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Genetic immune and inflammatory markers associated with diabetes in solid organ transplant recipients

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New-onset diabetes mellitus after transplantation (NODAT) is a complication following solid organ transplantation (SOT) and may be related to immune or inflammatory responses. We investigated whether single nucleotide polymorphisms (SNPs) within 158 immune- or inflammation-related genes contribute to NODAT in SOT recipients. The association between 263 SNPs and NODAT was investigated in a discovery sample of SOT recipients from the Swiss Transplant Cohort Study (STCS, $n_1 = 696$).

Abbreviations: DIAGRAM, DIAbetes Genetics Replication and Meta-Analysis; FBG, fasting blood glucose; GAMM, generalized additive mixed model; GIANT, Genetic Investigation of ANthropometric Traits; HCV, hepatitis C virus; HWE, Hardy-Weinberg equilibrium; IFNγ, interferon gamma; IL-6, interleukin 6; MAGIC, Meta-Analyses of Glucose and Insulin-related Traits; NODAT, new-onset diabetes mellitus after transplantation; SNP, single-nucleotide polymorphism; SOT, solid organ transplantation; STCS, Swiss Transplant Cohort Study; TNF-α, tumor necrosis factor alpha; VODI, veno-occlusive disease with immunodeficiency.

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Positive results were tested in a first STCS replication sample (n_2 = 489) and SNPs remaining significant after multiple test corrections were tested in a second SOT replication sample (n_3 = 156). Associations with diabetic traits were further tested in several large general population-based samples (n > 480 000). Only *SP110 rs2114592C>T* remained associated with NODAT in the STCS replication sample. Carriers of r*s2114592-TT* had 9.9 times (95% confidence interval [CI]: 3.22-30.5, *P* = .00006) higher risk for NODAT in the combined STCS samples (n = 1184). *rs2114592C>T* was further associated with NODAT in the second SOT sample (odds ratio: 4.8, 95% CI: 1.55-14.6, *P* = .006). On the other hand, *SP110 rs2114592C>T* was not associated with diabetic traits in population-based samples, suggesting a specific gene-environment interaction, possibly due to the use of specific medications (ie, immunosuppressants) in transplant patients and/or to the illness that may unmask the gene effect.

KEYWORDS
clinical research/practice, diabetes, genetics, new onset/posttransplant

1 | INTRODUCTION

New-onset diabetes mellitus after transplantation (NODAT) is a serious complication following solid organ transplantation (SOT)[1] affecting 2%-53% of transplanted patients.[2] It is considered that NODAT alone is associated with increased 3-year mortality and a significant reduction in graft survival.[2] NODAT is also associated with increased cardiovascular events, infectious complications, and graft loss,[3] which justifies the great interest in understanding the underlying mechanisms and risk factors contributing to the development of this disease. Obesity, increased age, hepatitis C virus (HCV) infection, family history of diabetes mellitus, and African-American or Hispanic descent are among the risk factors of NODAT.[4] Immunosuppressive drugs such as corticosteroids and calcineurin inhibitors, in particular tacrolimus, are also associated with an increased risk of NODAT.[1,2]

Both infections and NODAT are frequent complications in SOT and may be related to altered innate immune responses.[5] The innate immune system is the first line of defense against infections with offending pathogens. Cytokines released from immune cells can elicit local and systemic inflammatory responses that can contribute to the development of type 2 diabetes mellitus by enhancing insulin resistance. Several proinflammatory cytokines, such as tumor necrosis factor alpha (TNF-α), interleukin 6 (IL-6), and interferon gamma (IFNγ), were previously shown to decrease the expression of insulin-sensitive glucose transporters, insulin signaling, and to promote insulin resistance.[6] Also, in the absence of infection, obesity (pre- and posttransplant) is associated with chronic low-grade inflammation driven by adipokines derived from adipose tissue, which can lead to insulin resistance[7] and the development of NODAT.[8]

Genome-wide association studies conducted to date explain only 10% of type 2 diabetes mellitus heritability and more diabetes mellitus susceptibility genes remain to be discovered.[9] Regarding the genetics of NODAT, little is known and many inconsistent associations have been reported and are reviewed.[10] Therefore, we aimed to examine whether single nucleotide polymorphisms (SNPs) located in different genes mainly implicated in inflammation or in the immune responses to infectious diseases may contribute to the development of NODAT in SOT recipients. As control, analyses were extended to samples of the general population.

2 | MATERIALS AND METHODS

2.1 | Sample description

2.1.1 | The Swiss Transplant Cohort Study (discovery and first replication samples)

The Swiss Transplant Cohort Study (STCS) is an ongoing prospective multicenter nationwide cohort project with an extensive and structured data collection.[11] Full description of this nationwide Swiss cohort (Basel, Bern, Geneva, Lausanne, St. Gallen, and Zurich) is published elsewhere.[12] Briefly, all SOT recipients in Switzerland are prospectively registered since May 2008. All patients with an allotransplant are included and are followed in their respective centers. Clinical data are collected at baseline, 6 and 12 months, and yearly thereafter. For the discovery STCS cohort, patients transplanted from May 2008 to May 8, 2011 were included in the analyses (n = 1294). Patients from the STCS transplanted from May 9, 2011 to May 2013 were included in the first replication cohort (n = 759). Nonwhite patients and patients who were diagnosed with glucose intolerance or diabetes mellitus before transplantation, as reported in the medical files, were excluded from NODAT analyses. Recipients younger than 18 years old and recipients with multiple

organ transplantation were excluded from our study. If a patient received more than 1 transplantation during the inclusion period, only data from the first SOT were included in the analyses. NODAT was diagnosed if a patient needed antidiabetic treatments following transplantation or if such new metabolic event was reported in the case report forms. For the combined STCS sample, new-onset hyperlipidemia was diagnosed if patients needed hypolipidemic treatment post-SOT. Patients with previous hyperlipidemia, as reported in the medical files, were excluded from the new-onset hyperlipidemia analyses. Blood samples were obtained from all SOT recipients at the time of transplantation. The study was approved by the corresponding ethics committee and all patients gave their written informed consent to participate in the study.

2.1.2 | Second replication SOT sample

A full description of this cohort is published elsewhere.[12-14] Briefly, a total of 200 patients were enrolled between 2003 and 2005 from the outpatient clinic of the Transplant Center of the University Hospital of Lausanne, Switzerland. Clinical data were collected retrospectively from the medical files at the time of transplantation, at 1, 3, 6, 9, and 12 months posttransplantation, and yearly thereafter until 5 years posttransplantation. Nonwhite patients and patients who had glucose intolerance or diabetes mellitus before transplantation, as reported in the medical files, were excluded from the present study. NODAT was diagnosed if a patient needed antidiabetic treatment (either insulin or oral antidiabetic agents) following transplantation or fulfilled the criteria given by the World Health Organization (WHO) and American Diabetes Association (ADA) consensuses,[15] including fasting blood glucose (FBG) ≥ 7.0 mmol/L on ≥2 occasions and/or 2-hour plasma glucose ≥11.1 mmol/L during an oral glucose tolerance test. The study was approved by the ethics committee of the University of Lausanne, and all patients gave their written informed consent to participate in the study.

Overall, 1294, 759, and 200 patients from the STCS discovery sample, the first STCS replication sample, and the second SOT replication sample, respectively, were included in the study. After excluding patients younger than 18 years of age, patients with multiple organ transplantation, patients with previous diabetes mellitus or prediabetes mellitus, and keeping patients with both clinical and genetic data available, 696, 489, and 156 patients, respectively, remained included in the NODAT analysis.

2.1.3 | Population-based samples

Significant SNPs were tested for association with diabetes mellitus development in the general white population (UK biobank [6117 type 2 diabetes mellitus cases and 109 942 controls], the MAGIC [Meta-Analyses of Glucose and Insulin-related traits] consortium [n = 46 186], and the DIAGRAM [DIAbetes Genetics Replication and Meta-analysis] consortium [n = 69 033; 12 171 T2D cases and 56 862 controls]). Associations with obesity markers in the GIANT (Genetic Investigation of ANthropometric Traits) consortium (n = 250 596), lipid markers in the Genome Wide Associations Scans for Total Cholesterol, HDL-C, LDL-C, and triglycerides (n > 100 000) and with different metabolic parameters in the Cohorte Lausannoise (CoLaus, n = 5338) were also tested. More information is given in the Supporting Information.

2.2 | Polymorphism selection

Two hundred eighty-seven SNPs within 158 different genes (Table S1) implicated in the immune response to infectious pathogens or inflammation were selected from customized 384 Golden Gate Genotyping Assay (Illumina, San Diego, CA). More information about Polymorphism selection and genotyping is given in the Supporting Information.

2.3 | Statistical analyses

Quantitative data are presented as median and range unless otherwise mentioned, while qualitative data are presented as frequency and percentage. For association studies, the χ^2 test or the Fisher exact test for binomial variables were used. Differences in allele and genotype frequencies as well as deviation from Hardy-Weinberg equilibrium (HWE) were assessed using the χ^2 test. Data were analyzed using Stata 13 (StataCorp, College Station, TX) and R version 2.13.0 software (http://www.R-project.org).

2.3.1 | STCS samples (discovery and first replication sample)

NODAT in the STCS samples was analyzed using logistic regression models adjusted for recipient's age at transplantation, sex, HCV status, baseline body mass index (BMI), and the type of calcineurin inhibitor (variables identified through the univariate analysis with $P < .10$, Table 1). Associations between NODAT and selected SNPs with $P < .01$ in the STCS discovery sample were retained and analyzed in the STCS replication sample and in other cohorts as described. No correction for multiple testing was applied for the discovery sample because it was only used to explore associations between the selected SNPs and NODAT, and these associations were only considered significant if replicated in another cohort. Bonferroni correction for multiple testing, however, was applied for the replication cohorts.

In the combined STCS sample, logistic regression models adjusted for recipient's age at transplantation, sex, and baseline BMI were used to analyze other metabolic parameters including new-onset hyperlipidemia after transplantation. Because of nonlinearity of BMI and HDL-cholesterol models and the absence of any linear transformation, the association between the SNPs with BMI and HDL-cholesterol levels was assessed by fitting a generalized additive mixed model (GAMM) to allow a smooth trend for the response in time based on multiple observations for each patient (using a thin plate regression spline basis) adjusting for recipient's age at transplantation, sex, and baseline BMI (also adjusted for the use of hypolipidemic drugs for analysis of HDL-cholesterol). GAMMs were

TABLE 1 Clinical characteristics of the solid organ transplantation discovery sample (Swiss Transplant Cohort Study first sample)

Characteristic	Total	Non-NODAT	NODAT	*P* value
Sample size (%)	696	544 (78.2)	152 (21.8)	
Age at transplantation (y), median (range)	52 (18-79)	51 (18-79)	56 (19-73)	.0003
Recipient sex (male) (%)	65.6	63.7	72.4	.05
Living donor (%)	32.7	33.7	29.0	.27
Donor age (y) median (range)	52 (1-86)	52 (1-85)	54 (14-86)	.73
Donor sex (male) (%)	53.3	54.8	48.0	.14
Recipients anti-HCV (% of HCV positive)	7.7	6.5	11.8	.03
Organ (%)				
Kidney	69.1	70.6	63.8	.15
Liver	15.5	15.1	17.1	
Lung	9.2	9.2	9.2	
Heart	6.2	5.1	9.9	
Body mass index (kg/m^2), median (range)				
Pretransplant	24.4 (13.7-41.2)	24.2 (13.7-39.1)	25.8 (15.2-41.2)	.0001
One-y follow-up	25.1 (15.4-44.4)	24.7 (15.4-44.4)	26.6 (17.3-44.3)	.0005
Calcineurin inhibitors, (%)				.004
Tacrolimus	66.5	64.1	75.0	
Cyclosporine	26.2	29.1	15.8	
None	7.3	6.8	9.2	

HCV, hepatitis C virus; NODAT, new-onset diabetes mellitus after transplantation.

fitted using the mgcv package of R (settings were fixed at package defaults). In order to be more conservative, the uncertainty of estimated parameters was assessed by 1000 bootstraps at the subject level. Linear mixed models were used for total blood cholesterol and LDL-cholesterol analyses. These models were adjusted for recipient's age at transplantation, sex, baseline BMI, and hypolipidemic drug intake.

2.3.2 | Second replication SOT sample

Logistic regression models were applied for NODAT analyses adjusted for recipient's age at transplantation, sex, baseline BMI, and type of calcineurin inhibitors.

2.3.3 | Population-based samples

SNPs significantly associated with different metabolic traits in SOT samples were analyzed in population-based samples. Multivariate linear regression with allele dosage in which potential confounding factors such as age, sex, batch effect, 5 ancestry principal components, and smoking status were added as covariates. FBG, oral glucose tolerance test, the surrogate estimates of beta cell function (homeostatic model assessment beta cell [HOMA-B]), and insulin resistance (HOMA-IR) were analyzed in the MAGIC study, while FBG was investigated in the CoLaus study.

BMI, waist circumference, and waist-to-hip ratio were the only adiposity traits analyzed by the GIANT Consortium. Triglycerides, total cholesterol, HDL-cholesterol, and LDL-cholesterol were analyzed in the "Genome Wide Associations Scans for Total Cholesterol, HDL-C, LDL-C and triglycerides" study.[16]

2.4 | Estimation of the functional activity of the significant SNPs

The RegulomeDB[17] database was used to estimate the functional activity of the significant SNPs. This database annotates SNPs with known and predicted regulatory elements in the intergenic regions of the *Homo sapiens* genome and reveals known and predicted regulatory DNA elements including regions of DNase hypersensitivity, binding sites of transcription factors, and promoter regions that have been biochemically characterized to regulate transcription. The source of these data includes public datasets from GEO, the ENCODE project, and the published literature.

TABLE 2 Association between *SP110 rs2114592C>T* genotypes and NODAT in the Swiss Transplant Cohort Study (STCS) replication sample and the combined sample

	CC	CT	TT	T allele dosage
STCS replication sample				
Sample size	376	105	8	
Odds ratio (95% CI)	1 (Ref.)	1.26 (0.72-2.20)	8.90 (1.97-40.0)	1.70 (1.08-2.66)
P-corrected value[a]		1.0	.04	.20
Combined STCS samples				
Sample size	935	234	15	
Odds ratio (95% CI)	1 (Ref.)	1.16 (0.81-1.65)	9.90 (3.22-30.5)	1.53 (1.13-2.06)
P value		.42	.00006	.005

Results are expressed as odds ratio and (95% CI). Statistical analysis by logistic regression adjusting for age of recipient at transplantation (continuous variable), sex of the recipient, baseline BMI (continuous variable), type of calcineurin inhibitor (no calcineurin inhibitor/tacrolimus/cyclosporine). No adjustment was done for HCV status (as in the discovery sample) because data were not available.
BMI, body mass index; CI, confidence interval; HCV, hepatitis C virus; NODAT, new-onset diabetes mellitus after transplantation; STCS, Swiss Transplant Cohort Study.
[a]*P* values were corrected using Bonferroni correction for multiple testing (corrected *P* value = original *P* value × 10).

3 | RESULTS

3.1 | General characteristics of the study populations

The incidence of NODAT in the discovery sample was 21.8%. NODAT patients were older with higher baseline BMI and were more often prescribed tacrolimus-based maintenance immunosuppression compared to non-NODAT patients. A detailed description of this sample is presented in Table 1. Similar descriptions for the replication samples are presented in Tables S2 and S3.

3.2 | Polymorphisms in immune-related genes and NODAT in the STCS and the replication samples

The minor allele frequency was <5% for 12 SNPs in our discovery sample and they were removed from the analyses. Additionally, 12 SNPs did not meet HWE ($P < .05$); therefore, a total of 263 SNPs were finally analyzed (Table S1, Parts A, B, and C). Among these SNPs, 10 polymorphisms were associated with NODAT ($P < .01$ in additive models and/or allele dosage; Table S4). These SNPs were used for replication in the first replication sample (second subset of the STCS sample).

Among these 10 SNPs only 1 SNP, the *SP110 nuclear body protein* (*SP110*) *rs2114592C>T*, was associated with NODAT in the first replication sample after correction for multiple testing (Table 2) and was therefore investigated in the second SOT replication sample and in other population-based samples. A significant observation was also noticed when combining both STCS samples, with *SP110 rs2114592-TT* genotype carriers having 9.9 (95% confidence interval [CI] 3.22-30.5, $P = .00006$) higher risk of developing NODAT compared to reference genotype carriers (Table 2). Importantly, *SP110 rs2114592C>T* SNP was also associated with NODAT in the second SOT replication sample (n = 156, odds ratio [OR]: 4.8, 95% CI, 1.55-14.6, $P = .006$). Time to NODAT in the combined STCS sample by *SP110* genotype is shown in Figure 1, with an increased incidence of NODAT during the first year in *SP110 rs2114592-TT* genotype carriers ($P = .003$) compared to other genotypes.

A second SNP (*rs2907749A>G*) within the *nucleotide-binding oligomerization domain containing 1 (NOD1)* was associated with NODAT in the discovery and in the first replication samples, but this association did not remain significant after multiple testing correction (Table S5a). This SNP was not significantly associated with NODAT in the second SOT replication sample (data not shown), but was significantly associated with NODAT (n = 1156), new-onset hyperlipidemia (n = 1051), lower HDL-cholesterol (n = 1051), and higher total cholesterol levels (n = 1051) in the combined STCS sample (Table S5a and 5b).

3.3 | *SP110 rs2114592C>T* SNP and other metabolic syndrome complications in the combined STCS sample

The associations between *SP110 rs2114592C>T* SNP and other metabolic syndrome complications were investigated in the combined STCS sample (n = 1071). No association was observed between *SP110 rs2114592C>T* SNP and BMI (dichotomized into normal versus overweight or obese) at 12 months post-SOT or when analyzing the evolution of BMI over time between different genotype groups (data not shown). No associations were observed between *SP110 rs2114592 C>T* SNP and clinical characteristics listed in Table 1 (data not shown). Carriers of *SP110 rs2114592-TT* genotype presented

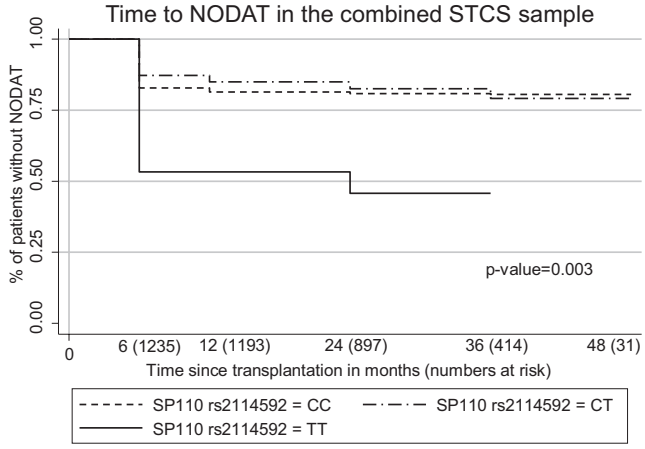

FIGURE 1 Time to new-onset diabetes after transplantation (NODAT) in the combined Swiss Transplant Cohort Study (STCS) sample by SP110 genotype for the rs2114592 polymorphism

3 times higher risk of developing new-onset hyperlipidemia after transplantation (OR: 3.41, 95% CI, 1.06-11.0, $P = .04$) when compared to the reference genotype (*SP110 rs2114592-CC*), but this genotype was not associated with differences of lipid blood levels (Table S6).

3.4 | *SP110 rs2114592C>T* SNP and metabolic markers in population-based samples

In the population-based samples, SP110 *rs2114592C>T* SNP was not associated with the different metabolic markers investigated, including available diabetes mellitus markers (Table S7).

3.5 | Tagging SNPs of *SP110* gene

Twenty-six tagging SNPs within the *SP110* gene were found using HapMap Genome Browser (Table S8). Only *SP110 rs7580900T>C* SNP was associated with NODAT in the discovery sample (n = 695, OR: 0.72, 95% CI, 0.55-0.95, $P = .02$ in additive model and OR: 0.43, 95% CI, 0.23-0.80, $P = .007$ for *rs7580900-CC* genotype compared to the reference genotype). This significant association between *SP110 rs7580900T>C* SNP and NODAT was not replicated in the different replication samples (data not shown).

3.5.1 | Estimation of the functional activity of *SP110 rs2114592C>T* SNP

The functional activity of *SP110 rs2114592C>T* SNP was investigated using the RegulomeDB database.[17] We found 22 SNPs to be in complete linkage disequilibrium ($r^2 = 1$) with *SP110 rs2114592C>T* SNP (intronic variant), among them the *SP110 rs75411703* SNP, which shows a deoxyribonuclease sensitivity quantitative trait loci (dsQTL) effect and is linked to the expression of the target gene.[18] Additionally, by using the eQTL gene browser-gene network database,[19] *SP110 rs2114592-T* allele is associated with a decrease of *SP110* expression in the blood ($P = 3.87E-26$). Altogether, these data suggest that *SP110 rs2114592C>T* SNP might have important regulatory functions.

4 | DISCUSSION

Inflammation is intimately linked with the development of diabetes mellitus in the general population (reviewed in refs. 20, 21) and recipients of SOT.[5] In our study, 10 polymorphisms within 10 different genes primarily implicated in the innate immune response or inflammation were significantly ($P < .01$) associated with NODAT in a discovery cohort of 696 patients from the STCS without a previous history of diabetes mellitus or glucose intolerance before SOT. These significant results were tested for replication in 2 samples with SOT (a second subset of the STCS and a third independent sample with SOT), and only 1 polymorphism in the intronic region of *SP110* gene *(rs2114592C>T)* was found to be significantly associated with NODAT in both replication samples. Carriers of the *rs2114592-TT* genotype had nearly 10 times increased risk of NODAT in the combined STCS sample. They also showed an increased risk of new-onset hyperlipidemia. However, *SP110 rs2114592C>T* was not associated with diabetes mellitus in large population-based samples.

SP110 nuclear body protein (encoded by the *SP110* gene) is an activator of gene transcription and may serve as a nuclear hormone receptor coactivator.[22] This gene is a member of the SP100/SP140 family of nuclear body proteins and is involved in several cellular processes such as apoptosis, cell cycle control, and a prototype type I interferon-induced gene that regulates immune responses, in particular in myeloid cells differentiation. Most of the studies investigating SP110 have shown its role in immunoprotective defenses against infectious organisms in humans, mainly tuberculosis.[23] Human SP110 is the closest homolog of mouse intracellular pathogen resistance-1 (Ipr1) protein, and previous animal studies showed that the expression of the *Ipr1* transgene in macrophages limits the multiplication of *Mycobacterium tuberculosis* and switches the infected macrophages toward an apoptotic cell death pathway.[23] Several studies in humans investigated the association between polymorphisms of *SP110* (among them the *rs2114592C>T*) and the risk of developing pulmonary tuberculosis, but results were inconsistent.[24,25] No previous studies related *SP110* or its polymorphisms to diabetes mellitus or NODAT. Interestingly, SP110, as a transcriptional coactivator, enhances transcription of genes with retinoic acid response elements,[22] and several of these genes such as *IL2RA*, *PEPCK1*, *UCP1*, and *CD38*[26] are known candidate genes for type 1 and type 2 diabetes mellitus. Retinoids are important in glucose and lipid metabolism, and the activation of the retinoic acid cascade through the retinoic acid response elements has been previously related to the development of diabetes mellitus, adipogenesis, and insulin resistance (reviewed in ref. 27). The functional activity of *SP110 rs2114592C>T* was investigated and was linked to the expression of the target gene. Moreover, it has several histone marks and influences the binding of several proteins, among them some important immune regulatory proteins (eg, IRF-4 and NF-ATc1), but also the transcription factor

E2-alpha (encoded by TCF3). TCF3 is a transcriptional regulator that binds to IEB1 and IEB2, which are short DNA sequences in the insulin gene transcription control region providing a new possible mechanism relating *SP110* to the development of diabetes mellitus and/or NODAT.

Homozygous loss of function mutations in *SP110* in humans cause hepatic veno-occlusive disease with immunodeficiency (VODI), a monogenic autosomal-recessive primary immunodeficiency associated with hepatic vascular occlusion and fibrosis[28] and a severe combined immunodeficiency despite normal lymphocyte numbers. VODI is associated with high mortality either due to hepatic failure but also to life-threatening infections, often pneumocystis.[28] SP110-deficient B cells have reduced ability to respond to T cell–dependent signals and to differentiate into plasmablasts, which might be the explanation for the observed hypogammaglobulinemia in VODI.[29] Metabolic alterations, especially diabetes mellitus, are not hallmarks of VODI; however, these patients often die within the first 12 months of life, so long-term metabolic complications of SP110 deficiency might be missed.[28] These data altogether suggest that SNPs within the *SP110* gene may be linked in populations at risk with an altered inflammatory response, leading to the development of NODAT.

We also tested whether the associations between *SP110 rs2114592C>T* SNP and diabetes mellitus are present in the general population. No effect was observed in 2 large population-based samples and a large case-control sample of patients with type 2 diabetes mellitus, pointing out the implication of this SNP only in populations at high risk of developing metabolic diseases including diabetes mellitus. Although there is a strong correlation between inflammation and type 2 diabetes mellitus in the general population, our results suggest a specific gene-environment interaction. We have previously shown an association between a SNP in the *CRTC2* gene and NODAT and other metabolic syndrome traits in transplanted patients. This association was also found in population-based samples, but with a very weak effect size.[12] Additionally, the same observation was noted for the *CRTC1* gene, for which an effect of a SNP was found on obesity in psychiatric patients, a population at high risk of metabolic syndrome, but not in population-based samples.[30]These observations point out that, in patients at risk of developing metabolic syndrome traits, because of the disease itself and/or the medication (eg, immunosuppressive or psychotropic medications), both diseases and/or medications can act as important triggers, unmasking different genetic effects. This study has several limitations and strengths. One of the major limitations of genetic studies in this field is the different definitions used for NODAT. However, it should be noted that NODAT definition in the discovery and second STCS replication samples was the same and the closest to the criteria given by the WHO and ADA consensuses,[15] which is a strength in our study. Even though the association between *SP110 rs2114592C>T* and NODAT was replicated in 2 cohorts, they are modest in size and not truly independent. The present finding must therefore be confirmed by future studies. The selection of the SNPs was based on SNPs already associated with infections and inflammation in the literature and does not cover all variants within these genes, so unknown variants within these candidate genes with important associations with NODAT or novel pro-inflammatory variants may have been missed. Our findings on samples in white subjects cannot be extrapolated to other ethnicities. The present results do not allow determining whether *SP110 rs2114592C>T* SNP is the causative variant or merely a proxy of 1 or more yet unidentified variants. Despite the important regulatory functions suggested by the RegulomeDB and the eQTL databases, further studies are needed to elucidate which precise mechanisms underlie the observed associations. Another limitation is the low allelic frequency of *SP110 rs2114592C>T* with only few homozygote carriers of *TT* genotype being observed (Table S9). Thus, *SP110 rs2114592C>T* contributes only to a small fraction of new-onset diabetes mellitus in the analyzed populations. On the other hand, the fact that these results were replicated in 2 independent samples with SOT strengthens the validity of our data. In addition, the high observed ORs suggest important clinical implications of *SP110 rs2114592C>T* SNP. Well-designed prospective clinical trials should therefore be conducted to test whether genetic testing before transplantation may offer more precise new-onset diabetes mellitus risk stratification. In addition, SP110 may be even more relevant in predicting diabetes mellitus in specific subgroups of at-risk SOT patients (eg, HCV-infected transplant recipients or overweight patients), but this remains to be analyzed.

In conclusion, our results link for the first time the *SP110* gene in the development of diabetes mellitus and other metabolic complications in SOT recipients. While the pathway by which the *SP110 rs2114592C>T* SNP is associated with diabetes mellitus remains undetermined, our study opens the door for further investigation of this SNP and provides a promising new candidate gene for diabetes mellitus development. Ultimately, an important question will be to determine whether personalized strategies based on genetic testing in pretransplant patients may help to reduce the risk of developing diabetes mellitus.

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AUTHORS' CONTRIBUTIONS

All authors reviewed, revised, and approved the final version of the manuscript. LQ obtained funding, did the statistical analysis, interpreted the data, and wrote the manuscript. WA did the genetic analysis (STCS cohort) and interpreted the data. PYB obtained funding, designed the genetic analysis (STCS cohort), and interpreted the data. SC collected and interpreted the data (second replication SOT sample). JPV, OM, DG, RL, NJM, IB, CvD, JS, PM, JFD, PMS, and MP obtained funding, designed the STCS study, and collected and interpreted the data. ZK supervised the statistical analysis and did the statistical analysis for the population-based cohorts. PMV, MR, CH, and FV interpreted the data. CBE was the chief investigator, obtained funding, designed the study, and interpreted the data.

DISCLOSURE

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SUPPORTING INFORMATION

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