabetes. There were several types of diabetes measures in the UK Biobank project. This work focused specifically on “diabetes diagnosed by doctor” with data collected from a touchscreen question “Has a doctor ever told you that you have diabetes?”.

If there is evidence for a statistically significant causal effect, we then further investigate whether there are shared causal SNPs between BMI and diabetes using COLOC, and therefore gain insights into the underlying mechanisms of diabetes (Fig. 1).

**Results**

**Higher BMI causes increased risk of diabetes.** We used summary statistics of 76 independent BMI associated SNP instruments (\(R^2 \leq 0.001, P < 5 \times 10^{-8}\)) and applied four existing methods (inverse variance weighted estimation, weighted median estimation, MR-Egger regression and MR-RAPS) in our MR analysis. All the MR results consistently showed evidence for a positive causal effect of BMI on diabetes (Table 1), which is agreement with existing literature. Table 1 consists of estimated odds ratios (ORs), confidence intervals (CIs) and corresponding p-values. The estimated ORs are in the range of (1.038, 1.057) and none of the 95% CIs include value 1. The estimated ORs are fairly precise, with the widest 95% CI (1.015, 1.082) from MR-Egger. Our MR results are strongly suggestive of a causal relationship between BMI and risk of diabetes. For example, in the MR-RAPS method (estimated OR:1.048, 95% CI: (1.042, 1.054)), the odds of an individual being diagnosed with diabetes will increase by 4.8% per 1-SD (or 4.5 kg/m²) increase in BMI. The estimated intercept from MR-Egger (estimate: \(-0.001, 95\% \text{ CI: } (-0.002, 0.001)\)), not significantly different from zero, suggests that the null hypothesis of zero average horizontal pleiotropic effect is not rejected. The left panel of Figure S1 is scatter plot of the estimated coefficients in the regression analysis of BMI and diabetes on the 76 independent SNPs included as instruments in our MR analysis. The magnitudes of the slopes of the regression lines correspond to the logarithms of the estimated ORs from the first three MR methods in Table 1.

In our MR analysis, as shown at the bottom of the left panel of Figure S1, we identified an outlier SNP rs7903146 (within gene TCF7L2). This SNP was already found to be an outlier and a horizontal pleiotropic instrument in previous studies on BMI-diabetes causal relationships, which was supported by previous findings that it...
was associated with both fasting glucose and BMI\(^3\). We then performed a sensitivity analysis by excluding the outlier. The results without this SNP in Table S1 and Figure S1 (right panel) show that rs7903146 has little impact on the estimated causal effect of BMI on diabetes. For example, in MR-RAPS, the estimated OR increased by less than 0.4% (from 1.048 to 1.052) and 95% CI changed from (1.042, 1.054) to (1.046, 1.057). We further carried out leave-one-out MR analysis. Again, the results changed little in all the MR methods, which suggests none of the SNP instruments influence the MR estimate disproportionately.

### Shared causal SNPs of BMI and diabetes are highlighted for four genes.

Although we have replicated evidence for a positive causal effect of BMI on diabetes, whether BMI is a mediator on the genetic causal pathways to diabetes is unknown. For such purpose, we used COLOC to test shared causal SNPs between BMI and diabetes. We first included 128 independent SNPs that were associated with either BMI or diabetes (P < 5 × 10\(^{-8}\)). Each of the SNPs and their neighbours (distance within 200 kb) were then utilized to define a test region. After merging overlapping regions, we tested for colocalization in 118 unique regions using R package coloc, \url{http://cran.r-project.org/web/packages/coloc}. Of these unique test regions, four regions in chromosomes 6, 10, 16 and 19 suggest a single causal SNP common to both BMI and diabetes (posterior probability of colocalization PP\(_4\) > 0.9; Table 2); one region in chromosome 3 suggests two distinct causal SNPs, one for BMI only and the other for diabetes only (posterior probability of distinct causal SNPs PP\(_3\) = 1; Table 2). In each of the five regions, we further calculated the posterior probability of each SNP being causal to both of the traits (PP\(_4\), Both, Fig. 2).

| Region | N     | PP\(_3\) | PP\(_4\) | Candidate causal SNP | Chr: position | Gene   | Alleles | EAF  | B_{BMI} | P_{BMI}  | B_{diabetes} | P_{diabetes} |
|--------|-------|----------|----------|----------------------|--------------|--------|---------|------|---------|-----------|----------------|----------------|
| chr6: 50600724-51065757 | 472    | 0.049    | 0.923    | rs987237             | 6: 50803050  | TFAP2B | G/A    | 0.09 | 0.044   | 1.07 × 10\(^{-3}\) | 0.001           | 5.67 × 10\(^{-8}\) |
| chr10: 114554779-114958159 | 447    | 0.001    | 0.999    | rs7903146            | 10: 114758349 | TCF7L2 | T/C    | 0.25 | -0.204  | 1.10 × 10\(^{-16}\) | 0.015           | 3.72 × 10\(^{-10}\) |
| chr16: 53604177-54000907 | 247    | 0.026    | 0.974    | rs1558902            | 16: 53803574 | FTO    | A/T    | 0.45 | 0.084   | 1.13 × 10\(^{-16}\) | 0.005           | 1.67 × 10\(^{-11}\) |
| chr19: 47369753-47761543 | 386    | 0.004    | 0.993    | rs3810291            | 19: 47569003 | ZC3H4 | A/G    | 0.63 | 0.029   | 6.35 × 10\(^{-16}\) | 0.003           | 1.24 × 10\(^{-8}\) |
| chr3: 185324933-186022133 | 112    | 1        | 0.000    | rs9816226*           | 3:185344499 | ETV5   | A/T    | 0.15 | -0.040  | 6.03 × 10\(^{-16}\) | 0.002           | 2.88 × 10\(^{-13}\) |
|         |       |          |          | rs1470580            | 3:185529174 | IGF2BP2 | A/T    | 0.29 | -0.014  | 1.03 × 10\(^{-8}\)  | 0.006           | 7.88 × 10\(^{-17}\) |

Table 2. Evidence for a shared causal SNP or two distinct causal SNPs between BMI and diabetes shown in five regions. PP\(_3\) denotes the posterior probability of two distinct causal SNPs, one for BMI only and the other for diabetes only. N is the number of SNPs included in the test region. Alleles (effect/reference), effect allele frequency (EAF), estimated coefficient (B) and p-value (P) are summary results from two independent GWASs: BMI from the GIANT study http://portals.broadinstitute.org/collaboration/giant/index.php/GIANT_consortium_data_files#GWAS_Anthropometric_2015_BMI, diabetes from the UK Biobank study http://www.nealelab.is/blog/2017/7/19/rapid-gwas-of-thousands-of-phenotypes-for-337000-samples-in-the-uk-biobank. * The SNP rs9816226 does not lie within a gene. ETV5 is its nearest gene.
Discussion
Interestingly, estimated causal effect of BMI on the risk of diabetes in our MR analysis were smaller but more precise than those reported in the literature. This could be partly explained by (i) the number of instruments. For example, we included 76 instruments in our analysis. However, Fall et al. used a single instrument in their study which could not separate mediation from pleiotropy4; (ii) sample size. We used GWAS summary data for diabetes from the UK Biobank on the basis of ~ 330,000 individuals which was almost five times the sample size.
in Corbin’s study; (iii) genotyping platforms. The summary results in this study were from recent GWASs which used more advanced genotyping technology which provides more accurate genotype measures.

MR, COLOC and eCAVIAR use summary statistics from large-scale GWASs. However, as they were designed for different purposes, each of which alone is insufficient to investigate mediators on the genetic causal pathways. We have applied an integrative approach by combining MR with COLOC to investigate BMI-mediated genetic causal pathways to diabetes. The five regions highlighted in COLOC have also been analyzed using eCAVIAR. Our results have consistently shown that if one shared causal SNP is present, then MR + COLOC and MR + eCAVIAR could serve as alternatives. However, where there is evidence for two distinct causal SNPs in a region, eCAVIAR has shown much weaker evidence for each of the distinct SNPs being causal to both BMI and diabetes. This might be because when multiple causal SNPs exist, assuming a single causal SNP results in an underestimation of the posterior probability of a shared causal SNP in eCAVIAR. Thus, if there exists at most one causal SNP to either trait, one may prefer COLOC as it provides relatively robust results. In addition to providing the posterior probability of colocalization, COLOC also computes the likelihood of distinct causal SNPs in a test region, which is another advantage comparing with eCAVIAR when there are two distinct causal SNPs in a test region.

In our analysis, both MR + COLOC and MR + eCAVIAR have consistently shown evidence for causal effects on diabetes mediated by BMI for four genes (TFAP2B, TCF7L2, FTO and ZC3H4). Previous studies have suggested that TFAP2B is associated with BMI in both Europeans and African Americans. This gene has been found associated with waist circumference which is known to be correlated with BMI and may modify the effect of dietary fat intake on weight loss and waist reduction. Evidence for its association with diabetes in the UK population and a candidate contributor of the susceptibility to diabetes has also been reported. Thus, the TFAP2B gene may play an important role of the underlying mechanisms of diabetes, where BMI is a mediator.

rs7903146, located in the intronic region of the TCF7L2 gene, is associated with BMI and diabetes in the European population. It was an outlier instrument in our study and in the literature. However, there was little change in estimated causal effect of BMI on diabetes after removing this SNP in our sensitivity analysis. It was identified as a potential pleiotropic but weak instrument. Thus, the MR analysis performed in this study without this SNP provided higher precision in the estimates.

FTO is associated with BMI and diabetes. In addition, rs9939609 in this gene has been identified to affect diabetes through BMI in the UK population. This SNP is in high linkage disequilibrium with the SNP rs1558902 (R² = 0.918) highlighted in our analysis. However, its association with diabetes is reported to be partly independent of its effect on BMI in the Scandinavian population and in east and south Asian. Such discrepancy might come from the heterogeneity across populations.

Rs3810291 (in ZC3H4) has been associated with BMI in European populations. Recent research has further shown that it is associated with both BMI and diabetes in children. Our analysis indicates that this SNP may causally affect diabetes through BMI in the Europeans.

In region chr3:185324933–186022133, there may exist two independent causal SNPs: rs9816226 (nearest gene: ETV5) for BMI and rs1470580 (in IGF2BP2) for diabetes. Previous GWASs have shown that genetic variation of ETV5 is predictive of BMI in multiple populations including the Europeans while IGF2BP2 predictive of the onset of diabetes in European populations and a Chinese Han population. Thus, IGF2BP2 may causally affect diabetes but not through BMI.

The MR + COLOC approach in our study assumes that the two non-overlapping samples of the two GWASs are from the same population. We have used the summary statistics from the UK Biobank (British population) and GIANT studies (individuals of all ancestries). One possible limitation of our study would be that if the individuals of the two studies came from different populations and/or partly overlap, our results may be biased due to violations of the assumption. The unavailability of the individual level data in this study has hindered us from testing the plausibility of this assumption.

Our analysis using MR + COLOC has highlighted, indirectly, four genes with putative BMI-mediated causal pathways to diabetes. These findings, however, need further validation investigations (e.g. mediation analysis using individual level data).

Another limitation is GWAS results of diabetes we downloaded from the Neale lab. They used linear regressions rather than logistic regressions for binary phenotypes, although bias from such model misspecification is not an important issue in our study thanks to a large number of diabetes cases (~17,000) and rare variants excluded as instruments.

Conclusion

In this paper, we have recommended an analytical approach MR + COLOC to investigate causal pathways to diabetes. The causal effects of four genes on diabetes were found mediated by BMI. Both COLOC and eCAVIAR have highlighted the same SNPs that are most likely to be shared causal signals when there is a single shared causal SNP. For a specific study, if we believe there exists at most one causal SNP for either trait or wish to test whether there exists two distinct causal SNPs in a genetic region, COLOC is recommended. If there are multiple shared causal SNPs, one may however prefer eCAVIAR.

The approach applied in the present study takes forward strengths of both MR and colocalization, which can be used as an indirect approach of investigating genetic causal pathways where a mediator conveys the genetic effects. This approach provides new insights into causal mechanisms of diabetes that could be further validated in other studies and ultimately help in the development of new and effective treatments.
Methods

Study design and data. Figure 1 depicts the workflow of our study design to investigate putative BMI-mediated causal pathways to diabetes. We used publicly available GWAS summary data of BMI and diabetes. BMI summary data (including major and minor alleles and allele frequencies on 2,554,637 SNPs from 339,224 individuals of all ancestries, estimated effects of allele dose and their standard errors and corresponding p-values) were downloaded from http://portals.broadinstitute.org/collaboration/giant/index.php/GIANT_consortium_data_files#GWAS_Anthropometric_2015_BMI. The same summary statistics of diabetes on 10,894,596 SNPs from 336,473 unrelated individuals from the UK were downloaded from https://www.dropbox.com/s/41q5uj1sa6v99y0/2443.assoc.tsv.gz?dl=0.

Investigation of causal relationship between BMI and diabetes using Mendelian randomization.

First, we used MR to test if BMI causally affects diabetes, where BMI associated SNPs were used as the instrumental variables (IVs). The assumptions of MR are represented by a directed acyclic graph in Fig. 3. An IV is: i) associated with the exposure (an arrow pointing from IV to BMI), ii) independent of the confounders (no arrow between IV and confounders) and iii) independent of the outcome, conditioning on the exposure and the confounders (no arrow pointing directly from IV to diabetes). The third condition assumes that there exists no direct effect of the IV on the outcome, i.e., no horizontal pleiotropy47, which can be relaxed, for example, in MR-Egger regression. A direct arrow from IV to diabetes is present in Fig. 3 because MR-Egger regression was included in our MR analysis. Of the four MR methods applied in this study, MR robust adjusted profile score (MR-RAPS) was designed to reduce weak instrument bias39.

MR requires that SNP instruments are mutually independent. We first filtered the 2,042 BMI associated SNPs (P < 5 × 10^{-8}) from the GIANT GWAS results by clumping on the MR-Base platform48 to ensure that the instruments in MR analysis were independent of each other. That is, the SNPs in LD (R^2 ≥ 0.001) were clumped together and only the one with the lowest p-value was retained. This led to 76 independent BMI-associated SNPs (Table S2) included as the IVs in MR analysis using four existing MR methods (inverse variance weighted estimation, weighted median estimation, MR-Egger regression and MR-RAPS) in R package TwoSampleMR, https://mrcieu.github.io/TwoSampleMR/. The SNP rs7903146 was detected as an outlier instrument, which was removed in our sensitivity analysis. Leave-one-out analysis was further carried out by leaving one instrument out at a time (Table S2) included as the IVs in MR analysis using four existing MR methods (inverse variance weighted estimation, weighted median estimation, MR-Egger regression and MR-RAPS) in R package TwoSampleMR, https://mrcieu.github.io/TwoSampleMR/. The SNP rs7903146 was detected as an outlier instrument, which was removed in our sensitivity analysis. Leave-one-out analysis was further carried out by leaving one instrument out at a time in our MR analysis. Of the four MR methods applied in this study, MR robust adjusted profile score (MR-RAPS) was designed to reduce weak instrument bias39.

Identification of shared causal genes between BMI and diabetes using COLOC.

Since the IVs are required to be associated with BMI but not necessarily in a causal way, one cannot decide whether such associations are causal. To investigate genetic causal pathways to diabetes, we applied COLOC to detect shared causal SNPs between the two traits: BMI and diabetes. This approach assumes that: (1) in each test region, there exists at most one causal SNP for either trait; (2) the probability that a SNP is causal is independent of the probability that any other SNP in the genome is causal; (3) all causal SNPs are genotyped or imputed and included in analysis. According to these assumptions, there are five mutually exclusive hypotheses for each test region: (1) there is no causal SNP for either trait (H_0); (2) there is one causal SNP for trait 1 only (H_1); (3) there is one causal SNP for trait 2 only (H_2); (4) there are two distinct causal SNPs, one for each trait (H_3); and (5) there is a causal SNP common to both traits (H_4). Our primary interest lies in the last hypothesis H_4 - colocalization. Support for each of the hypotheses is quantified by the posterior probability (PP), denoted by PP_0, PP_1, PP_2, PP_3, and PP_4 accordingly. These PPs were calculated from the priors and the approximate Bayes factors. We set the prior probability of each SNP that is causal to either of the traits to 1 × 10^{-8} (i.e., one in 10,000 SNPs in the genome are causal to either trait) and causal to both traits to 1 × 10^{-6} (i.e., one in 100 SNPs in the genome causal to one trait are causal to both traits). We used the GWAS summary statistics of BMI and diabetes to approximate the Bayes factors. COLOC treats trait 1 and trait 2 as two outcomes in no particular order. Thus, it has no capacity for examining relationships between BMI and diabetes.

To define test regions in COLOC, we included the 76 independent BMI-associated SNPs from our MR analysis and 52 independent diabetes-associated SNPs (P < 5 × 10^{-8}). Each of these 128 SNPs and their neighbor SNPs (distance within 200 kb on https://genome.ucsc.edu GRCh37/hg19) were used to define a test region. After
merging overlapping regions, we tested for colocalization in 118 unique regions that were associated with either BMI or diabetes in COLOC using R package coloc, http://cran.r-project.org/web/packages/coloc.

Colocalization analysis using eCAVIAR. Next, we applied another Bayesian method eCAVIAR to colocalization analysis. The aim of eCAVIAR is to quantify the likelihood of the number of shared causal SNPs of an exposure and a disease in a test region. This approach assumes that: 1) in each test region, at least one SNP is causal to either the exposure or the disease; 2) the probability that a SNP is causal to the exposure is independent of the probability that it is causal to the disease. It computes the colocalization posterior probability (CLPP) that the same SNP is causal to both the exposure and the disease. This approach allows for multiple shared causal SNPs and is therefore different from COLOC which assumes at most one shared causal SNP in each region. To ensure the results of the two approaches were comparable, we assumed only one SNP is causal to BMI or to diabetes in each of the five regions in eCAVIAR (https://github.com/fhormoz/caviar).

Ethics statement. Ethical approval was not required for this study that used publicly available GWAS summary statistics.

Data availability
The datasets analysed during the current study are publicly available from http://portals.broadinstitute.org/collaboration/giant/index.php/GIANT_consortium_data_files#GWAS_Anthropometric_2015_BMI by clicking on “Download BMI All Ancestry GZIP” for BMI and https://www.dropbox.com/s/41q5uj1sa6v99y0/2443.assoc.tsv.gz?dl=0 for diabetes.

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References
1. International Diabetes Federation. IDF Diabetes Atlas Eighth Edition 2017. International Diabetes Federation https://www.idf.org/news/94-new-idf-figures-show-continued-increase-in-diabetes-across-the-globe-reiterating-the-need-for-urgent-action.html (2017).
2. Cains, J. C. et al. Childhood adiposity and risk of type 1 diabetes: A Mendelian randomization study. PLoS Med. 14, e1002362 (2017).
3. Corbin, L. J. et al. BMI as a modifiable risk factor for type 2 diabetes: Refining and understanding causal estimates using mendelian randomization. Diabetes 65, 3002–3007 (2016).
4. Fall, T. et al. The role of adiposity in cardiometabolic traits: A Mendelian randomization analysis. PLoS Med. 10, e1001474 (2013).
5. Holmes, M. V. et al. Causal effects of body mass index on cardiometabolic traits and events: A Mendelian randomization analysis. Am. J. Hum. Genet. 94, 198–208 (2014).
6. Lyall, D. M. et al. Association of body mass index with cardiometabolic disease in the UK biobank: A mendelian randomization study. JAMA Cardiol 2, 882–889 (2017).
7. Geng, T., Smith, C. E., Li, C. & Huang, T. Childhood BMI and adult type 2 diabetes, coronary artery diseases, chronic kidney disease, and cardiometabolic traits: A Mendelian randomization analysis. Diabetes Care 41, 1089–1096 (2018).
8. Cheng, L. et al. Exposing the causal effect of body mass index on the risk of type 2 diabetes mellitus: A mendelian randomization study. Front. Genet 10, 1–10 (2019).
9. Zhu, Z. et al. Integration of summary data from GWAS and eQTL studies predicts complex trait gene targets. Nat. Genet. 48, 481–487 (2016).
10. Locke, A. E. et al. Genetic studies of body mass index yield new insights for obesity biology. Nature 518, 197–206 (2015).
11. Gong, J. et al. Fine mapping and identification of BMI loci in African Americans. Am. J. Hum. Genet. 93, 661–671 (2013).
12. Stocks, T. et al. TFA2P2B influences the effect of dietary fat on weight loss under energy restriction. PLoS One 7, e43212 (2012).
13. Maeda, S. et al. Genetic variations in the gene encoding TFA2P2B are associated with type 2 diabetes mellitus. J. Hum. Genet. 50, 283–292 (2005).
14. Grant, S. F. A. et al. Variant of transcription factor 7-like 2 (TCF7L2) gene confers risk of type 2 diabetes. Nat. Genet. 38, 320–323 (2006).
15. Frayling, T. M. et al. A common variant in the FTO gene is associated with body mass index and predisposes to childhood and adult obesity. Science. 316, 889–894 (2007).
16. Burton, P. R. et al. Genome-wide association study of 14,000 cases of seven common diseases and 3,000 shared controls. Nature 447, 661–678 (2007).
17. Visscher, P. M., Brown, M. A., McCarthy, M. I. & Yang, J. Five years of GWAS discovery. Am. J. Hum. Genet. 90, 7–24 (2012).
18. Rietveld, C. A. et al. GWAS of 126,559 individuals identifies genetic variants associated with educational attainment. Science 340, 1467–1471 (2013).
19. Ripeke, S. et al. Genome-wide association analysis identifies 13 new risk loci for schizophrenia. Nat. Genet. 45, 1150–1159 (2013).
20. Bowden, J., Smith, G. D. & Burgess, S. Mendelian randomization with invalid instruments: Effect estimation and bias detection through Egger regression. Int. J. Epidemiol. 44, 512–525 (2015).
21. Bowden, J., Davey Smith, G., Haycock, P. C. & Burgess, S. Consistent estimation in Mendelian randomization with some invalid instruments using a weighted median estimator. Genet. Epidemiol. 40, 304–314 (2016).
22. Smith, G. D. & Ebrahim, S. ‘Mendelian randomization’: Can genetic epidemiology contribute to understanding environmental determinants of disease? Int. J. Epidemiol. 32, 1–22 (2003).
23. Giambardomei, C. et al. Bayesian test for colocalisation between pairs of genetic association studies using summary statistics. PLoS Genet. 10, e1004383 (2014).
24. Guo, H. et al. Integration of disease association and eQTL data using a Bayesian colocalisation approach highlights six candidate causal genes in immune-mediated diseases. Hum. Mol. Genet. 24, 3305–3313 (2015).
25. Hormozdari, F. et al. Colocalization of GWAS and eQTL signals detects target genes. Am. J. Hum. Genet. 99, 1245–1260 (2016).
26. Pickrell, J. K. et al. Detection and interpretation of shared genetic influences on 42 human traits. Nat. Genet. 48, 709–717 (2016).
27. Burgess, S., Foley, C. N. & Zuber, V. Inferential causal relationships between risk factors and outcomes from genome-wide association study data. Annu. Rev. Genomics Hum. Genet. 19, 303–327 (2018).
28. Richardson, T. G. et al. Mendelian randomization analysis identifies CpG sites as putative mediators for genetic influences on cardiovascular disease risk. Am. J. Hum. Genet. 101, 590–602 (2017).
29. Hemani, G., Bowden, J. & Davey Smith, G. Evaluating the potential role of pleiotropy in Mendelian randomization studies. Hum. Mol. Genet. 27, R195–R208 (2018).
30. McGowan, L. M., Davey Smith, G., Gaunt, T. R. & Richardson, T. G. Integrating Mendelian randomization and multiple-trait colocalization to uncover cell-specific inflammatory drivers of autoimmune and atopic disease. Hum. Mol. Genet. 28, 3293–3300 (2019).
31. The Neale Lab. UK Biobank GWAS Results. http://www.nealelab.is/uk-biobank (2018).
32. Manning, A. K. et al. A genome-wide approach accounting for body mass index identifies genetic variants influencing fasting glycemic traits and insulin resistance. Nat. Genet. 44, 659–669 (2012).
33. Flegal, K. M. et al. Comparisons of percentage body fat, body mass index, waist circumference, and waist-stature ratio in adults. Am. J. Clin. Nutr. 89, 500–508 (2009).
34. Thorleifsson, G. et al. Genome-wide association yields new sequence variants at seven loci that associate with measures of obesity. Nat. Genet. 41, 18–24 (2009).
35. Dina, C. et al. Variation in FTO contributes to childhood obesity and severe adult obesity. Nat. Genet. 39, 724–726 (2007).
36. Scuteri, A. et al. Genome-wide association scan shows genetic variants in the FTO gene are associated with obesity-related traits. PLoS Genet. 3, e115 (2007).
37. Scott, L. J. et al. A genome-wide association study of type 2 diabetes in Finns detects multiple susceptibility variants. Science. 316, 1341–1345 (2007).
38. Hertel, J. K. et al. FTO, type 2 diabetes, and weight gain throughout adult life: A meta-analysis of 41,504 subjects from the scandinavian HUNT, MDC, and MPP studies. Diabetes 60, 1637–1644 (2011).
39. Li, H. et al. Association of genetic variation in FTO with risk of obesity and type 2 diabetes with data from 96,531 East and South Asians. Diabetologia 55, 981–995 (2012).
40. Sovio, U. et al. Association between common variation at the FTO locus and changes in body mass index from infancy to late childhood: The complex nature of genetic association through growth and development. PLoS Genet. 7, e1001307 (2011).
41. Clayton, D. G. et al. Population structure, differential bias and genomic control in a large-scale, case-control association study. Nat. Genet. 37, 1243–1246 (2005).
42. Lohmueller, K. E., Pearce, C. L., Pike, M., Lander, E. S. & Hirschhorn, J. N. Meta-analysis of genetic association studies supports a contribution of common variants to susceptibility to common disease. Nat. Genet. 33, 177–182 (2003).
43. Speliotes, E. K. et al. Association analyses of 249,796 individuals reveal 18 new loci associated with body mass index. Nat. Genet. 42, 937–948 (2010).
44. Willer, C. J. et al. Six new loci associated with body mass index highlight a neuronal influence on body weight regulation. Nat. Genet. 41, 23–34 (2009).
45. Sladek, R. et al. A genome-wide association study identifies novel risk loci for type 2 diabetes. Nature 445, 881–885 (2007).
46. Wu, Y. et al. Common variants in CDKAL1, CDKN2A/B, IGF2BP2, SLC30A8, and HHEX/IDE genes are associated with type 2 diabetes and impaired fasting glucose in a Chinese Han population. Diabetes 57, 2834–2842 (2008).
47. Verbanck, M., Chen, C. Y., Neale, B. & Do, R. Detection of widespread horizontal pleiotropy in causal relationships inferred from Mendelian randomization between complex traits and diseases. Nat. Genet. 50, 693–698 (2018).
48. Hemani, G. et al. The MR-base platform supports systematic causal inference across the human phenotype. Elife 7, e34408 (2018).

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Author contributions
Q.L. conceived and designed the study, performed statistical analysis, wrote the first version of the manuscript, interpreted the data and contributed to the final manuscript. H.G. conceived and designed the study, supervised statistical analysis, wrote the first version of the manuscript, interpreted the data and contributed to the final manuscript. J.P. interpreted the data and contributed to the final manuscript. C.B. interpreted the data and contributed to the final manuscript. M.K.R interpreted the data and contributed to the final manuscript.

Competing interests
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Additional information
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