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LETTER TO THE EDITOR

T cell receptor sequencing reveals reduced clonal breadth of T-cell responses against SARS-CoV-2 after natural infection and vaccination in allogeneic hematopoietic stem cell transplant recipients

Allogeneic hematopoietic stem cell transplantation (HSCT) recipients have a higher risk of developing severe coronavirus disease (COVID-19) and a higher mortality rate compared with the general population (Ljungman et al.¹) potentially also as a consequence of their reduced ability to respond to vaccination (Mamez et al.²; Redjoul et al.³; Einarsdottir et al.⁴). To evaluate the magnitude and breadth of T-cell responses against SARS-CoV-2 in allogeneic HSCT recipients, we carried out high-throughput T cell receptor (TCR) repertoire profiling on cells recovered from allogeneic HSCT recipients or healthy controls (HC) after COVID-19 natural infection or messenger RNA (mRNA)-based vaccination.

Peripheral blood samples were obtained after COVID-19 infection from allogeneic HSCT recipients ($n = 11$; Supplementary Tables S1 and S2, available at <https://doi.org/10.1016/j.annonc.2022.09.153>) or HC ($n = 10$; Supplementary Material, available at <https://doi.org/10.1016/j.annonc.2022.09.153>). A total of 6 out of 11 patients were under immunosuppression for active ($n = 3$) or resolved ($n = 3$) graft-versus-host disease. T-cell receptor (TCR) beta sequencing (Supplementary Material, available at <https://doi.org/10.1016/j.annonc.2022.09.153>) identified SARS-CoV-2 specific T-cell clonotypes in both HC and HSCT recipients after COVID-19 infection (Figure 1A). No difference was observed in the proportion of T cells specific for SARS-CoV-2 between HSCT recipients and HC (data not shown). The diversity of the SARS-CoV-2-specific T-cell clonotypes, a measure previously shown to be inversely associated with severity of the disease (Elyanow et al.⁵), however, was significantly reduced in HSCT recipients compared with HC (Figure 1B). Enzyme-Linked ImmunoSpot (ELISpot) assay (Supplementary Material, available at <https://doi.org/10.1016/j.annonc.2022.09.153>) showed significantly lower numbers of interferon- γ (IFN- γ) spot forming units (SFU) after stimulation of PBMCs from HSCT recipients with peptides from both the SARS-CoV-2 spike (S) protein (Figure 1B, upper panel) and the membrane glycoprotein (M) plus the nucleocapsid phosphoprotein (N) proteins (data not shown) compared with HC. A significant positive correlation between SARS-CoV-2-specific T-cell clonotypes and IFN- γ SFU was observed (Figure 1B, upper panel). Conversely, we detected no significant difference in anti-S immunoglobulin G (IgG) titers and no correlation between antibody titers and different clonotypes (Figure 1B, middle panel). HSCT recipients displayed a less

diverse TCR repertoire compared with HC as revealed by higher Simpson clonality and the Simpson clonality negatively correlated with the number of different SARS-CoV-2-specific T-cell clonotypes (Figure 1B, lower panel).

We next carried out the same analysis on samples recovered from allogeneic HSCT recipients ($n = 11$; Supplementary Tables S1 and S2, available at <https://doi.org/10.1016/j.annonc.2022.09.153>) or from healthy controls ($n = 10$) after vaccination with three doses of mRNA-based SARS-CoV-2 vaccines (Supplementary Material, available at <https://doi.org/10.1016/j.annonc.2022.09.153>). We observed a significant reduction in different S-protein-specific T-cell clonotypes in allogeneic HSCT recipients compared with HC (Figure 1C and D). ELISpot analysis revealed significantly lower numbers of IFN- γ SFU in HSCT recipients compared with HC and a slightly significant positive correlation between the ELISpot and the TCR-seq results (Figure 1D, upper panel). We observed slightly reduced anti-S titers in HSCT recipients compared with HC and a trend toward a positive correlation between S-specific clonotypes and anti-S titers (Figure 1D, middle panels). We detected a negative correlation between the Simpson clonality and the number of different S-protein-specific T-cell clonotypes after vaccination (Figure 1D).

Our results indicate that allogeneic HSCT recipients display reduced breadth of SARS-CoV-2-specific T-cell clonotypes after COVID-19 infection and vaccination. No clear correlation was detected between TCR clonal breadth and anti-S IgG titers. The clonal breadth defect was associated with increased T-cell clonality after HSCT, pointing to the reduced diversity of the TCR repertoire as a mechanism leading to impaired cellular responses against SARS-CoV-2 in HSCT recipients.

A. Pradier^{1,2,†}, AC. Mamez^{1,*†}, C. Stephan¹, F. Giannotti¹, S. Masouridi-Levrat¹, S. Wang², S. Morin¹, D. Neofytos³, D. L. Vu³, A. Melotti², I. Arm⁴, C. S. Eberhardt⁵, J. Tamburini², L. Kaiser^{3,4,6}, Y. Chalandon^{1,2} & F. Simonetta^{1,2*}

¹Division of Hematology, Department of Oncology, Geneva University Hospitals, University of Geneva, Geneva;

²Translational Research Centre in Onco-Haematology, Faculty of Medicine, University of Geneva, Geneva;

³Division of Infectious Diseases, Geneva University Hospitals, Geneva;

⁴Laboratory of Virology, Division of Infectious Diseases and Division of Laboratory Medicine, University Hospitals of Geneva & Faculty of Medicine, University of Geneva, Geneva;

⁵Center for Vaccinology, University Hospitals of Geneva Division of General Pediatrics, Department of Woman, Child and Adolescent Medicine, Faculty of Medicine, University of Geneva, Geneva;

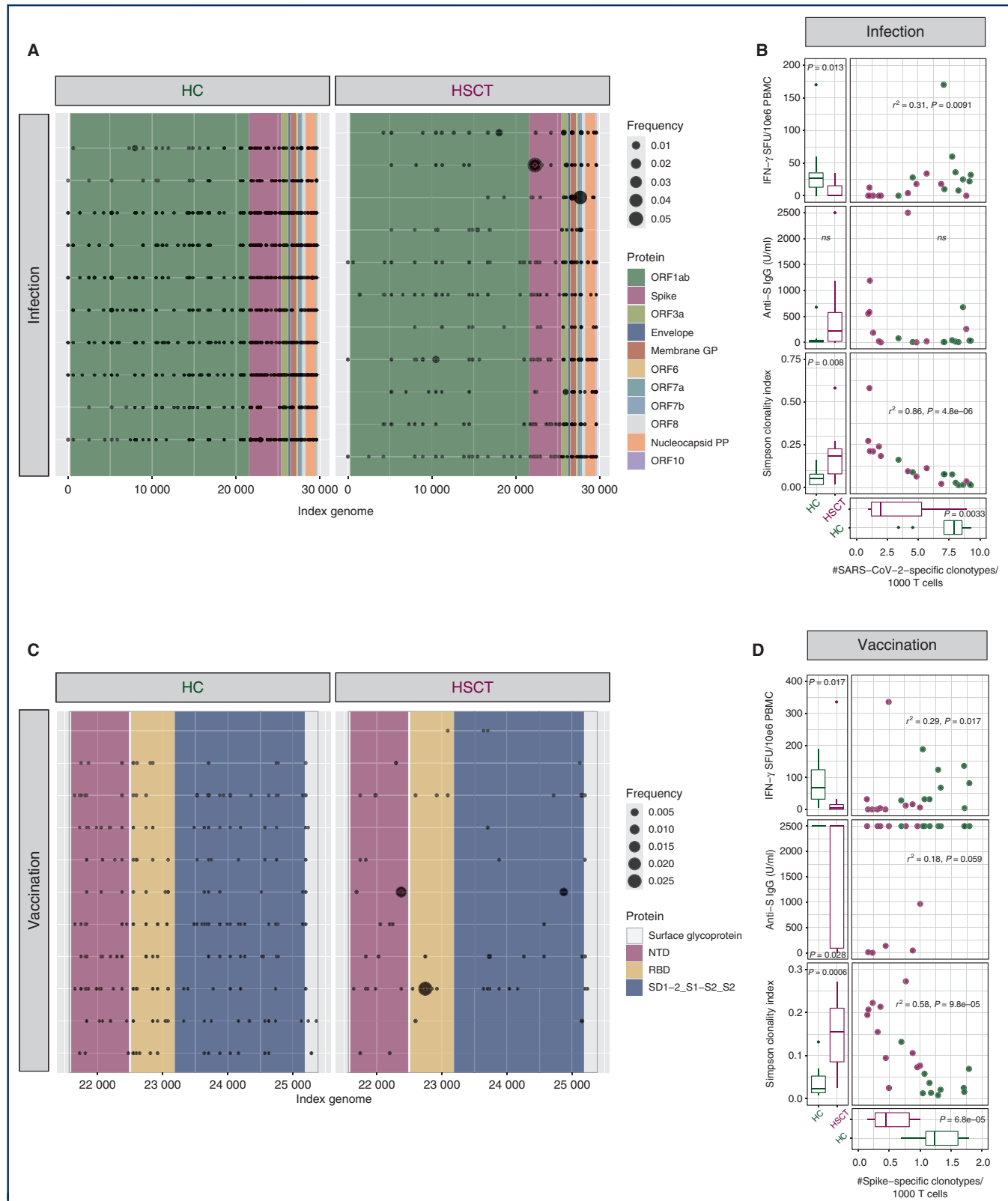


Figure 1. Reduced SARS-CoV2-specific T-cell clonotypes after COVID-19 infection and vaccination in allogeneic HSCT recipients. (A, C) SARS-CoV-2-specific T-cell clonotypes visualized based on the putative sequence of the SARS-CoV-2 genome recognized. (B, D) Scatter plots and marginal bar plots correlating and comparing the number of different SARS-CoV-2-specific T-cell clonotypes/1000 T cells, the anti-S IFN- γ SFU, the anti-S IgG titers and the Simpson clonality index in HC and HSCT. Differences between groups were assessed using the Mann–Whitney U test. Correlations were evaluated using a Spearman rank correlation coefficient test. GP, glycoprotein; HC, healthy controls; HSCT, hematopoietic stem cell transplantation; IFN, interferon; IgG, immunoglobulin G; NTD, N-terminal domain; PP, phosphoprotein; RBD, receptor binding domain; SFU, spot forming units.

⁶Geneva Centre for Emerging Viral Diseases, Geneva University Hospitals, Geneva, Switzerland
 (*E-mail: anne-claire.mamez@hcuge.ch)
 (*E-mail: federico.simonetta@unige.ch).

[†]These authors contributed equally to this work.

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DISCLOSURE

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