Interdisciplinary Research in Medical Sciences Specialty
ISSN: 3062-4401
2025, Volume 5, Issue 2, Page No: 220-232

Available online at: www.galaxypub.co/page/journals

Galaxy Publication

A Transcriptome-Wide Mendelian Randomization and Colocalization Analysis of Immune Cell Gene Expression in Type 1 Diabetes

Kristian Eriksen[1]*, Solveig Berg[1], Magnus Dahl[2]

[1]Department of Immunogenetics and Mendelian Randomization, Faculty of Medicine, University of Oslo, Oslo, Norway.
[2]Department of Type 1 Diabetes Research and Transcriptomics, Faculty of Medicine, University of Bergen, Bergen, Norway.

***E-mail** ✉ kristian.eriksen@medisin.uio.no

Received: 25 August 2025; **Revised:** 03 November 2025; **Accepted:** 10 November 2025

ABSTRACT

Strategies to avert type 1 diabetes (T1D), a condition driven by autoimmune processes and necessitating continuous lifelong therapy, are still constrained. This investigation aimed to uncover new candidate therapeutic targets for T1D by employing Mendelian randomization together with colocalization analyses that relied on genetic instruments originating from immune cells. We identified cis-acting genetic variants across 14 transcriptomic datasets to instrument the expression of 8998 genes profiled in various immune cell populations. Association data for the outcome were derived from a large T1D genome-wide association study encompassing 18,942 cases and 501,638 controls. Secondary evaluations examined potential horizontal pleiotropy, assessed novelty of signals, and conducted phenome-wide scans combined with colocalization (PheWAS-coloc) using the instrumental variants. We highlighted 21 genes (CLNK, EED, LZTFL1, MGAT4A, NAA38, NFKB1, PHACTR4, PHLPP2, PLEKHA1, P2RY12, REST, RGS14, SERPINB6, SESN3, SLC25A29, SPAG1, STIM2, THEMIS, TMEM80, VSIR, ZNF217) that had not previously emerged in T1D genome-wide association studies. Of particular interest, increased genetically determined expression of VSIR (which codes for the immune checkpoint molecule VISTA) correlated with lower T1D susceptibility. This genetic evidence from humans reinforces and extends animal model data indicating a protective function of VISTA against autoimmune pathology. Another prioritized gene, P2RY12, is already the target of multiple approved pharmaceuticals, suggesting a feasible drug repurposing strategy. Complementary PheWAS-coloc results associated P2RY12 expression with antibody responses to Epstein-Barr virus EBNA-1, thereby implicating an autoimmune-relevant biological pathway. The present results open fresh opportunities for developing and repositioning therapeutic agents intended to prevent or postpone the emergence of T1D.

Keywords: Type 1 diabetes, Mendelian randomization, Genetic epidemiology, Drug development, Transcriptomics

How to Cite This Article: Eriksen K, Berg S, Dahl M. A Transcriptome-Wide Mendelian Randomization and Colocalization Analysis of Immune Cell Gene Expression in Type 1 Diabetes. Interdiscip Res Med Sci Spec. 2025;5(2):220-32. https://doi.org/10.51847/unZGcEVWy0

Introduction

Type 1 diabetes (T1D) results from the autoimmune destruction of insulin-secreting β cells located in the pancreatic islets. Diagnosis commonly occurs in childhood, after which patients face a lifetime of intensive daily disease management accompanied by heightened risks of early morbidity and mortality [1]. Beyond its effects on patients, T1D generates major societal burdens through healthcare costs and diminished workforce participation [2]. These challenges are expected to intensify as the incidence of T1D continues to climb [3]. Effective prevention of disease onset would therefore substantially lessen both personal suffering and broader economic consequences. Clinical evaluation of teplizumab, an anti-CD3 antibody, has shown that intervening against an individual molecular target can postpone T1D development in high-risk family members of affected individuals [4]. However, further therapeutic approaches are urgently needed, as 43% of treated participants still progressed to

clinical disease within a median observation period of 2 years. Mendelian randomization (MR) represents a powerful method for nominating candidate drug targets through evaluation of their causal influence on disease susceptibility [5]. By exploiting the random allocation of genetic variants at conception, MR helps circumvent confounding and reverse causation issues that commonly affect observational research [6]. In addition, conclusions drawn from human genetic variation frequently align more closely with human physiology than findings from experimental animal systems. Supporting this view, compounds directed at proteins backed by robust human genetic data demonstrate elevated success rates in achieving regulatory approval [7].

Earlier applications of MR to T1D have illuminated aspects of disease causation using instruments that modify plasma protein abundances [8, 9]. Despite their value, circulating proteins may not capture the most relevant biology for T1D compared with expression patterns in immune cells or other disease-related tissues. To address this gap and identify additional potential drug targets, we conducted a transcriptomic Mendelian randomization study of T1D that leveraged eQTL instruments derived specifically from purified immune cell populations.

Materials and Methods

Mendelian randomization assumptions

Valid Mendelian randomization inference rests on three core assumptions: relevance, independence, and exclusion restriction. The relevance assumption is met when the chosen genetic instruments display robust associations with the exposure variable. The independence assumption requires that instruments remain unassociated with confounders of the exposure–outcome link. As an illustration, population stratification could lead a variant to show elevated frequency in a particular ancestral group that also carries higher disease risk, thereby distorting the apparent relationship between variant and outcome. The exclusion restriction assumption specifies that any effect of the instruments on the outcome must operate solely through the exposure of interest, without involvement of alternative horizontal pleiotropic routes.

Genetic instruments and outcome associations

Selection of genetic instruments relied on 29 eQTL datasets generated from 14 independent investigations [10-23] that profiled gene expression in multiple immune cell lineages. All datasets were retrieved from the eQTL Catalog [24], which compiles publicly released gene expression quantitative trait loci resources. Donor cohorts across these 14 studies were composed entirely or overwhelmingly of European-ancestry individuals. The 29 datasets included two measuring B-cell expression, three assessing CD4+ T cells, two evaluating CD8+ T cells, five examining lymphoblastoid cell lines (LCLs), four focusing on macrophages, one on microglia, six on monocytes, three on neutrophils, one on natural killer (NK) cells, one on total T cells, and one on regulatory T-cell memory cells (Treg memory).

All cis-acting eQTLs achieving genome-wide significance ($P < 5 \times 10{-}8$) were retained to fulfill the relevance criterion. These variants underwent LD clumping ($r^2 < 0.1$) using plink (v1.07) to retain largely independent instruments for each gene in each dataset [25]. F-statistics were calculated for every instrument, and Steiger filtering was applied to verify directional consistency. All retained instruments exhibited F-statistics > 10 and satisfied Steiger filtering at $P < 0.05$. The analysis was limited to protein-coding genes identified by UniProt accession numbers. Variants mapping to the major histocompatibility complex (MHC) region (chr6:25,726,063–33,400,644) were omitted owing to its intricate linkage disequilibrium architecture [8, 26].

Summary statistics linking the instruments to T1D risk were drawn from the extensive GWAS meta-analysis by Chiou *et al*. [27], encompassing 18,942 cases and 501,638 controls of European descent—the most comprehensive public T1D GWAS currently available. Although this GWAS is confined to European participants, the matching ancestry with the eQTL data strengthens adherence to the MR independence assumption.

Mendelian randomization analysis

Single-variant instruments were analyzed using the Wald ratio approach. For genes with multiple independent variants within one dataset, fixed-effect inverse-variance weighted meta-analysis was employed to combine the individual variant estimates. Heterogeneity among variant-specific effects was quantified using Cochran's Q test P-value and the I^2 statistic for multi-variant instruments, which may signal underlying pleiotropy. Variants were retained regardless of detected heterogeneity. A dataset-specific Bonferroni threshold, adjusted for the total

number of genes tested in each of the 29 datasets, defined statistical significance. All analyses were executed with the TwoSampleMR R package [28] (https://github.com/MRCIEU/TwoSampleMR) under R version 4.3.1.

Follow-up analyses

Drug-target MR studies employing cis-QTL instruments typically yield sparse instruments after LD pruning [29]. In this analysis, merely 1.0% of instruments included four or more cis-eQTLs. Traditional pleiotropy-robust methods such as MR-Egger and weighted median therefore have restricted utility, as they require instruments with many variants [29]. To compensate, we implemented a series of targeted follow-up procedures detailed in the following sections.

Pairwise conditional colocalization

A significant MR association may occasionally stem from linkage disequilibrium between the instrumental variant and an independent causal variant for T1D, rather than a direct causal relationship with the queried gene (LD confounding). To investigate this scenario, pairwise conditional colocalization (PWCoCo) [30] was performed between relevant eQTL summary data (± 500 kb of each instrumental variant) and T1D GWAS statistics for all associations meeting the MR significance criteria.

This Bayesian colocalization framework generates five posterior probabilities: PPH0 (no signal for either trait), PPH1 (signal for gene expression alone), PPH2 (signal for T1D alone), PPH3 (distinct signals for both traits), and PPH4 (a common causal signal for both traits). Default prior probabilities were applied ($p1 = 1 \times 10^{-4}$, $p2 = 1 \times 10^{-4}$, $p12 = 5 \times 10^{-5}$). PWCoCo begins with standard marginal colocalization [31] and, when PPH4 < 0.8, advances to conditional analysis via GCTA-COJO [32] using appropriate LD reference data. This step mitigates interference from multiple independent signals that violate the single-causal-variant assumption of conventional colocalization. Colocalization support was defined primarily as PPH4 > 0.8, with sensitivity checks at PPH4 > 0.7 and PPH4 > 0.9.

LD reference information for GCTA-COJO was obtained from 487,395 UK Biobank participants genotyped using two custom Affymetrix arrays [33]. Data were phased with SHAPEIT3 [34] and imputed with the Haplotype Reference Consortium and UK10K reference panels via IMPUTE4 [33]. Quality filtering removed variants with MAF < 1%, Hardy-Weinberg equilibrium $P < 1 \times 10^{-6}$, or strand-ambiguous alleles (C/G, A/T). Samples exhibiting > 5% missingness, outlier heterozygosity rates, or sex mismatches were also excluded.

Assessment of horizontal pleiotropy

Focusing exclusively on cis-eQTLs raises the chance that detected relationships between genetic variants and phenotypes reflect downstream consequences of gene expression changes (vertical pleiotropy). Such effects remain compatible with the exclusion restriction assumption. However, an eQTL located in a regulatory sequence might simultaneously modulate the transcription of several adjacent genes. Under these circumstances, it can be challenging to identify the specific gene whose altered expression is causally responsible for the observed disease association. We treated such instances as a manifestation of horizontal pleiotropy. As an illustration, at the 1q23.1 locus, eQTLs influencing both TTC24 (profiled in regulatory T-cell memory cells) and GPATCH4 (profiled in CD4+ T cells) colocalized with the same T1D risk signal, and the variant rs35091159 functioned as an instrument for both genes. To address this issue, we clustered significant results by genomic region—defined as instrumental variants belonging to different genes and positioned within 250 kb of each other. Any region containing signals for more than one distinct gene was interpreted as evidence of horizontal pleiotropy.

Assessment of novelty

A result was considered novel when none of its instrumental variants displayed correlation ($r^2 < 0.2$ based on the 1000 Genomes EUR panel) or physical proximity (± 250 kb) to any genome-wide significant T1D variants ($P < 5 \times 10{-}8$) identified in five prior association studies [27, 35-38], and when the gene had not been implicated in two earlier Mendelian randomization analyses [8, 9].

Phenome-wide scans with colocalization

Phenome-wide association studies (PheWAS) enable the identification of potential on-target consequences for traits unrelated to T1D, such as adverse safety signals that merit early consideration. They can also highlight biologically intermediate phenotypes connected to T1D etiology, thereby providing mechanistic understanding

and bolstering confidence in causal claims. For this reason, novel results that stood alone within their genomic region were examined using the MRbase PheWAS resource [28], a repository encompassing summary statistics from over 50,000 publicly released GWAS covering diverse phenotypes. In addition, associations with plasma concentrations of 4,775 proteins assayed by the SomaLogic platform in a sample of 10,708 participants were investigated [39, 40] (data accessible at www.omicscience.org). For genes instrumented by several independent variants, the one exhibiting the strongest link to gene expression was chosen for these queries. Reported associations were those reaching $P < 5 \times 10^{-5}$. PWCoCo analyses were also performed between the eQTL data and PheWAS statistics within a ± 500 kb window around the queried variant to help differentiate true shared causality from linkage disequilibrium confounding. Notably, these PheWAS-coloc examinations offer further utility in detecting horizontal pleiotropy by uncovering shared causal influences between the instrumental variants and the expression of nearby genes or proteins, drawing upon both the queried SomaLogic protein measurements and the MR-Base platform, which contains whole-blood expression GWAS for nearly 20,000 genes contributed by the eQTLGen Consortium [41].

Replication in African ancestry

To evaluate the generalizability of findings, MR analyses were repeated in participants of African ancestry (AA). Instruments were obtained from whole-blood expression data generated in 757 AA individuals by Kachuri *et al*. [42], since immune-cell-specific eQTL resources for AA populations are currently unavailable in the public domain. Variant selection followed the same procedures as the main analysis, retaining associations at $P < 5 \times 10^{-8}$ and ensuring independence at $r^2 < 0.1$. T1D outcome associations were derived by meta-analyzing summary statistics from two independent AA GWAS [43, 44] using METAL software [45]. Verma *et al*. contributed data on 6,451 cases identified through ICD coding in Veterans Health Administration records paired with 109,410 controls. Michalek *et al*. provided results from 409 confirmed T1D cases and 482 controls recruited by the Type 1 Diabetes Genetics Consortium [46].

Results and Discussion

Study design

An overview of the complete analytical framework is presented in **Figure 1**. Locally acting cis-eQTLs serving as instruments for gene expression in immune cells were extracted from 14 studies (yielding 29 datasets) available through the eQTL Catalog [24]. Association statistics for T1D were taken from a major GWAS that included 18,942 cases and 501,638 controls [27]. Results meeting the study-specific Bonferroni-corrected MR significance level—adjusted for the count of unique genes examined per dataset—were subjected to pairwise conditional colocalization (PWCoCo) [30] to investigate potential confounding by linkage disequilibrium.

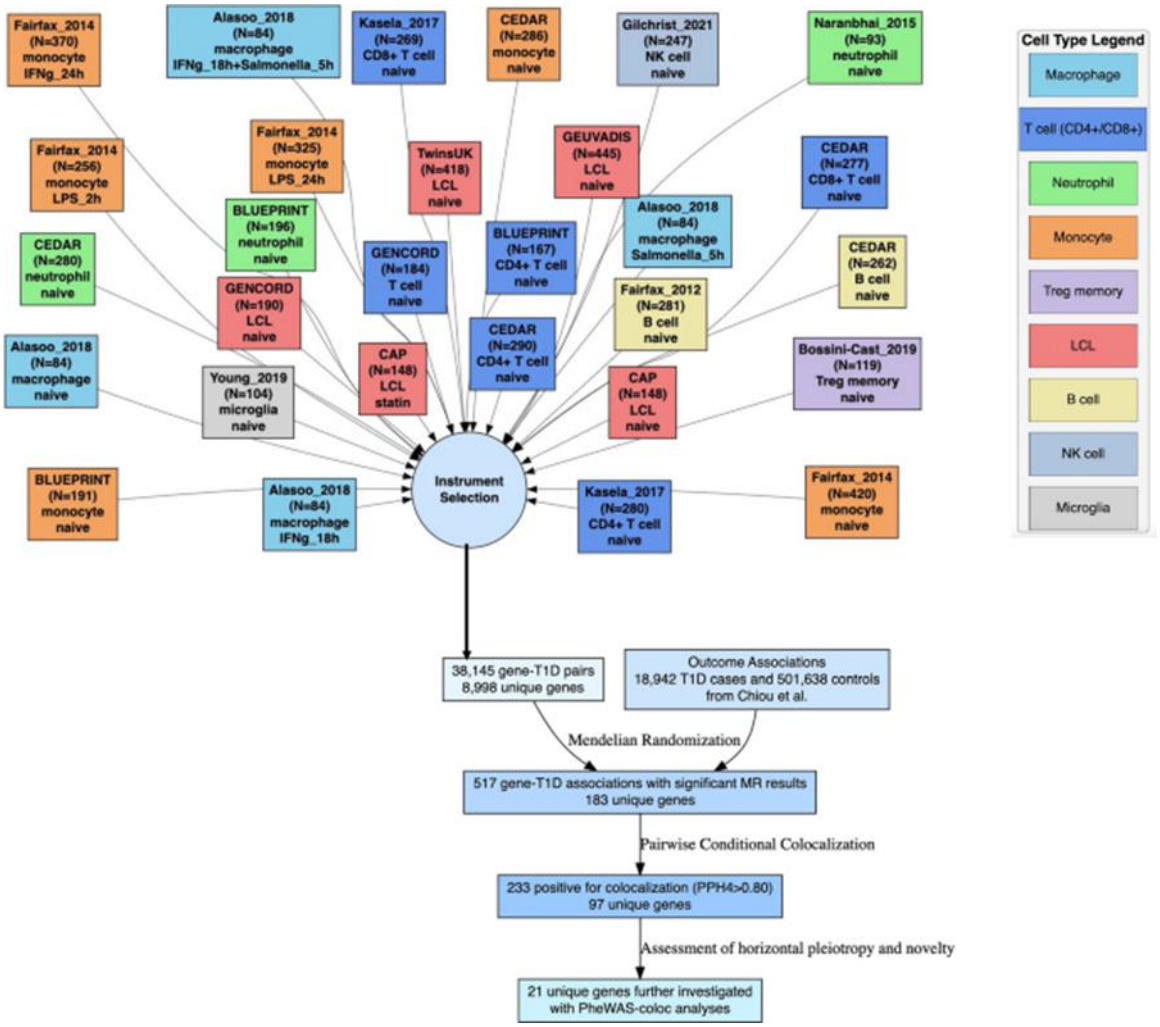

Figure 1. Overall study design.

The figure summarizes the 29 datasets utilized (specifying study source, sample size, immune cell type, and experimental conditions) for the identification of locally acting eQTLs (cis-eQTLs) as genetic instruments for gene expression. T1D association data originated from Chiou *et al*. [27]. Pairwise conditional colocalization was carried out using PWCoCo [30]. PheWAS-coloc indicates the phenome-wide association scans incorporating colocalization, as detailed in the Methods.

Mendelian randomization and colocalization

A total of 38,145 tests examining the relationship between genes and T1D were performed using data from the 29 available datasets. Most instruments (90.6%) relied on a single cis-eQTL, with 4.1% utilizing two variants, 4.3% using three, and 1.0% employing four or more. Only two genes possessed instruments present across every one of the 29 eQTL datasets, in contrast to 2,478 genes that were instrumented in merely a single dataset. Overall, 517 gene–T1D associations achieved statistical significance in the MR analyses. Of these, 233 also provided supportive evidence from colocalization (**Figure 2)**. When focusing on distinct genes, 8,998 were evaluated in total, 183 reached MR significance, and 97 demonstrated colocalization in addition **(Figure 2)**. Among the 97 colocalizing genes, 75 met the threshold based on marginal summary statistics, while conditional analysis identified a further 22. Alternative PPH4 thresholds resulted in 105 genes at > 0.7 and 69 genes at > 0.9.

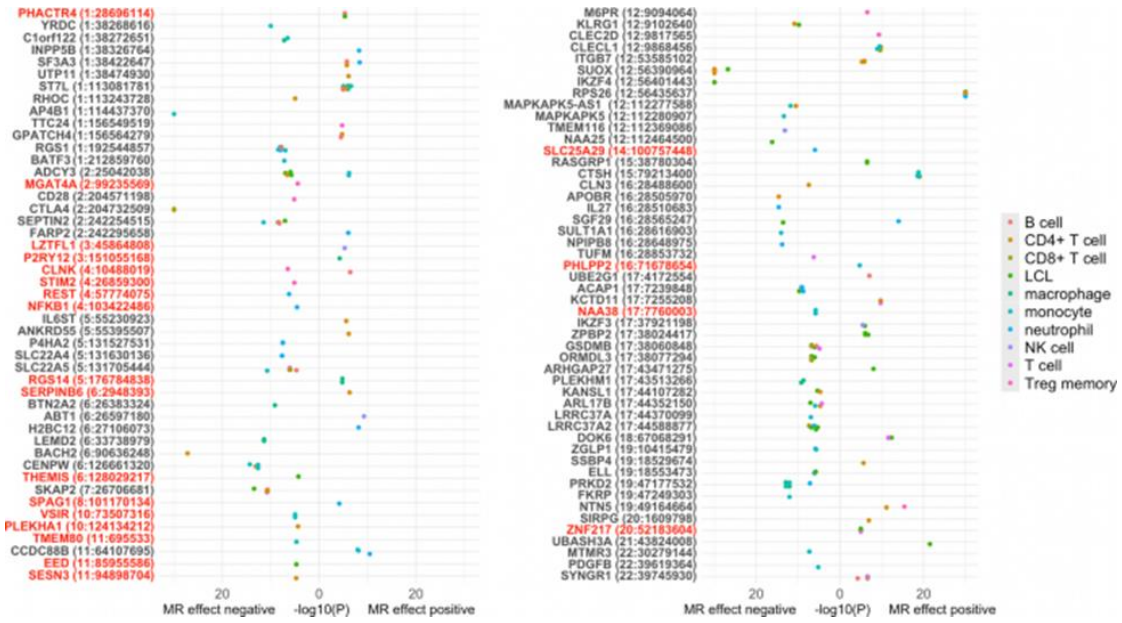

Figure 2. Bidirectional plot of associations meeting both Mendelian randomization significance and colocalization criteria.

The plot presents 233 gene–T1D pairs organized by unique gene (indicated by chromosome and transcription start site), with colors denoting the immune cell type source of the instrumental variants. MR analysis –log10(P-values) are displayed to the right for positive effect directions and to the left for negative directions, with values truncated at 30. Red-colored genes signify those that qualify as both the sole signal in their genomic region and novel. LCL, lymphoblastoid cell line. NK, natural killer. Treg memory, regulatory-T-cell memory cells.

Instruments developed from B cells and T cells showed some degree of enrichment for significant findings relative to other cell types. Among 1,757 unique genes tested in B cells, 17 (0.97%) satisfied both MR and colocalization requirements. The success rates were 0.95% in T cells, 0.77% in regulatory T-cell memory cells (Treg memory), 0.75% in neutrophils, 0.72% in lymphoblastoid cell lines (LCL), 0.67% in macrophages, 0.61% in monocytes, 0.36% in natural killer (NK) cells, and 0% in microglia. For genes passing MR significance, colocalization confirmation was most frequent in B cells (56.7%) and T cells (54.9%), and least frequent in NK cells (28.6%) and microglia (0%); **(Figure 3)**.

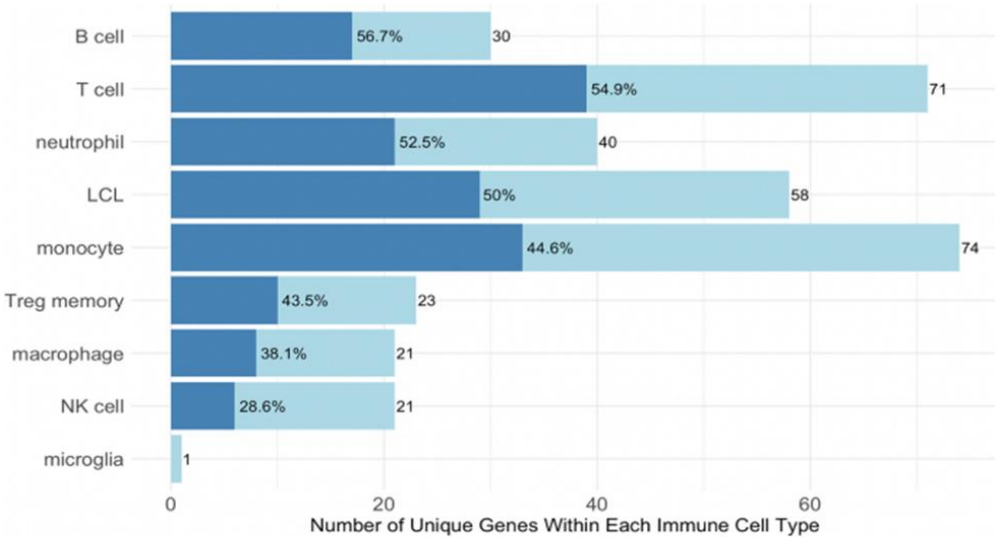

Figure 3. Genes attaining Mendelian randomization significance along with colocalization support, categorized by immune cell type.

Light blue bars display the count of unique genes reaching significance per cell type, whereas dark blue bars show the additional number that passed colocalization. Datasets derived from equivalent immune cell categories (such

as CD4+ T cells, CD8+ T cells, and total T cells) were aggregated when determining unique gene counts. LCL, lymphoblastoid cell line. NK, natural killer. Treg memory, regulatory-T-cell memory cells.

Prioritizing genes that are novel and the lone result in a region

Twenty-seven of the 97 colocalizing genes were judged novel, having escaped detection in prior T1D GWAS and MR reports. Among all 97 genes, 56 lay within 250 kb of another significant association, resulting in 41 isolated regional signals. The final set of 21 genes fulfilling both novelty and isolation criteria consisted of CLNK, EED, LZTFL1, MGAT4A, NAA38, NFKB1, PHACTR4, PHLPP2, PLEKHA1, P2RY12, REST, RGS14, SERPINB6, SESN3, SLC25A29, SPAG1, STIM2, THEMIS, TMEM80, VSIR, and ZNF217 **(Figure 2)**. No heterogeneity was detected in the variant-level estimates for these 21 genes. ZNF217 had instruments available in six datasets, all of which produced significant MR associations with T1D and five of which also colocalized (spanning B cells, LCL, T cells, and monocytes); **(Figure 4)**. Conversely, although 16 TMEM80–T1D tests were conducted, only one met both MR and colocalization standards. Of the six genes (CLNK, NAA38, PHACTR4, RGS14, VSIR, ZNF217) demonstrating multiple successful dataset-level signals **(Figure 4)**, directional consistency was observed in every instance except CLNK. For CLNK, instruments from Treg memory cells (rs4295251) and B cells (rs9291444) exhibited high correlation ($r^2 = 0.99$) but opposing influences on expression levels. Increased predicted CLNK expression from Treg memory cells linked to decreased T1D susceptibility, while the same from B cells linked to elevated susceptibility.

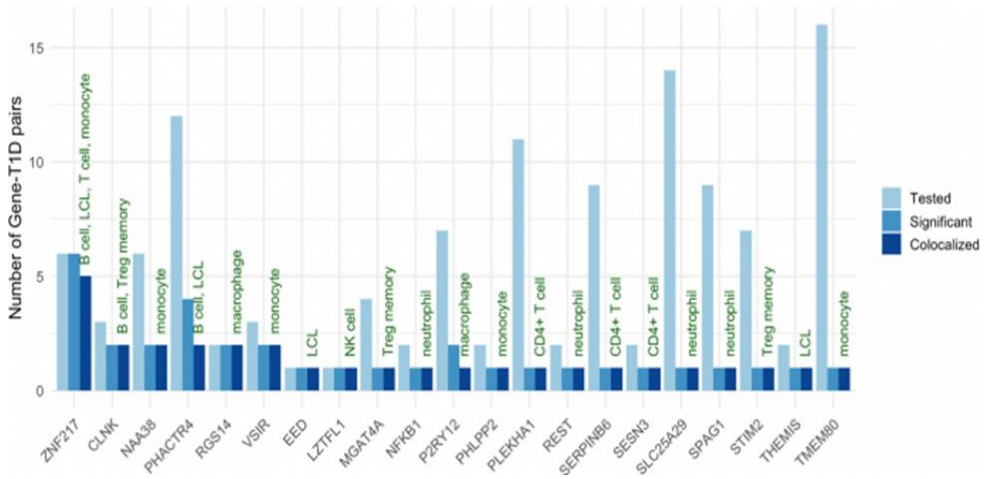

Figure 4. Prioritized genes shown across contributing eQTL datasets.

Light blue bars reflect the total gene–T1D MR tests performed (corresponding to datasets with available cis-eQTL instruments), medium blue bars mark those achieving MR significance, and dark blue bars highlight those that further colocalized. Green text identifies the distinct immune cell types providing instruments that succeeded in both MR and colocalization. LCL, lymphoblastoid cell line. NK, natural killer. Treg memory, regulatory-T-cell memory cells.

Phenome-wide scans of instrumental variants with conditional colocalization

PheWAS-coloc testing was carried out on instrumental variants belonging to the 21 prioritized genes. For genes with multiple successful datasets, the variant with the strongest T1D association was selected, with the exception of CLNK, for which separate queries were made using the cell-type-specific instruments due to opposing directions. Several genes displayed PheWAS-coloc signals ($P < 5 \times 10^{-5}$ and PPH4 > 0.8) involving other immune disorders, such as primary sclerosing cholangitis (CLNK, NFKB1), myasthenia gravis (SESN3), Graves' disease (CLNK, SESN3), rheumatoid arthritis (CLNK, SESN3), and asthma (EED, NFKB1, REST).

Key examples of PheWAS-coloc findings are depicted in **Figure 5** for VSIR (monocyte eQTLs), P2RY12 (macrophage eQTLs), CLNK (Treg memory and B-cell eQTLs), and SESN3 (T-cell eQTLs), where estimates correspond to elevated genetically predicted expression. Predicted VSIR levels associated with circulating C-X-C motif chemokine 16 (CXCL16, encoded on chromosome 17) and C-C motif chemokine 17 (CCL17, encoded on chromosome 16). Given their trans configuration and genomic distance from VSIR, these links likely represent downstream consequences of VSIR activity (vertical pleiotropy). VSIR further correlated positively with basal

cell carcinoma risk and negatively with abundance of a Blautia bacterial strain in stool [47]. P2RY12 demonstrated a positive relationship with Epstein-Barr virus EBNA-1 antibody levels and suggested horizontal pleiotropy through colocalization with the adjacent SUCNR1 gene; conditional analysis, however, reinforced P2RY12 as the primary signal. Both SESN3 and CLNK connected to various autoimmune traits. Elevated SESN3 prediction additionally linked to diminished CD3 expression on resting CD4 regulatory T cells and reduced CD28 on CD45RA+ CD4+ T cells.

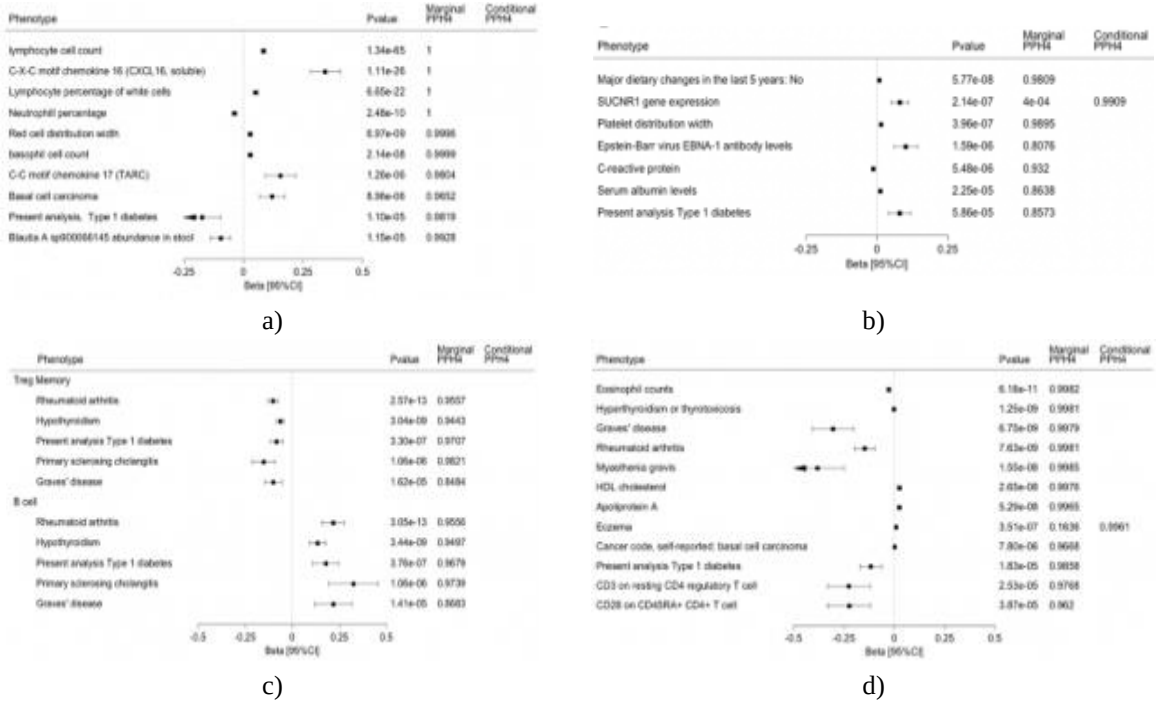

Figure 5. PheWAS-coloc findings for A VSIR, B P2RY12, C CLNK, and D SESN3.

Panels show A rs748113 (monocyte cis-eQTL for VSIR), B rs10513398 (macrophage cis-eQTL for P2RY12), C top panel rs9291444 (CLNK in regulatory T-cell memory cells) and bottom panel rs4295251 (CLNK in B cells), and D rs4409785 (T-cell cis-eQTL for SESN3). Beta values indicate MR estimates for higher predicted gene expression. Queries utilized the MRBase PheWAS platform for traits and omicscience.org for plasma proteins. Colocalization assessments (± 500 kb) between eQTLs and PheWAS data employed PWCoCo. Displayed associations satisfy $P < 5 \times 10^{-5}$ and PPH4 > 0.80. Conditional PPH4 refers to the probability after conditional analysis.

Replication in African ancestry individuals

MR analyses were subsequently conducted for the 21 prioritized genes within a cohort of individuals of African ancestry. Genetic instruments were drawn from whole-blood gene expression data reported by Kachuri *et al*. [42], while T1D outcome associations were derived from a meta-analysis of two publicly accessible GWAS [43, 44]. Among the 21 prioritized genes, 14 (NAA38, NFKB1, PHACTR4, PHLPP2, PLEKHA1, RGS14, SERPINB6, SESN3, SLC25A29, SPAG1, STIM2, TMEM80, VSIR, ZNF217) possessed a suitable cis-eQTL instrument. Four associations reached nominal significance at $P < 0.1$. Three of these (SESN3, TMEM80, and VSIR) displayed effect directions consistent with the main European-ancestry analysis, whereas one (SPAG1) showed discordance. Successful prevention of type 1 diabetes would substantially alleviate the profound effects of this condition on quality of life, long-term health complications, and survival for millions of people worldwide. In pursuit of candidate therapeutic targets, the current study applied genetic instruments originating from immune cells to conduct Mendelian randomization and colocalization analyses involving nearly 9,000 genes in relation to T1D risk. This effort identified 21 genes absent from previous genome-wide association and Mendelian randomization reports, while PheWAS-coloc evaluations yielded additional biological insights. Taken together, the results point toward promising new strategies for pharmacological development and repurposing aimed at averting or postponing T1D onset.

A particularly noteworthy finding concerns VSIR, the gene encoding the V-domain immunoglobulin suppressor of T cell activation (also termed GI24 or VISTA), given its established function in modulating immune responses. VISTA functions as an inhibitory immune checkpoint molecule primarily expressed on myeloid cells, where it dampens reactivity against self-antigens [48]. As a member of the B7 family of ligands—similar to programmed death-ligand 1 (PD-L1)—it attenuates activation of both CD4+ and CD8+ T cells [49]. Substantial preclinical evidence, largely from murine models, supports a protective role for VISTA in autoimmune conditions. Mice lacking VISTA (VISTA−/−) exhibit heightened susceptibility and severity of experimental autoimmune encephalomyelitis, systemic lupus erythematosus, and lupus-like disease when crossed with autoimmune-prone strains [50]. Treatment with anti-VISTA antibodies exacerbated disease progression in models of multiple sclerosis [51]. In contrast, VISTA agonism ameliorated pathology in mouse models of systemic lupus erythematosus [52] and psoriasis [53]. The present study extends these observations by demonstrating that the influence of VSIR on autoimmune risk, previously documented in animals, receives corroboration from human genetic evidence—a critical factor associated with translational success of targeted therapies. Moreover, pharmacological modulation of VISTA appears clinically feasible in the near future, as an oral small-molecule VISTA agonist (M351-0056) has already been described [53].

PheWAS-coloc results offered further mechanistic context for the VSIR association. Genetically predicted VSIR expression correlated with plasma concentrations of the chemokines CXCL16 and CCL17, both of which participate in immune cell trafficking and responses [54, 55] and may therefore mediate effects on T1D susceptibility. In addition, VSIR showed an inverse relationship with fecal abundance of a Blautia strain, consistent with emerging hypotheses implicating the gut–immune axis in T1D development [56]. Reduced Blautia levels have previously been observed in children diagnosed with T1D [56]. Finally, positive associations between elevated VSIR expression and basal cell carcinoma risk raise potential safety considerations for VISTA agonism, possibly reflecting impaired anti-tumor immune surveillance. This pattern aligns with increased basal cell carcinoma incidence following immunosuppressive therapy after renal transplantation [57] and parallels associations observed for CTLA4 [58], another negative regulator of T cell activation. Abatacept, a CTLA-4 fusion protein approved for rheumatoid arthritis and previously evaluated in T1D [59], did not show a significant increase in basal cell carcinoma incidence across nine randomized trials compared with placebo [60]. Thus, any such risk associated with VISTA agonism may similarly be modest. Nonetheless, these genetic findings underscore the importance of vigilant dermatologic monitoring should long-term VISTA-targeted therapies be pursued for T1D prevention.

The study also highlighted P2RY12 as a compelling novel target for T1D intervention. This receptor is already modulated by several licensed medications, such as clopidogrel, which are used to reduce thrombotic events following myocardial infarction or stroke. Although platelet-related pathways may not intuitively relate to T1D pathogenesis, the observed connection to Epstein-Barr virus EBNA-1 antibody levels is notable, given EBV's established links to multiple autoimmune disorders [61], including T1D [62]. Elevated EBNA-1 seropositivity has been documented in individuals at the time of T1D diagnosis relative to healthy controls [63]. Molecular mimicry between EBNA-1 and host proteins such as GlialCAM has been proposed as a contributor to autoimmunity in multiple sclerosis [64]. Existing functional studies align with the current genetic findings: P2RY12 deficiency protected mice against autoimmune hepatitis [65] and preserved islet integrity while improving glycemic control in models of induced T1D [66]. These observations warrant further exploration of P2RY12 inhibition, particularly in the context of EBV-related mechanisms in T1D.

Additional prioritized genes, including SESN3 and CLNK, also influence susceptibility to other autoimmune conditions. Sestrin 3, encoded by SESN3, serves as a stress-responsive protein that safeguards cells against oxidative injury. Fine-mapping in human T cells has indicated that the rheumatoid arthritis risk signal overlapping with one for T1D at the SESN3 locus operates through chromatin interactions that control SESN3 expression [67]. The present results suggest that SESN3 may downregulate surface levels of CD3—a critical co-receptor for T cell activation and the target of teplizumab, which is approved to delay progression to stage 3 T1D [4]—as well as CD28, an important co-stimulatory molecule. Notably, genetically predicted CLNK expression displayed opposing directional effects on autoimmune risk, including T1D, depending on whether instruments were derived from Treg memory cells or B cells. As CLNK participates in immune signaling cascades, additional research will be required to clarify its context-dependent contributions to autoimmunity.

Key strengths of this work include the utilization of eQTL instruments obtained from purified immune cell populations, which are centrally involved in T1D pathogenesis. The application of conditional colocalization to

both T1D and PheWAS outcomes provided rigorous assessment of potential linkage disequilibrium confounding. Finally, systematic follow-up evaluations ensured careful consideration of novelty, horizontal pleiotropy, and possible on-target or mediating effects.

Several limitations should be acknowledged. The primary analyses were conducted in European-ancestry populations, and the African-ancestry replication attempt was statistically underpowered. The replication outcome dataset contained only 6,860 T1D cases of African ancestry, the large majority (94%) originating from an electronic health record-based GWAS prone to diagnostic misclassification. For instance, ICD codes for T1D demonstrated a positive predictive value of only 41% in one major healthcare system, with most coded cases actually representing type 2 diabetes [68]. By comparison, the primary GWAS meta-analysis by Chiou *et al*. incorporated more stringently phenotyped participants. Furthermore, African-ancestry instruments were limited to whole-blood eQTLs, as no public resources currently exist for immune-cell eQTLs in this population. These considerations emphasize the continued necessity for expanded genetic research in diverse, non-European populations, including in T1D.

Additional constraints merit mention. Gene expression levels do not always correspond directly to functional protein abundance, yet proteins constitute the primary targets of most therapeutic agents. For example, compensatory upregulation of gene expression in response to dysfunctional protein variants could produce misleading genetic associations. Moreover, summary-level MR methods assume linear relationships between exposure and outcome, which may not fully capture potential non-linear effects of gene expression on T1D risk.

Conclusion

This study uncovered multiple novel genes exhibiting putative causal relationships with T1D that represent promising candidates for therapeutic intervention to prevent or postpone disease onset. Notable examples include VSIR and P2RY12, both of which are supported by prior functional evidence in autoimmune models. The addition of human genetic support is particularly valuable, as it enhances the likelihood of successful clinical development. Continued investigation into therapeutic modulation of VSIR, P2RY12, and the remaining prioritized genes is therefore justified.

Acknowledgments: None

Conflict of Interest: None

Financial Support: None

Ethics Statement: None

References

1. Quattrin T, Mastrandrea LD, Walker LSK. Type 1 diabetes. Lancet. 2023;401(10394):2149–62.
2. Sussman M, Benner J, Haller MJ, Rewers M, Griffiths R. Estimated lifetime economic burden of type 1 diabetes. Diabetes Technol Ther. 2020;22(2):121–30. doi:10.1089/dia.2019.0398
3. Gregory GA, Robinson TIG, Linklater SE, Wang F, Colagiuri S, de Beaufort C, et al. Global incidence, prevalence, and mortality of type 1 diabetes in 2021 with projection to 2040: A modelling study. Lancet Diabetes Endocrinol. 2022;10(10):741–60.
4. Herold KC, Bundy BN, Long SA, Bluestone JA, DiMeglio LA, Dufort MJ, et al. An anti-CD3 antibody, teplizumab, in relatives at risk for type 1 diabetes. N Engl J Med. 2019;381(7):603–13.
5. Schmidt AF, Finan C, Gordillo-Marañón M, Asselbergs FW, Freitag DF, Patel RS, et al. Genetic drug target validation using Mendelian randomisation. Nat Commun. 2020;11(1):3255.
6. Smith GD, Ebrahim S. Mendelian randomization': can genetic epidemiology contribute to understanding environmental determinants of disease? Int J Epidemiol. 2003;32(1):1–22.
7. Nelson MR, Tipney H, Painter JL, Shen J, Nicoletti P, Shen Y, et al. The support of human genetic evidence for approved drug indications. Nat Genet. 2015;47(8):856–60.

8. Yazdanpanah N, Yazdanpanah M, Wang Y, Forgetta V, Pollak M, Polychronakos C, et al. Clinically relevant circulating protein biomarkers for type 1 diabetes: evidence from a two-sample Mendelian randomization study. Diabetes Care. 2022;45(1):169–77.
9. Lu T, Manousaki D, Sun L, Paterson AD. Integrative Proteogenomic Analyses Provide Novel Interpretations of Type 1 Diabetes Risk Loci Through Circulating Proteins. Diabetes. 2025;74(4):630–9.
10. Alasoo K, Rodrigues J, Mukhopadhyay S, Knights AJ, Mann AL, Kundu K, et al. Shared genetic effects on chromatin and gene expression indicate a role for enhancer priming in immune response. Nat Genet. 2018;50(3):424–31.
11. Bossini-Castillo L, Glinos DA, Kunowska N, Golda G, Lamikanra AA, Spitzer M, et al. Immune disease variants modulate gene expression in regulatory CD4(+) T cells. Cell Genom. 2022;2(4).
12. Buil A, Brown AA, Lappalainen T, Viñuela A, Davies MN, Zheng HF, et al. Gene-gene and gene-environment interactions detected by transcriptome sequence analysis in twins. Nat Genet. 2015;47(1):88–91.
13. Chen L, Ge B, Casale FP, Vasquez L, Kwan T, Garrido-Martín D, et al. Genetic drivers of epigenetic and transcriptional variation in human immune cells. Cell. 2016;167(5):1398-414.e24.
14. Fairfax BP, Humburg P, Makino S, Naranbhai V, Wong D, Lau E, et al. Innate immune activity conditions the effect of regulatory variants upon monocyte gene expression. Science. 2014;343(6175):1246949.
15. Fairfax BP, Makino S, Radhakrishnan J, Plant K, Leslie S, Dilthey A, et al. Genetics of gene expression in primary immune cells identifies cell type-specific master regulators and roles of HLA alleles. Nat Genet. 2012;44(5):502–10.
16. Gilchrist JJ, Makino S, Naranbhai V, Sharma PK, Koturan S, Tong O, et al. Natural killer cells demonstrate distinct eQTL and transcriptome-wide disease associations, highlighting their role in autoimmunity. Nat Commun. 2022;13(1):4073.
17. Gutierrez-Arcelus M, Lappalainen T, Montgomery SB, Buil A, Ongen H, Yurovsky A, et al. Passive and active DNA methylation and the interplay with genetic variation in gene regulation. Elife. 2013;2:e00523. doi:10.7554/eLife.00523
18. Kasela S, Kisand K, Tserel L, Kaleviste E, Remm A, Fischer K, et al. Pathogenic implications for autoimmune mechanisms derived by comparative eQTL analysis of CD4+ versus CD8+ T cells. PLoS Genet. 2017;13(3):e1006643. doi:10.1371/journal.pgen.1006643
19. Lappalainen T, Sammeth M, Friedländer MR, t Hoen PA, Monlong J, Rivas MA, et al. Transcriptome and genome sequencing uncovers functional variation in humans. Nature. 2013;501(7468):506–11.
20. Momozawa Y, Dmitrieva J, Théâtre E, Deffontaine V, Rahmouni S, Charloteaux B, et al. IBD risk loci are enriched in multigenic regulatory modules encompassing putative causative genes. Nat Commun. 2018;9(1):2427.
21. Naranbhai V, Fairfax BP, Makino S, Humburg P, Wong D, Ng E, et al. Genomic modulators of gene expression in human neutrophils. Nat Commun. 2015;6:7545.
22. Theusch E, Chen YI, Rotter JI, Krauss RM, Medina MW. Genetic variants modulate gene expression statin response in human lymphoblastoid cell lines. BMC Genomics. 2020;21(1):555.
23. Young AMH, Kumasaka N, Calvert F, Hammond TR, Knights A, Panousis N, et al. A map of transcriptional heterogeneity and regulatory variation in human microglia. Nat Genet. 2021;53(6):861–8.
24. Kerimov N, Tambets R, Hayhurst JD, Rahu I, Kolberg P, Raudvere U, et al. eQTL catalogue 2023: new datasets, X chromosome QTLs, and improved detection and visualisation of transcript-level QTLs. PLoS Genet. 2023;19(9):e1010932. doi:10.1371/journal.pgen.1010932
25. Stacey D, Gaziano L, Eldi P, Toben C, Benyamin B, Lee SH, et al. A transcriptome-wide Mendelian randomization study in isolated human immune cells highlights risk genes involved in viral infections and potential drug repurposing opportunities for schizophrenia. Am J Med Genet B Neuropsychiatr Genet. 2025;198(6):19–31.
26. Zheng J, Haberland V, Baird D, Walker V, Haycock PC, Hurle MR, et al. Phenome-wide Mendelian randomization mapping the influence of the plasma proteome on complex diseases. Nat Genet. 2020;52(10):1122–31.
27. Chiou J, Geusz RJ, Okino ML, Han JY, Miller M, Melton R, et al. Interpreting type 1 diabetes risk with genetics and single-cell epigenomics. Nature. 2021;594(7863):398–402.

28. Hemani G, Zheng J, Elsworth B, Wade KH, Haberland V, Baird D, et al. The MR-Base platform supports systematic causal inference across the human phenome. Elife. 2018. doi:10.7554/eLife.34408
29. Burgess S, Mason AM, Grant AJ, Slob EAW, Gkatzionis A, Zuber V, et al. Using genetic association data to guide drug discovery and development: review of methods and applications. Am J Hum Genet. 2023;110(2):195–214.
30. Robinson JW, Hemani G, Babaei MS, Huang Y, Baird DA, Tsai EA, et al. An efficient and robust tool for colocalisation: Pair-wise Conditional and Colocalisation (PWCoCo). bioRxiv. 2022.
31. Giambartolomei C, Vukcevic D, Schadt EE, Franke L, Hingorani AD, Wallace C, et al. Bayesian test for colocalisation between pairs of genetic association studies using summary statistics. PLoS Genet. 2014;10(5):e1004383. doi:10.1371/journal.pgen.1004383
32. Yang J, Lee SH, Goddard ME, Visscher PM. GCTA: A tool for genome-wide complex trait analysis. Am J Hum Genet. 2011;88(1):76–82. doi:10.1016/j.ajhg.2010.11.011
33. Bycroft C, Freeman C, Petkova D, Band G, Elliott LT, Sharp K, et al. The UK Biobank resource with deep phenotyping and genomic data. Nature. 2018;562(7726):203–9.
34. O'Connell J, Sharp K, Shrine N, Wain L, Hall I, Tobin M, et al. Haplotype estimation for biobank-scale data sets. Nat Genet. 2016;48(7):817–20.
35. Forgetta V, Manousaki D, Istomine R, Ross S, Tessier MC, Marchand L, et al. Rare genetic variants of large effect influence risk of type 1 diabetes. Diabetes. 2020;69(4):784–95.
36. Barrett JC, Clayton DG, Concannon P, Akolkar B, Cooper JD, Erlich HA, et al. Genome-wide association study and meta-analysis find that over 40 loci affect risk of type 1 diabetes. Nat Genet. 2009;41(6):703–7.
37. Robertson CC, Inshaw JRJ, Onengut-Gumuscu S, Chen WM, Santa Cruz DF, Yang H, et al. Fine-mapping, trans-ancestral and genomic analyses identify causal variants, cells, genes and drug targets for type 1 diabetes. Nat Genet. 2021;53(7):962–71.
38. Onengut-Gumuscu S, Chen WM, Burren O, Cooper NJ, Quinlan AR, Mychaleckyj JC, et al. Fine mapping of type 1 diabetes susceptibility loci and evidence for colocalization of causal variants with lymphoid gene enhancers. Nat Genet. 2015;47(4):381–6.
39. Pietzner M, Wheeler E, Carrasco-Zanini J, Cortes A, Koprulu M, Wörheide MA, et al. Mapping the proteo-genomic convergence of human diseases. Science. 2021;374(6569):eabj1541.
40. Pietzner M, Wheeler E, Carrasco-Zanini J, Raffler J, Kerrison ND, Oerton E, et al. Genetic architecture of host proteins involved in SARS-CoV-2 infection. Nat Commun. 2020;11(1):6397.
41. Võsa U, Claringbould A, Westra HJ, Bonder MJ, Deelen P, Zeng B, et al. Large-scale cis- and trans-eQTL analyses identify thousands of genetic loci and polygenic scores that regulate blood gene expression. Nat Genet. 2021;53(9):1300–10.
42. Kachuri L, Mak ACY, Hu D, Eng C, Huntsman S, Elhawary JR, et al. Gene expression in African Americans, Puerto Ricans and Mexican Americans reveals ancestry-specific patterns of genetic architecture. Nat Genet. 2023;55(6):952–63.
43. Verma A, Huffman JE, Rodriguez A, Conery M, Liu M, Ho YL, et al. Diversity and scale: Genetic architecture of 2068 traits in the VA Million Veteran Program. Science. 2024;385(6706):eadj1182.
44. Michalek DA, Tern C, Zhou W, Robertson CC, Farber E, Campolieto P, et al. A multi-ancestry genome-wide association study in type 1 diabetes. Hum Mol Genet. 2024;33(11):958–68.
45. Willer CJ, Li Y, Abecasis GR. METAL: fast and efficient meta-analysis of genomewide association scans. Bioinformatics. 2010;26(17):2190–1.
46. Onengut-Gumuscu S, Concannon P, Akolkar B, Erlich HA, Julier C, Morahan G, et al. Type 1 Diabetes Genetics Consortium. J Clin Endocrinol Metab. 2025;110(6):1505–13.
47. Liu X, Mao B, Gu J, Wu J, Cui S, Wang G, et al. Blautia-a new functional genus with potential probiotic properties? Gut Microbes. 2021;13(1):1–21.
48. Lines JL, Pantazi E, Mak J, Sempere LF, Wang L, O'Connell S, et al. VISTA is an immune checkpoint molecule for human T cells. Cancer Res. 2014;74(7):1924–32.
49. Zheng M, Zhang Z, Yu L, Wang Z, Dong Y, Tong A, et al. Immune-checkpoint protein VISTA in allergic, autoimmune disease and transplant rejection. Front Immunol. 2023;14:1194421.
50. Wang L, Le Mercier I, Putra J, Chen W, Liu J, Schenk AD, et al. Disruption of the immune-checkpoint VISTA gene imparts a proinflammatory phenotype with predisposition to the development of autoimmunity. Proc Natl Acad Sci U S A. 2014;111(41):14846–51.

51. Wang L, Rubinstein R, Lines JL, Wasiuk A, Ahonen C, Guo Y, et al. VISTA, a novel mouse Ig superfamily ligand that negatively regulates T cell responses. J Exp Med. 2011;208(3):577–92.
52. Yang L, Zhang T, Wang P, Chen W, Liu W, He X, et al. Imatinib and M351–0056 enhance the function of VISTA and ameliorate the development of SLE via IFN-I and noncanonical NF-κB pathway. Cell Biol Toxicol. 2023;39(6):3287–304.
53. Hu X, Qie C, Jiang J, Xie X, Chen W, Liu W, et al. M351-0056 is a novel low MW compound modulating the actions of the immune-checkpoint protein VISTA. Br J Pharmacol. 2021;178(6):1445–58.
54. Bao N, Fu B, Zhong X, Jia S, Ren Z, Wang H, et al. Role of the CXCR6/CXCL16 axis in autoimmune diseases. Int Immunopharmacol. 2023;121:110530.
55. Lupancu TJ, Eivazitork M, Hamilton JA, Achuthan AA, Lee KM. CCL17/TARC in autoimmunity and inflammation-not just a T-cell chemokine. Immunol Cell Biol. 2023;101(7):600–9.
56. Dedrick S, Sundaresh B, Huang Q, Brady C, Yoo T, Cronin C, et al. The role of gut microbiota and environmental factors in type 1 diabetes pathogenesis. Front Endocrinol (Lausanne). 2020;11:78.
57. Rangwala S, Tsai KY. Roles of the immune system in skin cancer. Br J Dermatol. 2011;165(5):953–65.
58. Choquet H, Jiang C, Yin J, Kim Y, Hoffmann TJ, Jorgenson E, et al. Multi-ancestry genome-wide meta-analysis identifies novel basal cell carcinoma loci and shared genetic effects with squamous cell carcinoma. Commun Biol. 2024;7(1):33.
59. Russell WE, Bundy BN, Anderson MS, Cooney LA, Gitelman SE, Goland RS, et al. Abatacept for delay of type 1 diabetes progression in stage 1 relatives at risk: a randomized, double-masked, controlled trial. Diabetes Care. 2023;46(5):1005–13. doi:10.2337/dc22-2200
60. Simon TA, Dong L, Suissa S, Michaud K, Pedro S, Hochberg M, et al. Abatacept and non-melanoma skin cancer in patients with rheumatoid arthritis: a comprehensive evaluation of randomised controlled trials and observational studies. Ann Rheum Dis. 2024;83(2):177–83.
61. Robinson WH, Younis S, Love ZZ, Steinman L, Lanz TV. Epstein-Barr virus as a potentiator of autoimmune diseases. Nat Rev Rheumatol. 2024;20(11):729–40.
62. Harley JB, Chen X, Pujato M, Miller D, Maddox A, Forney C, et al. Transcription factors operate across disease loci, with EBNA2 implicated in autoimmunity. Nat Genet. 2018;50(5):699–707.
63. Bian X, Wallstrom G, Davis A, Wang J, Park J, Throop A, et al. Immunoproteomic profiling of antiviral antibodies in new-onset type 1 diabetes using protein arrays. Diabetes. 2016;65(1):285–96.
64. Lanz TV, Brewer RC, Ho PP, Moon JS, Jude KM, Fernandez D, et al. Clonally expanded B cells in multiple sclerosis bind EBV EBNA1 and GlialCAM. Nature. 2022;603(7900):321–7.
65. Zhuang W, Liu X, Liu G, Lv J, Qin H, Wang C, et al. Purinergic receptor P2Y12 boosts autoimmune hepatitis through hexokinase 2-dependent glycolysis in T cells. Int J Biol Sci. 2023;19(11):3576–94.
66. Qin C, Zhou J, Gao Y, Lai W, Yang C, Cai Y, et al. Critical role of P2Y(12) receptor in regulation of Th17 differentiation and experimental autoimmune encephalomyelitis pathogenesis. J Immunol. 2017;199(1):72–81.
67. Shi C, Zhao D, Butler J, Frantzeskos A, Rossi S, Ding J, et al. Multi-omics analysis in primary T cells elucidates mechanisms behind disease-associated genetic loci. Genome Biol. 2025;26(1):26.
68. Klompas M, Eggleston E, McVetta J, Lazarus R, Li L, Platt R. Automated detection and classification of type 1 versus type 2 diabetes using electronic health record data. Diabetes Care. 2013;36(4):914–21.