Impact of polymorphisms in genes orchestrating innate immune responses on replication kinetics of Torque teno virus after kidney transplantation

Natalia Redondo1,2*, Isabel Rodríguez-Goncer1,2, Patricia Parra1, Eliseo Albert3, Estela Giménez2,3, Tamara Ruiz-Merlo1, Francisco López-Medrano1,2,4, Rafael San Juan1,2,4, Esther González2, Ángel Sevillano5, Amado Andrés1,5, David Navarro2,3,6, José María Aguado1,2,4 and Mario Fernández-Ruiz1,2,4

1Unit of Infectious Diseases, Hospital Universitario ‘12 de Octubre’, Instituto de Investigación Sanitaria Hospital ‘12 de Octubre’ (imas12), Madrid, Spain, 2Centro de Investigación Biomédica en Red de Enfermedades Infecciosas (CIBERINFEC), Instituto de Salud Carlos III, Madrid, Spain, 3Department of Microbiology, Instituto de Investigación Sanitaria INCLIVA, Hospital Clínico Universitario, Valencia, Spain, 4Department of Medicine, School of Medicine, Universidad Complutense, Madrid, Spain, 5Department of Nephrology, Instituto de Investigación Sanitaria Hospital ‘12 de Octubre’ (imas12), Hospital Universitario ‘12 de Octubre’, Madrid, Spain, 6Department of Microbiology, School of Medicine, University of Valencia, Valencia, Spain

Background: Torque teno virus (TTV) DNAemia has been proposed as a surrogate marker of immunosuppression after kidney transplantation (KT), under the assumption that the control of viral replication is mainly exerted by T-cell-mediated immunity. However, the impact on post-transplant TTV kinetics of single genetic polymorphisms (SNPs) in genes orchestrating innate immune responses remains unknown. We aimed to characterize the potential association between 14 of these SNPs and TTV DNA levels in a single-center cohort of KT recipients.

Methods: Plasma TTV DNAemia was quantified by real-time PCR in 221 KT recipients before transplantation (baseline) and regularly through the first 12 post-transplant months. We performed genotyping of the following SNPs: CTLA4 (rs5742909, rs231775), TLR3 (rs3775291), TLR9 (rs5743836, rs352139), CD209 (rs735240, rs4804803), IFNL3 (rs12979860, rs8099917).

Abbreviations: AUC, area under the curve; BKPyV, BK polyomavirus; CI, confidence interval; CpG, cytosine-phosphate-guanine; CMV, cytomegalovirus; D, donor; dsRNA, double-stranded RNA; ESRD, end-stage renal disease; HBV, hepatitis B virus; HCV, hepatitis C virus; HIV, human immunodeficiency virus; HSCT, hematopoietic stem cell transplantation; IQR, interquartile range; KT, kidney transplantation; LLod, lower limit of detection; OR, odds ratio; PAMP, pathogen-associated molecular pattern; PCR, polymerase chain reaction; R, recipient; PRR, pattern recognition receptor; SD, standard deviation; SNP, single-nucleotide polymorphism; SOT, solid organ transplantation; TLR, toll-like receptor; TTV, Torque teno virus.
Results: The presence of the minor G allele of CD209 (rs4804803) in the homozygous state was associated with undetectable TTV DNAemia at the pre-transplant assessment (adjusted odds ratio: 36.96; 95% confidence interval: 4.72–289.67; p-value = 0.001). After applying correction for multiple comparisons, no significant differences across SNP genotypes were observed for any of the variables of post-transplant TTV DNAemia analyzed (mean and peak values, areas under the curve during discrete periods, or absolute increments from baseline to day 15 and months 1, 3, 6 and 12 after transplantation).

Conclusion: The minor G allele of CD209 (rs4804803) seems to exert a recessive protective effect against TTV infection in non-immunocompromised patients. However, no associations were observed between the SNPs analyzed and post-transplant kinetics of TTV DNAemia. These negative results would suggest that post-transplant TTV replication is mainly influenced by immunosuppressive therapy rather than by underlying genetic predisposition, reinforcing its clinical application as a biomarker of adaptive immunity.

KEYWORDS single-nucleotide polymorphisms, Torque teno virus, TTV replication kinetics, kidney transplantation, SNP

Introduction

The study of the human virome in health and disease has gained growing attention over recent years (Webb et al., 2020; Dodi et al., 2021). Viruses belonging to Anelloviridae family are the most abundant eukaryotic viruses in the virome and may be detected in a variety of samples, such as blood, plasma, urine or saliva (Kaczorowska and van der Hoek, 2020; Arze et al., 2021). Anelloviruses are non-enveloped viruses with small circular replication-associated protein-encoding single-stranded DNA genomes (Biagini, 2009; Kaczorowska and van der Hoek, 2020), which lack attributable pathogenic roles (“orphan viruses”) (Focosi et al., 2016; Rezahosseini et al., 2019). Once primary infection occurs at early stages of life, anelloviruses remain in different body compartments and fluids—including peripheral blood mononuclear cells, feces, semen, throat swabs, umbilical cord blood, lungs, kidneys or cerebrospinal fluid—under the control of the immune system, resulting in a prevalence as high as 90% in the adult population (Redondo et al., 2022a). The precise underlying mechanisms on how this immune control is carried out largely remain to be determined, although a major role has been proposed for the cellular arm. Belonging to the Alphatorquevirus genus and discovered in 1997 (Nishizawa et al., 1997), Torque teno virus (TTV) has been proven by us and others to serve as a convenient surrogate marker of the overall status of immunosuppression after solid organ (SOT) and allogeneic hematopoietic stem cell transplantation (HSCT) (Fernandez-Ruiz et al., 2019; Rezahosseini et al., 2019; Mouton et al., 2020; Redondo et al., 2022a; Jaksch et al., 2022).

The innate immunity acts as a frontline defense against viruses through an orchestrated response, that is, triggered upon recognition of viral motifs by pathogen recognition receptors (PRRs) present in macrophages and dendritic cells (Takeuchi and Akira, 2010). The rationale for the use of TTV DNAemia as a biomarker of immune competence after SOT lies on the assumption that the viral kinetics is mainly dictated by the T-cell-mediated immunity (Redondo et al., 2022a; Jaksch et al., 2022). Indeed, various studies have shown a direct correlation between TTV DNA loads and calcineurin inhibitors trough levels (Gorzer et al., 2014; Jaksch et al., 2018). The role played by the innate immune arm in the setting of ongoing immunosuppression remains largely unknown, as is the potential impact of polymorphisms in genes coding for PRRs (such as toll-like receptors [TLRs]), interleukins (IL) or interferons (IFNs) (Prasetyo et al., 2015; Ramzi et al., 2019; Ramzi et al., 2021). Evidence of an individual genetic susceptibility to TTV regardless of the amount of immunosuppressive therapy would question the reliability of viral replication as clinical biomarker in the SOT population.

We aimed to investigate the association between 14 single genetic polymorphisms (SNPs) in different genes mainly involved in the orchestration of innate immune responses (Table 1) and TTV DNA levels at baseline and various points during the first post-transplant year in a well characterized cohort of kidney transplant (KT) recipients (Fernandez-Ruiz et al., 2019). The selection of these SNPs was dictated by
| Gene    | Encoded protein                                                                 | Biological function                                                                 | SNP ID number | Nucleotide substitution (reference allele/alternative allele) | Global allele frequency<sup>a</sup> | Impact of the SNP on the susceptibility to infection in previous studies<sup>b</sup>                                                                 |
|---------|----------------------------------------------------------------------------------|-------------------------------------------------------------------------------------|---------------|----------------------------------------------------------------|-------------------------------------|-----------------------------------------------------------------------------------|
| **CTLA4** | Cytotoxic T-lymphocyte antigen 4 (CTLA-4/CD152)                                | T-cell co-inhibitory receptor                                                       | rs5742909     | C / T                                                          | C = 0.91755                        | Increased risk of CMV after SOT (Misra et al. 2015)                                  |
|         |                                                                                 |                                                                                     | rs231775      | A / G                                                          | T = 0.08245                        | Increased risk of chronic HCV in the general population (Ali et al. 2022) and CMV after SOT (Misra et al. 2015) |
| **TLR3** | Toll-like receptor 3: endosomal PRR                                             | Endocytic pathogen recognition receptor of single and double-stranded RNA            | rs3775291     | C / T                                                          | C = 0.716526                       | Increased risk of CMV (Redondo et al. 2022b) and BKPyV after SOT (Redondo et al. 2022c), increased risk of dengue (Singh et al. 2021) and HBV in the general population (Ye et al. 2020) |
| **TLR9** | Toll-like receptor 9: endosomal PRR                                             | Recognition of unmethylated CpG motif-containing DNA                                  | rs5743836     | A / G                                                          | A = 0.80444                        | Protection against TB (Varshney et al. 2022), higher HBV viral load (Chihab et al. 2019) in the general population |
|         |                                                                                 |                                                                                     | rs352139      | T / C                                                          | T = 0.458978                       | Increased risk of CMV after SOT (Redondo et al. 2022b), increased risk of EBV-related IM in the general population (Jablonska et al. 2020) |
| **CD209** | Dendritic cell-specific ICAM 3-grabbing nonintegrin (DC-SIGN/CD209): endosomal C-type lectin receptor | Recognition of carbohydrates present in viruses, bacteria, fungi and parasites and DAMPs in damaged host T-cells | rs735240      | G / A                                                          | G = 0.57414                        | Increased risk of CMV after SOT (Fernandez-Ruiz et al. 2015) and HSCT (Mezger et al. 2008) |
|         |                                                                                 |                                                                                     | rs4804803     | A / G                                                          | A = 0.786719                       | Protection against BKPyV after SOT (Redondo et al. 2022c), protection against severe dengue (Sakuntabhai et al. 2002) and TBE (Czupryna et al. 2017) and increased risk of symptomatic CHIKV (Chaaithanya et al. 2016) in the general population |
| **IFNL4** | Interferon-λ3 (IL28B), type III interferon: soluble immune mediator             | Antiviral cytokine                                                                   | rs12979860    | C / T                                                          | C = 0.672446                       | Lower HCV clearance upon IFN-a therapy in the general population (Bliri et al. 2021), protection against CMV after SOT (Fernandez-Ruiz et al. 2015) and HSCT (Bravo et al. 2014) |
|         |                                                                                 |                                                                                     | rs8099917     | T / G                                                          | T = 0.808472                       | Lower HCV clearance upon IFN-a therapy in the general population (Li et al. 2016), protection against CMV after SOT (Egh et al. 2014) |
| **TNF**  | Tumor necrosis factor                                                            | Pro-inflammatory cytokine                                                            | rs1800829     | G / A                                                          | G = 0.847933                       | Increased risk of severe influenza (Aliagaras et al. 2021) and COVID-19 (Gupta et al. 2022) in the general population |
| **IL10** | Pleiotropic cytokine                                                             |                                                                                     | rs1800872     | T / G                                                          | T = 0.29385                        |                                                                                     |

(Continued on following page)
previous research showing a potential impact on the susceptibility to viral infections. In the case of TLR3 (rs3775291), various pieces of evidence have shown an effect on the incidence of infection by cytomegalovirus (CMV) or BK polyomavirus (BKPyV), two relevant viral pathogens in the KT scenario, but also tick-borne encephalitis, BK polyomavirus (BKPyV), two relevant viral pathogens in

Material and methods

Study population and setting

The present research was performed as a post hoc retrospective analysis of a previous study that investigated the role of TTV DNA levels to predict the occurrence of serious and opportunistic infection and de novo malignancy in a cohort of KT recipients recruited at the University Hospital “12 de Octubre” (a 1,300-bed tertiary care center in Madrid with an active KT program since 1990) between November 2014 and December 2016 (Fernandez-Ruiz et al., 2019). As detailed elsewhere, adult patients with end-stage renal disease (ESRD) undergoing KT during the study period and providing informed consent were eligible for inclusion. Exclusion criteria included double organ transplantation and primary graft non-function. By applying these criteria, 221 KT recipients were eventually included. The study was performed in accordance with the ethical standards laid down in the Declarations of Helsinki and Istanbul. The local Clinical Research Ethics Committee approved the study protocol.

Study design

Participants were enrolled at the time of KT and followed-up for at least 12 months, unless graft loss (retransplantation or return to dialysis) or death occurred earlier. Plasma TTV DNA load was quantified at baseline (i.e., within 6 h prior to the transplant procedure), day 7, and months 1, 3, 6 and 12 by a

TABLE 1 (Continued) Candidate SNPs selected for the present study.

| Gene                  | Encoded protein                  | Biological function                                                                 | SNP ID number | Nucleotide substitution (reference allele/alternative allele) | Global allele frequency* | Impact of the SNP on the susceptibility to infection in previous studies† |
|-----------------------|----------------------------------|-------------------------------------------------------------------------------------|---------------|---------------------------------------------------------------|--------------------------|-----------------------------------------------------------------------|
| Interleukin-10: human cytokine | rs1878672 G / C                  | Increased risk of BKPyV after SOT (Redondo et al. 2022c)                           | **G = 0.90165** | Increased risk of BKPyV after SOT (Redondo et al. 2022c)     |                          | Increased risk of BKPyV after SOT (Redondo et al. 2022c)                           |
| IL12B: human cytokine  | rs3121227 T / G                  | No apparent impact on the risk of CNV after HSCT (Corrales et al. 2015)           | **C = 0.3111** | No apparent impact on the risk of CNV after HSCT (Corrales et al. 2015) |                          | No apparent impact on the risk of CNV after HSCT (Corrales et al. 2015) |
| IL17: human cytokine   | rs2275913 G / A                  | Increased risk of BKPyV after SOT (Redondo et al. 2022c)                           | **A = 0.334257** | Increased risk of BKPyV after SOT (Redondo et al. 2022c)     |                          | Increased risk of BKPyV after SOT (Redondo et al. 2022c)                           |

*Obtained from ALFA Allele Frequency (available at: https://www.ncbi.nlm.nih.gov/snp/).
†The clinical effect associated with the minor (alternative) allele of the corresponding SNP is detailed.

BKPyV, BK polyomavirus; CHIK, chikungunya virus; CMV, cytomegalovirus; COVID-19, coronavirus disease 2019; CTLA-4, cytotoxic T-lymphocyte antigen 4; DAMP, damage-associated molecular pattern; HBV, hepatitis B virus; HCV, hepatitis C virus; HSCT, hematopoietic stem cell transplantation; IFN, interferon; IL, interleukin; IM, infectious mononucleosis; NK, natural killer; PRR, pattern recognition receptor; SD, standard deviation; SNP, single-nucleotide polymorphism; SOT, solid organ transplantation; TB, tuberculosis; TLR, toll-like receptor; TTF, tumor necrosis factor; TTV, torque teno virus.
polymerase chain reaction (PCR)-based quantitative nucleic acid amplification test. Immunosuppression and prophylaxis regimens are detailed as Supplementary Material.

Single genetic polymorphisms genotyping

Whole blood specimens that have been stored at −70°C were retrieved for SNP genotyping. DNA was extracted with the KingFisher Duo Prime system using the MagMax DNA Multi-Sample Ultra 2.0 kit (Thermo Fisher Scientific, Waltham, MA) following the manufacturer’s instructions. CTLA4 (rs5742909, rs231775), TLR3 (rs3775291), TLR9 (rs5743836, rs352139), CD209 (rs735240, rs4804803), IFNL3
DNAemia through discrete time periods (1, 3, and 6 months). DNA levels and areas under the curve (AUCs) for TTV parameters were compared across SNPs: peak plasma TTV (i.e. plasma DNA levels). In addition, other viral kinetic T-Student or U-Mann-Whitney tests for continuous variables undetectable [below the LLoD] DNAemia), or by the Fisher

Quantitative data were reported as the mean ± standard deviation (SD) or the median with interquartile range (IQR). Qualitative variables were given as absolute and relative frequencies. Normality of the distributions was tested with the Kolgomorov-Smirnov test. Deviation from the Hardy-Weinberg equilibrium for each SNP was evaluated by the χ² test with one degree of freedom. Comparisons of TTV kinetics at different points across SNP genotypes were performed by the χ² test or the Fisher’s exact test for qualitative variables (i.e. detectable or undetectable [below the LLoD] DNAemia), or by the T-Student or U-Mann-Whitney tests for continuous variables (i.e. plasma DNA levels). In addition, other viral kinetic parameters were compared across SNPs: peak plasma TTV DNA levels and areas under the curve (AUCs) for TTV DNAemia through discrete time periods (1, 3, and 6 months after transplantation), and increments (Δ) in DNA levels from baseline to day 15 and months 1, 3, 6 and 12. Additional pairwise comparisons were conducted between different SNP genotype groups, either individually or in combination. The independent impact of selected SNPs on the probability of having undetectable TTV DNAemia was confirmed by logistic regression, with associations given as odds ratios (ORs) and 95% CIs. All the significance tests were two-tailed and considered as significant at a p-value < 0.05. To control for p-value inflation due to multiple comparisons, the Bonferroni method (corrected α value = nominal α value/total number of comparisons) was applied. Statistical analysis was performed using SPSS version 21 (Statistical Package for Social Sciences, Chicago, IL).

Results

We included 221 KT recipients, whose demographics, clinical characteristics and patient and graft outcomes are detailed in Table 2. Samples from all the patients were successfully genotyped for the 14 SNPs considered. The median number of assessments for plasma TTV DNA per patient was 5 (IQR: 4–5). The majority of recipients had detectable TTV DNAemia (i.e. above the LLoD) at every time point, ranging from 96.3% (180/187) at baseline to 99.4% (176/177) at post-transplant month 6. The genotypic frequencies of candidate SNPs are shown in Supplementary Table S1. The observed genotype frequency distributions did not deviate from those expected according to the Hardy-Weinberg equilibrium except for TLR9 (rs5743836) and IFNL3 (rs12979860).

First, the effect of studied polymorphisms on plasma TTV DNAemia at discrete time points was investigated. In particular, we explored the impact of the minor alleles in each SNP in both dominant (heterozygous and homozygous) and recessive (homozygous only) models. Across the 14 SNPs considered, we did not find significant differences in TTV DNA levels at any of the monitoring points (Table 3).

Next, we analyzed if there was any association between candidate SNPs and the presence of undetectable plasma TTV DNAemia at baseline (before the initiation of immunosuppressive therapy). Seven (3.7%) patients had pre-transplant TTV DNA levels below the LLoD. We observed that carriers of the minor C allele of the IL10 (rs1878672) SNP in the homozygous state (CC) were more likely to have undetectable baseline TTV DNAemia compared to recipients bearing the reference G allele (GG/GC) [12.5% (3/24) versus 2.5% (4/163), respectively; nominal p-value = 0.046]. There were also significant differences within the TLR3 (rs3775291) SNP, since all the 7 patients with undetectable TTV DNAemia harbored the minor T allele either in the heterozygous or the homozygous state [7.1% (7/99) versus 0.0% (0/88) for CT/TT and CC carriers; nominal p-value = 0.015]. Finally, the minor allele of CD209 (rs4804803) in the homozygous state was also associated with
| SNP (ID number) | Model | Plasma TTV DNA level, log_{10} copies/mL (mean ± SD) |
|----------------|-------|----------------------------------------------------|
|******|
| **CTLA4 (rs5742909)** | Dominant | CC 2.9 ± 1.6 0.284 3.2 ± 1.6 0.389 4.4 ± 1.7 0.521 5.9 ± 1.8 0.495 | |
| | | CT/TT 2.6 ± 1.5 2.9 ± 1.9 4.2 ± 1.6 5.7 ± 1.5 0.389 4.0 ± 1.6 5.5 ± 1.9 | |
| | Recessive | CC/CT 2.9 ± 1.6 0.643 3.1 ± 1.7 0.821 4.4 ± 1.7 0.441 5.9 ± 1.7 0.620 | |
| | | TT 3.5 ± 1.0 3.3 ± 1.0 5.0 ± 1.2 6.3 ± 1.8 0.495 4.6 ± 1.8 5.8 ± 1.8 | |
| **CTLA4 (rs231775)** | Dominant | AA 2.8 ± 1.4 0.705 3.1 ± 1.6 0.972 4.5 ± 1.5 0.364 5.9 ± 1.7 0.961 | |
| | | AG/GG 2.9 ± 1.8 3.1 ± 1.7 4.3 ± 1.9 5.9 ± 1.8 0.496 4.6 ± 1.8 5.6 ± 1.7 | |
| | Recessive | AA/AG 2.9 ± 1.6 0.601 3.1 ± 1.7 0.689 4.5 ± 1.7 0.233 5.9 ± 1.7 0.527 | |
| | | GG 3.1 ± 1.7 3.3 ± 1.2 4.0 ± 1.9 6.1 ± 2.0 0.496 4.6 ± 1.8 5.8 ± 1.7 | |
| **TLR3 (rs3775281)** | Dominant | CC 2.9 ± 1.3 0.703 3.1 ± 1.7 0.730 4.2 ± 1.6 0.147 5.9 ± 1.7 0.441 | |
| | | CT/TT 2.8 ± 1.9 3.2 ± 1.6 4.6 ± 1.8 5.7 ± 1.6 0.364 4.6 ± 1.8 5.6 ± 1.7 | |
| | Recessive | CC/CT 2.8 ± 1.6 0.500 3.1 ± 1.6 0.917 4.4 ± 1.7 0.923 5.9 ± 1.8 0.908 | |
| | | TT 3.1 ± 2.0 3.1 ± 2.0 4.4 ± 2.0 5.8 ± 1.4 0.496 4.6 ± 1.8 4.9 ± 2.2 | |
| **TLR9 (rs5743836)** | Dominant | AA 2.9 ± 1.6 0.337 3.2 ± 1.6 0.692 4.4 ± 1.7 0.829 5.9 ± 1.8 0.566 | |
| | | AG/GG 2.7 ± 1.8 3.1 ± 1.8 4.4 ± 1.6 6.0 ± 1.7 0.829 5.2 ± 1.5 5.4 ± 1.9 | |
| | Recessive | AA/AG 2.9 ± 1.6 0.733 3.1 ± 1.7 0.902 4.4 ± 1.7 0.640 5.9 ± 1.7 0.116 | |
| | | GG 2.7 ± 1.0 3.2 ± 1.5 4.2 ± 1.4 5.0 ± 1.4 0.640 5.1 ± 1.8 | |
| **TLR9 (rs352139)** | Dominant | TT 3.0 ± 1.8 0.388 3.1 ± 2.0 0.935 4.3 ± 2.1 0.488 6.2 ± 1.9 0.132 | |
| | | CT/CC 2.8 ± 1.6 3.1 ± 1.5 4.5 ± 1.5 5.8 ± 1.7 0.488 5.1 ± 1.8 | |
| | Recessive | TT/TC 2.9 ± 1.6 0.508 3.1 ± 1.6 0.521 4.3 ± 1.8 0.184 6.0 ± 1.8 0.111 | |
| | | CC 2.7 ± 1.7 3.3 ± 1.7 4.7 ± 1.5 5.6 ± 1.6 0.184 5.1 ± 1.8 | |
| **CD209 (rs735240)** | Dominant | GG 2.8 ± 1.8 0.560 3.0 ± 1.9 0.532 4.6 ± 1.8 0.376 5.9 ± 1.8 0.883 | |
| | | GA/AA 2.9 ± 1.5 3.2 ± 1.5 4.3 ± 1.7 5.9 ± 1.7 0.376 5.2 ± 2.6 5.3 ± 2.2 | |
| | Recessive | GG/GA 2.8 ± 1.6 0.163 3.1 ± 1.7 0.808 4.4 ± 1.8 0.486 5.9 ± 1.7 0.907 | |
| | | AA 3.2 ± 1.7 3.2 ± 1.4 4.6 ± 1.4 5.9 ± 1.8 0.486 5.2 ± 2.1 5.3 ± 2.2 | |
| **CD209 (rs4804803)** | Dominant | AA 2.9 ± 1.6 0.760 3.1 ± 1.5 0.726 4.4 ± 1.6 0.724 5.9 ± 1.6 0.927 | |
| | | AG/GG 2.8 ± 1.7 3.2 ± 1.8 4.4 ± 1.9 5.9 ± 1.9 0.724 5.1 ± 2.2 5.6 ± 2.5 0.927 4.6 ± 1.7 | |
| | Recessive | AA/AG 2.9 ± 1.5 0.294 3.2 ± 1.6 0.373 4.4 ± 1.7 0.924 5.9 ± 1.7 0.658 | |
| | | GG 1.6 ± 3.2 2.5 ± 2.5 4.5 ± 2.6 6.1 ± 2.0 0.924 5.2 ± 2.8 | |
TABLE 3 (Continued) Plasma TTV DNA levels at different post-transplant time points according to candidate SNPs.

| SNP (ID number) | Model     | Plasma TTV DNA level, log_{10} copies/mL (mean ± SD) | Baseline | p-value | Day 15 | p-value | Month 1 | p-value | Month 3 | p-value | Month 6 | p-value | Month 12 | p-value |
|----------------|-----------|-----------------------------------------------------|----------|---------|--------|---------|---------|---------|---------|---------|---------|---------|---------|---------|
|                | Baseline  |                                                    |          |         |        |         |         |         |         |         |         |         |         |         |
|                |           |                                                    |          |         |        |         |         |         |         |         |         |         |         |         |
| Recessive      | TT/TG     | 2.8 ± 1.6                                          | 3.1 ± 1.7| 0.583   | 4.4 ± 1.7| 0.913   | 5.8 ± 1.7| 0.227   | 5.3 ± 2.3| 0.610   | 4.6 ± 1.9| 0.682   |
|                | GG        | 3.3 ± 1.5                                          | 3.4 ± 1.1| 0.326   | 4.3 ± 1.1| 0.348   | 5.6 ± 2.0| 0.952   | 5.3 ± 1.9| 0.498   | 4.9 ± 1.7| 0.777   |
| Dominant       | TNF (rs1800629) | 2.9 ± 1.5                                        | 3.2 ± 1.6| 0.737   | 4.5 ± 1.7| 0.837   | 6.0 ± 2.0| 0.644   | 5.4 ± 2.3| 0.394   | 4.8 ± 1.7| 0.813   |
|                | GA/AA     | 2.7 ± 1.9                                          | 2.8 ± 1.7| 0.496   | 4.0 ± 1.6| 0.913   | 5.6 ± 2.0| 0.392   | 5.3 ± 2.3| 0.698   | 4.9 ± 1.7| 0.787   |
|                | Recessive | GG/GA                                              | 2.9 ± 1.6| 3.1 ± 1.6| 0.696   | 4.4 ± 1.7| 0.913   | 5.9 ± 1.7| 0.197   | 5.3 ± 2.3| 0.394   | 4.8 ± 1.7| 0.777   |
|                | AA        | 3.6 ± 1.6                                          | 3.7 ± 1.5| 0.858   | 5.1 ± 0.9| 0.371   | 6.4 ± 1.8| 0.682   | 5.4 ± 0.8| 0.682   | 4.6 ± 1.8| 0.682   |
| Dominant       | IL10 (rs1800872) | 2.6 ± 2.1                                        | 2.9 ± 1.2| 0.737   | 4.3 ± 1.5| 0.837   | 5.8 ± 1.7| 0.644   | 6.0 ± 2.0| 0.644   | 5.4 ± 1.7| 0.813   |
|                | TT        | 2.9 ± 1.6                                          | 3.2 ± 1.7| 0.496   | 4.4 ± 1.7| 0.913   | 5.9 ± 1.7| 0.197   | 5.4 ± 2.3| 0.394   | 5.0 ± 1.7| 0.813   |
|                | GG        | 3.3 ± 1.5                                          | 3.4 ± 1.1| 0.326   | 4.3 ± 1.1| 0.348   | 5.6 ± 2.0| 0.952   | 5.3 ± 1.9| 0.498   | 4.9 ± 1.7| 0.777   |
| Recessive      | IL10 (rs1878672) | 2.9 ± 1.6                                        | 3.2 ± 1.8| 0.696   | 4.4 ± 1.7| 0.913   | 5.9 ± 1.7| 0.197   | 5.4 ± 2.3| 0.394   | 4.8 ± 1.7| 0.813   |
|                | TT/TG     | 2.9 ± 1.6                                          | 3.2 ± 1.8| 0.696   | 4.4 ± 1.7| 0.913   | 5.9 ± 1.7| 0.197   | 5.4 ± 2.3| 0.394   | 4.8 ± 1.7| 0.813   |
|                | TG/CC     | 2.9 ± 1.6                                          | 3.2 ± 1.8| 0.696   | 4.4 ± 1.7| 0.913   | 5.9 ± 1.7| 0.197   | 5.4 ± 2.3| 0.394   | 4.8 ± 1.7| 0.813   |
|                | GG        | 3.3 ± 1.5                                          | 3.4 ± 1.1| 0.326   | 4.3 ± 1.1| 0.348   | 5.6 ± 2.0| 0.952   | 5.3 ± 1.9| 0.498   | 4.9 ± 1.7| 0.777   |
| Dominant       | IL12B (rs3212227) | 2.9 ± 1.6                                   | 3.2 ± 1.8| 0.696   | 4.4 ± 1.7| 0.913   | 5.9 ± 1.7| 0.197   | 5.4 ± 2.3| 0.394   | 4.8 ± 1.7| 0.813   |
|                | TT        | 2.9 ± 1.6                                          | 3.2 ± 1.8| 0.696   | 4.4 ± 1.7| 0.913   | 5.9 ± 1.7| 0.197   | 5.4 ± 2.3| 0.394   | 4.8 ± 1.7| 0.813   |
|                | TG/CC     | 2.8 ± 1.4                                          | 3.1 ± 1.4| 0.614   | 4.4 ± 1.6| 0.840   | 5.8 ± 1.8| 0.306   | 5.2 ± 2.3| 0.470   | 4.7 ± 2.0| 0.698   |
|                | Recessive | TT/TG                                              | 3.1 ± 1.7| 4.4 ± 1.7| 0.761   | 5.9 ± 1.7| 0.700   | 5.3 ± 2.3| 0.870   | 4.7 ± 1.9| 0.207   | 4.2 ± 1.7| 0.160   |
|                | GG        | 3.1 ± 1.0                                          | 4.3 ± 1.6| 0.60 ± 0.0| 5.6 ± 0.0| 0.930   | 5.3 ± 2.2| 0.268   | 5.3 ± 2.2| 0.268   | 5.1 ± 2.1| 0.268   |
| Dominant       | IL17A (rs2275913) | 2.7 ± 1.7                                         | 3.1 ± 1.6| 0.696   | 4.4 ± 1.7| 0.913   | 5.9 ± 1.7| 0.197   | 5.3 ± 2.3| 0.394   | 4.8 ± 1.7| 0.813   |
|                | GG        | 2.9 ± 1.6                                          | 3.2 ± 1.6| 0.696   | 4.4 ± 1.7| 0.913   | 5.9 ± 1.7| 0.197   | 5.4 ± 2.3| 0.394   | 4.8 ± 1.7| 0.813   |
|                | Recessive | GG/GA                                             | 2.9 ± 1.6| 3.2 ± 1.6| 0.696   | 4.4 ± 1.7| 0.913   | 5.9 ± 1.7| 0.197   | 5.4 ± 2.3| 0.394   | 4.8 ± 1.7| 0.813   |
|                | GA/AA     | 3.0 ± 1.6                                          | 3.2 ± 1.7| 0.696   | 4.4 ± 1.7| 0.913   | 5.9 ± 1.7| 0.197   | 5.4 ± 2.3| 0.394   | 4.8 ± 1.7| 0.813   |
|                | AA        | 2.6 ± 1.7                                          | 2.7 ± 1.9| 0.696   | 4.4 ± 1.7| 0.913   | 5.9 ± 1.7| 0.197   | 5.4 ± 2.3| 0.394   | 4.8 ± 1.7| 0.813   |

CTLA-4, cytotoxic T-lymphocyte antigen 4; IL, interleukin; SD, standard deviation; SNP, single-nucleotide polymorphism; TLR, toll-like receptor; TNF, tumor necrosis factor; TTV, torque teno virus.
| SNP (ID number) | Model | Genotype | Undetectable TTV DNAemia at baseline [n (%)] | p-value |
|----------------|-------|----------|--------------------------------------------|---------|
|                |       |          | No (n = 180)                  | Yes (n = 7) |
|                |       |          |                                |          |
| **CTLA4 (rs5742909)** | Dominant | CC       | 149 (82.8)                  | 6 (85.7)  | 0.840 |
|                |       | CT/TT    | 31 (17.2)                  | 1 (14.3)  | 0.690 |
|                | Recessive | CC/CT    | 176 (97.8)                  | 7 (100.0) | 0.394 |
|                |       | TT       | 4 (2.2)                    | 0 (0.0)   | 0.940 |
| **CTLA4 (rs231775)** | Dominant | AA       | 95 (52.8)                  | 3 (42.9)  | 0.606 |
|                |       | AG/GG    | 85 (47.2)                  | 4 (57.1)  | 0.394 |
|                | Recessive | AA/AG    | 163 (90.6)                 | 7 (100.0) | 0.394 |
|                |       | GG       | 17 (9.4)                   | 0 (0.0)   | 0.940 |
| **TLR3 (rs3775291)** | Dominant | CC       | 88 (48.9)                  | 0 (0.0)   | 0.015 |
|                |       | CT/TT    | 92 (51.1)                  | 7 (100.0) | 0.015 |
|                | Recessive | CC/CT    | 145 (80.6)                 | 5 (71.4)  | 0.304 |
|                |       | TT       | 26 (14.9)                  | 2 (28.6)  | 0.286 |
| **TLR9 (rs5743836)** | Dominant | AA       | 130 (72.2)                 | 4 (57.1)  | 0.385 |
|                |       | AG/GG    | 50 (27.8)                  | 3 (42.9)  | 0.385 |
|                | Recessive | AA/AG    | 171 (95)                  | 7 (100.0) | 0.345 |
|                |       | GG       | 9 (5)                      | 0 (0.0)   | 0.050 |
| **TLR9 (rs352139)** | Dominant | TT       | 47 (26.1)                  | 3 (42.9)  | 0.326 |
|                |       | TC/CC    | 133 (73.9)                 | 4 (57.1)  | 0.405 |
|                | Recessive | TT/TC    | 129 (71.7)                 | 4 (57.1)  | 0.405 |
|                |       | CC       | 51 (28.3)                  | 3 (42.9)  | 0.345 |
| **CD209 (rs735240)** | Dominant | GG       | 48 (26.7)                  | 3 (42.9)  | 0.345 |
|                |       | GA/AA    | 132 (73.3)                 | 4 (57.1)  | 0.500 |
|                | Recessive | GG/GA    | 134 (74.4)                 | 6 (4.3)   | 0.500 |
|                |       | AA       | 46 (25.6)                  | 1 (2.1)   | 0.286 |
| **CD209 (rs4804803)** | Dominant | AA       | 105 (58.3)                 | 3 (42.9)  | 0.416 |
|                |       | AG/GG    | 75 (41.7)                  | 4 (57.1)  | 0.416 |
|                | Recessive | AA/AG    | 175 (97.2)                 | 4 (57.1)  | 0.0017 |
|                |       | GG       | 5 (2.8)                    | 3 (42.9)  | 0.258 |
| **IFNL3 (rs12979860)** | Dominant | CC       | 83 (46.1)                  | 3 (42.9)  | 0.865 |
|                |       | CT/TT    | 97 (53.9)                  | 4 (57.1)  | 0.258 |
|                | Recessive | CC/CT    | 152 (84.4)                 | 7 (4.4)   | 0.258 |
|                |       | TT       | 28 (15.6)                  | 0 (0.0)   | 0.258 |
| **IFNL3 (rs8999917)** | Dominant | TT       | 129 (71.7)                 | 3 (42.9)  | 0.101 |
|                |       | TG/GG    | 51 (28.3)                  | 4 (57.1)  | 0.569 |
|                | Recessive | TT/TG    | 172 (95.6)                 | 7 (100.0) | 0.569 |
|                |       | GG       | 8 (4.4)                    | 0 (0.0)   | 0.050 |
| **TNF (rs1800629)** | Dominant | GG       | 137 (76.1)                 | 4 (57.1)  | 0.253 |
|                |       | GA/AA    | 43 (23.9)                  | 3 (42.9)  | 0.253 |
|                | Recessive | GG/GA    | 178 (98.9)                 | 7 (3.8)   | 0.779 |
|                |       | AA       | 2 (1.1)                    | 0 (0.0)   | 0.779 |
| **IL10 (rs1800872)** | Dominant | TT       | 16 (8.9)                   | 2 (28.6)  | 0.083 |
|                |       | TG/GG    | 164 (91.1)                 | 5 (71.4)  | 0.083 |
|                | Recessive | TT/TG    | 109 (60.6)                 | 4 (57.1)  | 0.856 |
|                |       | GG       | 71 (39.4)                  | 3 (42.9)  | 0.856 |

(Continued on following page)
TABLE 4 (Continued) Association between undetectable TTV DNAemia at the baseline (pre-transplant) assessment and candidate SNPs in dominant (heterozygous and homozygous) and recessive (homozygous only) models.

| SNP (ID number) | Model | Genotype | Undetectable TTV DNAemia at baseline [n (%)] | p-value |
|-----------------|-------|----------|---------------------------------------------|---------|
|                 |       |          | No (n = 180) | Yes (n = 7) |
| **IL10 (rs1878672)** | Dominant | GG       | 66 (36.7) | 3 (42.9) | 0.739 |
|                 |       | GC/CC    | 114 (63.3) | 4 (57.1)  |
|                 | Recessive | GG/GC    | 159 (88.3) | 4 (57.1)  | 0.046 |
|                 |       | CC       | 21 (11.7)  | 3 (42.9)  |
| **IL12B (rs3212227)** | Dominant | TT       | 89 (49.4)  | 4 (57.1)  | 0.689 |
|                 |       | TG/GG    | 91 (50.6)  | 3 (42.9)  |
|                 | Recessive | TT/TG    | 165 (91.7) | 7 (100.0) | 0.426 |
|                 |       | GG       | 15 (8.3)   | 0 (0.0)   |
| **IL17A (rs2275913)** | Dominant | GG       | 76 (42.2)  | 4 (57.1)  | 0.434 |
|                 |       | GA/AA    | 104 (57.8) | 3 (42.9)  |
|                 | Recessive | GG/GA    | 157 (87.2) | 5 (71.4)  | 0.228 |
|                 |       | AA       | 23 (12.8)  | 2 (28.6)  |

CTLA-4, cytotoxic T-lymphocyte antigen 4; IL, interleukin; SNP, single-nucleotide polymorphism; TLR, toll-like receptor; TNF, tumor necrosis factor; TTV, torque teno virus.

undetectable TTV DNAemia before transplantation [37.5% (5/8) versus 2.2% (4/179) for GG and AA/AG carriers; nominal p-value = 0.0017]. Nevertheless, it should be noted that only the latter association was below the Bonferroni-corrected p-value threshold for statistical significance (which was settled at 0.00178) (Table 4). We further assessed whether the impact of the CD209 (rs4804803) SNP remained significant after adjusting for recipient demographics and pre-transplant clinical characteristics also associated with undetectable TTV DNAemia at baseline (Supplementary Table S2). In a logistic regression model that included recipient age and previous renal replacement therapy as covariates, the presence of the minor G allele of CD209 (rs4804803) in the homozygous state was still significantly associated with pre-transplant TTV DNA levels below the LLoD (adjusted OR: 36.96; 95% CI: 4.72–289.67; p-value = 0.001).

In order to better characterize the genetic determinants of post-transplant TTV viral kinetics, we compared peak TTV DNA levels through different time intervals according to candidate SNPs. The only apparent correlation was observed for TNF (rs1800629), with carriers of the minor allele either in the heterozygous or homozygous state showing lower peak levels during the first 3 post-transplant months (4.2 ± 1.5 versus 5.0 ± 1.8 log10 copies/mL for GA/AA and GG carriers; nominal p-value = 0.008) (Supplementary Table S3). This comparison, however, did not attain the Bonferroni-corrected significance level (settled at 0.00059). Accordingly, recipients bearing the minor A allele of this SNP also showed a non-significant trend—by applying the Bonferroni correction—towards a lower AUC for plasma TTV DNAemia through month 6 (5.8 ± 1.7 versus 6.8 ± 1.7 log10 copies/mL for GA/AA and GG carriers; nominal p-value = 0.007) (Supplementary Table S4).

Finally, no significant differences at the Bonferroni-adjusted α level were found in increments (Δ) in TTV DNA levels from baseline to day 15 and months 1, 3, 6 and 12 after transplantation either (Supplementary Table S5).

**Discussion**

There is increasing evidence on the usefulness of TTV as a surrogate marker of the immune status in a variety of clinical scenarios (Martin-Lopez et al., 2020; Honorato et al., 2021; Studenic et al., 2021), in particular SOT (Fernandez-Ruiz et al., 2019; Redondo et al., 2022a; Eldar-Yedidia et al., 2022; Jaksch et al., 2022), under the rationale that the T-cell-mediated immunity plays an instrumental role in controlling viral replication. Nevertheless, the relative contribution of the innate system—and its genetic determinants—has not been characterized so far. To our knowledge only three previous works have analyzed the impact of genetic polymorphisms on TTV replication in HSCT recipients (with two studies from the same group) and people living with human immunodeficiency virus (HIV) (Prasetyo et al., 2015; Ramzi et al., 2019; Ramzi et al., 2021). Ramzi et al. (2019); Ramzi et al. (2021) found a correlation between SNPs in IL10, CTLA4 and TNF genes and TTV infection in allogeneic HSCT recipients. In detail, the heterozygote genotypes of IL10 rs1878672 (−592C/A) and CTLA4 rs231775 (+49 A/G) were associated with a higher prevalence of TTV DNAemia, whereas the A allele of TNF rs1800629 (−308G/A) had a protective effect. On the other hand, Prasetyo et al. reported a correlation between the APOBEC3B deletion polymorphism...
status and TTV, HBV and HCV infection among HIV patients (Prasetyo et al., 2015; Ramzi et al., 2019; Ramzi et al., 2021). The present investigation is the first to evaluate to what extent the kinetics of TTV DNA levels following KT are influenced by SNPs in genes coding for PRRs (TLR3, TLR9 and CD209), ILs and cytokines (IL-12B, IL-17, IL-10, TNF), IFN-γ (IL-28B) and the costimulatory receptor CTLA4. These candidate SNPs were chosen on the basis of prior studies performed in the HSCT population (Ramzi et al., 2019; Ramzi et al., 2021) or due to their well-established involvement in other viral infections in SOT recipients (Redondo et al., 2022d).

We found no clear association between any of the SNP genotypes considered and various parameters reflecting viral kinetics after transplantation, such as mean and peak DNA levels or AUCs for plasma TTV DNAemia during discrete periods, or absolute increments from baseline. The significant differences observed for the minor A allele of TNF (rs1800629) in terms of lower peak DNA levels and AUC differences observed for the minor A allele of CD209 (rs4804803) after KT (Redondo et al., 2022c). In addition, the minor G allele of CD209 (rs4804803) SNP was associated with undetectable TTV DNAemia in a single-center cohort of HSCT recipients (OR: 0.46; 95% CI: 0.22–0.96; p-value = 0.025), although the timing for monitoring was unclear and no correction for multiple testing was performed. Interestingly, Ramzi et al. (2010) reported that the A allele of the TNF (rs1800629) SNP was associated with undetectable TTV DNAemia in a single-center cohort of HSCT recipients (OR: 0.46; 95% CI: 0.22–0.96; p-value = 0.025), although the timing for monitoring was unclear and no correction for multiple testing was performed. In line with our results, the same group observed no apparent impact of genotypes of CTLA4 (rs5742909) on the incidence of TTV infection after HSCT (Ramzi et al., 2019; Ramzi et al., 2021).

In addition to the longitudinal post-transplant monitoring of TTV replication, we have specifically investigated the associations between candidate SNPs and the presence of TTV infection at the baseline assessment, before immunosuppressive therapy was initiated. The cross-sectional comparison at this time point would reveal the potential role of genetic predisposition to TTV among ESRD patients in the absence of iatrogenic immunosuppression. In contrast to the negative results observed for the post-transplant period, we found that the minor G allele of CD209 (rs4804803) SNP was associated with undetectable TTV DNAemia in a single-center cohort of HSCT recipients (OR: 0.46; 95% CI: 0.22–0.96; p-value = 0.025), although the timing for monitoring was unclear and no correction for multiple testing was performed. In line with our results, the same group observed no apparent impact of genotypes of CTLA4 (rs5742909) on the incidence of TTV infection after HSCT (Ramzi et al., 2019; Ramzi et al., 2021).

Data availability statement

The data that support the findings of this study are available from the corresponding author upon reasonable request. SNP genotyping data are registered in the BioProject database under the ID PRJNA898147.

Ethics statement

The studies involving human participants were reviewed and approved by the Clinical Research Ethics Committee Hospital
12 de Octubre (Study protocol number 14/030). The patients/ participants provided their written informed consent to participate in this study.

Author contributions

NR and MF-R designed the study, performed statistical analyses and wrote the manuscript; EA, PP, and EG performed laboratory analyses; TR-M collected patient samples; IR-G, FL-M, RS, EG, NP, and AA participated in patient recruitment and performed data collection; AA, DN, and JMA critically reviewed the manuscript and provided significant input and feedback. All authors read and approved the final manuscript.

Funding

This study has been funded by Instituto de Salud Carlos III (ISICIII), Spanish Ministry of Science and Innovation, through the projects PIE13/00045, PI15/01953, and P119/01300—co-founded by European Regional Development Fund/European Social Fund “A way to make Europe”/“Investing in your future”. IR-G holds a research training contract “Rio Hortega” (CM19/00163) and MF-R holds a research contract “Miguel Servei” (CP18/00073), both from the ISICIII and also co-funded by the European Union.

References

Alagarasu, K., Kaushal, H., Shinde, P., Kakade, M., Chaudhary, U., Padhidiri, V., et al. (2021). TNFA and IL10 polymorphisms and IL-6 and IL-10 levels influence disease severity in influenza A(H1N1)pdm09 virus infected patients. Gene. 52 (12), 2019. doi:10.3390/genes12121914

Ali, E. S. G., Bassouni, R. H., Abdedaleem, O. O., Hassan, E. A., and Gaber, S. N. (2022). Association between SNPs of Cytotoxic T-lymphocyte-associated protein 4 (CTLA-4), programmed cell death 1 (PD-1) and the susceptibility to chronic Hepatitis C infection in virus C-infected patients. Virus Res. 310, 198684. doi:10.1016/j.viruses.2022.198684

Arze, C. A., Springer, S., Dudas, G., Patel, S., Bhattacharyya, A., Swaminathan, H., et al. (2021). Global genome analysis reveals a vast and dynamic anellovirus landscape within the human virome. Cell. Host Microbe 29 (8), 1305–1313.e6. doi:10.1016/j.chom.2021.07.001

Biagini, P. (2009). Classification of TTV and related viruses (anelloviruses). Curr. Top. Microbiol. Immunol. 331, 21–33. doi:10.1007/978-3-540-70972-9_2

Bravo, D., Solano, C., Gimenez, E., Remigia, M. J., Corrales, I., Amat, P., et al. (2014). Effect of the IL28B Rs12980352 C/T polymorphism on the incidence and features of active cytomegalovirus infection in allogeneic stem cell transplant patients. J. Med. Virol. 86 (5), 838–844. doi:10.1002/jmv.23865

Bucardo, F., Reyes, Y., Morales, M., Brinceno, R., Gonzalez, F., Lundkvist, A., et al. (2021). Association of genetic polymorphisms in DC-SIGN, toll-like receptor 3, and tumor necrosis factor alpha genes and the Lewis-negative phenotype with chikungunya infection and disease in Nicaragua. J. Infect. Dis. 223 (2), 278–286. doi:10.1093/infdis/jiaa364

Chaitthanyan, I. K., Muruganandan, N., Surya, P., Arwesh, M., Alagarasu, K., and Vijayarachy, P. (2016). Association of oligoadenylate synthetase gene cluster and DC-SIGN (CD209) gene polymorphisms with clinical symptoms in chikungunya virus infection. DNA Cell. Biol. 35 (1), 44–50. doi:10.1089/dna.2015.2819

Chihah, H., Zaidane, I., Elhabazi, A., Jaidid, F. Z., El Fihri, R., Elmesnaouidi-Idrissi, M., et al. (2019). Toll-like receptor 9 polymorphisms and Hepatitis B virus clearance in Moroccan chronic carriers. Gene. 607, 212–218. doi:10.1016/j.gene.2018.11.041

Corrales, J., Gimenez, E., Solano, C., Amat, P., de la Camara, R., Nieto, J., et al. (2015). Incidence and dynamics of active cytomegalovirus infection in allogeneic stem cell transplant patients according to single nucleotide polymorphisms in donor and recipient CCR5, MCP-1, IL-10, and TLR9 genes. J. Med. Virol. 87 (2), 248–255. doi:10.1002/jmv.24050

Czupryna, P., Parczewski, M., Gryniewicz, S., Pacwicz, S., Zajkowska, J., Dunaj, J., et al. (2017). Analysis of the relationship between single nucleotide polymorphism of the CD209, IL-10, IL-28 and CCR5 D32 genes with the human predisposition to developing tick-borne encephalitis. Postepy Hig. Med. Dosw. 71 (1), 788–796. doi:10.5604/201.0001.0010.03856

Dodi, G., Attanasi, M., Di Filippo, P., Di Pillo, S., and Chiarelli, F. (2021). Virome in the lungs: The role of anelloviruses in childhood respiratory diseases. Microorganisms 9 (7), 1357. doi:10.3390/microorganisms9071357

Egli, A., Levin, A., Santer, D. M., Joyce, M., O’Shea, D., Thomas, B. S., et al. (2014). Immuno-modulatory Function of Interleukin 28B during primary infection with cytomegalovirus. J. Infect. Dis. 210 (5), 717–727. doi:10.1093/infdis/jiu14

Eldar-Yedidia, Y., Ben-Shalom, E., Hillel, M., Belostotsky, R., Megged, O., Freier-Dror, Y., et al. (2022). Association of post-transplantation anellovirus viral load with kidney transplant rejection in children. Pediatr. Nephrol. 37, 1905–1914. doi:10.1007/s00467-021-05356-w

Eskandari-Nasab, E., Moghadampour, M., Tahmassebi, A., and Asadi-Saghandi, A. (2018). Interleukin-17 A and F gene polymorphisms affect the risk of tuberculosis: An updated meta-analysis. Indian J. Tuberc. 65 (3), 200–207. doi:10.1016/j.ijtb.2017.08.027

Fernandez-Ruiz, M., Albert, E., Gimenez, E., Ruiz-Merlo, T., Parra, P., Lopez-Medrano, F., et al. (2019). Monitoring of alphatorquevirus DNA

Acknowledgments

The authors gratefully acknowledge all patients recruited in the institutional cohort of kidney transplant recipients for their participation.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher’s note

All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fgene.2022.1069890/full#supplementary-material
levels for the prediction of immunosuppression-related complications after kidney transplantation. Am. J. Transpl. 19 (4), 1139–1149. doi:10.1111/ajt.15145

Fernandez-Ruiz, M., Corrales, I., Arias, M., Campistol, J., M., Gimenez, E., Creepo, J., et al. (2015). Association between individual and combined SNPs in genes related to innate immunity and incidence of CMV infection in seropositive kidney transplant recipients. Am. J. Transpl. 15 (5), 1323–1335. doi:10.1002/ajt.13107

Fischer, J., Kookoski, E., Schott, E., Fulop, B., Heyne, R., Berg, T., et al. (2018). Polymorphisms in the Toll-like receptor 3 (TLR3) gene are associated with the natural course of Hepatitis B virus infection in Caucasian population. Sci. Rep. 8 (1), 12737. doi:10.1038/s41598-018-31065-6

Focosi, D., Antonelli, G., Ptasleko, M., and Maggi, F. (2016). Torquepoxivirus: The human virome from bench to bedside. Clin. Microbiol. Infect. 22 (7), 589–593. doi:10.1016/j.cmi.2016.04.007

Ge, D., Fellay, J., Thompson, A. J., Simon, J. S., Shianna, K. V., Urban, T. J., et al. (2009). Genetic variation in IL28B predicts hepatitis C treatment-induced viral clearance. Nature 461 (7262), 399–401. doi:10.1038/nature08309

Geng, P. L., Song, L. X., An, H., Huang, J. Y., Li, S., and Zeng, X. T. (2016). Toll-like receptor 3 is associated with the risk of HCV infection and HBV-related diseases. Med. Baltim. 95 (21), e2302. doi:10.1097/MDB.0000000000002302

Goncalves de Albuquerque, S. D. C., da Costa Oliveira, C. N., Vairaktaris-Antao, V., Silva, A. C., Luna, C. F., de Lorena, V. M. B., et al. (2019). Study of association of the rs2275913 IL-17A single nucleotide polymorphism and susceptibility to cutaneous leishmaniasis caused by Leishmania braziliensis. Cytoine 123, 154784. doi:10.1016/j.cyto.2019.154784

Gorz, I., Haloscham, M., Jakoch, P., Klepetko, W., and Puchhammer-Stickl, E. (2014). Plasma DNA levels of Torque teno virus and immunosuppression after lung transplantation. J. Heart Lung Transpl. 33 (3), 320–323. doi:10.1016/j.healun.2013.12.007

Gupta, K., Kaur, G., Pathak, T., and Banerjee, I. (2022). Systematic review and meta-analysis of human genetic variants contributing to COVID-19 susceptibility and severity. Gene 768. doi:10.1016/j.gene.2021.149281

Honorato, L., Witkin, S. S., Mendes-Correa, M. C., Conde Toscano, A. L., Li, M., da Paula, A. V., et al. (2021). The torque teno virus titer in saliva reflects the level of circulating CD4(+) T lymphocytes and HIV in individuals undergoing antiretroviral maintenance therapy. Front. Pathol. Med. 8, 809312. doi:10.3898/fpm.2021.809312

Jablonska, A., Studzinska, M., Szenborn, L., Wisniewska-Ligier, M., Herytar, S., et al. (2015). The APOBEC3B deletion polymorphism is associated with prevalence of Hepatitis B virus, hepatitis C virus, Torque Teno Virus, and Toxoplasma gondii co-infection among HIV-infected individuals. J. Clin. Virol. 76, 67–71. doi:10.1016/j.jcv.2015.07.009

Ramzi, M., Arandi, N., Zarei, T., Saadi, M. I., Yaghobi, R., Moghadam, M., et al. (2019). Genetic variation of TNF-alpha and IL-10, IL-17 genes and association with torque teno virus infection post hematopoietic stem cell transplantation. Acta Virol. 63 (2), 186–194. doi:10.14499/av.2019.210

Ramzi, M., Iravani Saadi, M., Zarei, T., Yaghobi, R., and Arandi, N. (2021). Association between cytotoxic T-lymphocyte antigen 4 gene polymorphisms and torque teno virus infection after hematopoietic stem cell transplantation. Exp. Clin. Transpl. 19 (3), 259–263. doi:10.6002/ect.2017.0105

Redondo, N., Navarro, D., Aguado, J. M., and Fernandez-Ruiz, M. (2022). Human genetic polymorphisms and risk of viral infection after solid organ transplantation. Transpl. Res. 36 (1), 100669. doi:10.1016/jŕtr.2021.100669

Redondo, N., Navarro, D., Aguado, J. M., and Fernandez-Ruiz, M. (2022). Viruses, friends, and foes: The case of Torque Teno Virus and the net state of immunosuppression. Transpl. Infect. Dis. 24 (2), e13778. doi:10.1111/tid.13778

Redondo, N., Gonzalez-Roncer, I., Parra, P., Lopez-Medrano, F., Gonzalez, E., Hernandez, A., et al. (2022). Genetic polymorphisms in TLR3, IL10 and TRAF1 polymorphisms are associated with severity of dengue disease. Front. Immunol. 1346. doi:10.3389/fimmu.2022.88995

Redondo, N., Rodriguez-Ruiz, M., Parra, P., Lopez-Medrano, F., Gonzalez, E., Hernandez, A., et al. (2022). Genetic polymorphisms in TLR3, IL10 and TRAF1 influence the risk of BK polyomavirus infection after kidney transplantation. Scien. Rep. 12 (1), 11328. doi:10.1038/s41598-022-15406-0

Singh, A. K., Prakash, S., Garg, R. K., Jain, P., Kumar, R., and Iain, A. (2021). Study of single nucleotide polymorphisms in endosomal toll-like receptors-3, 7, and 9 genes in patients with dengue: A case-control study. Cureus 13 (5), e4883. doi:10.7554/cureus.14883

Studenic, P., Bond, G., Korschbaumer, A., Becede, M., Pavelka, K., Karateev, D., et al. (2021). Torque Teno Virus quantification for monitoring of immunosuppression with biologic compounds in the treatment of rheumatoid arthritis. Rheumatol. Ox. 61, 2815–2825. doi:10.1093/rheumatology/keh839
Studzinska, M., Jablonska, A., Wisniewska-Ligier, M., Nowakowska, D., Gaj, Z., Lesnikowski, Z. J., et al. (2017). Association of TLR3 L412F polymorphism with cytomegalovirus infection in children. *PLoS One* 12 (1), e0169420. doi:10.1371/journal.pone.0169420

Takeuchi, O., and Akira, S. (2010). Pattern recognition receptors and inflammation. *Cell.* 140 (6), 805–820. doi:10.1016/j.cell.2010.01.022

Thomas, D. L., Thio, C. L., Martin, M. P., Qi, Y., Ge, D., O’Huigin, C., et al. (2009). Genetic variation in IL28B and spontaneous clearance of hepatitis C virus. *Nature* 461 (7265), 798–801. doi:10.1038/nature08463

Vannberg, F. O., Chapman, S. J., Khor, C. C., Tosh, K., Floyd, S., Jackson-Sillah, D., et al. (2008). CD209 genetic polymorphism and tuberculosis disease. *PLoS One* 3 (1), e1388. doi:10.1371/journal.pone.0001388

Vargas-Castillo, A. B., Ruiz-Tovar, K., Vivanco-Cid, H., Quirce-Cruz, S., Escobar-Gutierrez, A., Cerna-Cortes, J. F., et al. (2018). Association of single-nucleotide polymorphisms in immune-related genes with development of dengue hemorrhagic fever in a Mexican population. *Viral Immunol.* 31 (3), 249–255. doi:10.1089/vim.2017.0069

Varshney, D., Singh, S., Sinha, E., Mohanty, K. K., Kumar, S., Kumar Barik, S., et al. (2022). Systematic review and meta-analysis of human Toll-like receptors genetic polymorphisms for susceptibility to tuberculosis infection. *Cytokine* 152, 155791. doi:10.1016/j.cyto.2021.155791

Webb, B., Rakibuzzaman, A., and Ramamoorthy, S. (2020). Torque teno viruses in health and disease. *Virus Res.* 285, 198013. doi:10.1016/j.virusres.2020.198013

Ye, S., Zhang, X., Zhang, Y. B., Tian, X., Liu, A., Cui, C., et al. (2020). Association of TLR3 (rs3775291) and IL-10 (rs1800871) gene polymorphisms with susceptibility to hepatitis B infection: A meta-analysis. *Epidemiol. Infect.* 148, e228. doi:10.1017/S0950268820002101