Monitoring of naturally acquired and vaccine-induced SARS-CoV-2 specific cellular immune responses

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Introduction

The global outbreak of the COVID-19 pandemic has emphasized the importance of immune monitoring technologies to reveal the dynamics of the immunological response in naturally acquired immunity, vaccination, and hybrid immunity. Immune monitoring technologies can help provide guidance to clinicians and health authorities about the type, magnitude, and duration of cellular immunity and thus improve the decision-making about future vaccine programs and roll out of booster doses.

Our aim was to develop SARS-CoV-2-specific immune monitoring assays based on the Dextramer® technology to detect and characterize virus-specific T cell responses. The applicability of the Dextramer assay for immune monitoring in SARS-CoV-2 was demonstrated by a longitudinal case study on an individual with hybrid immunity.

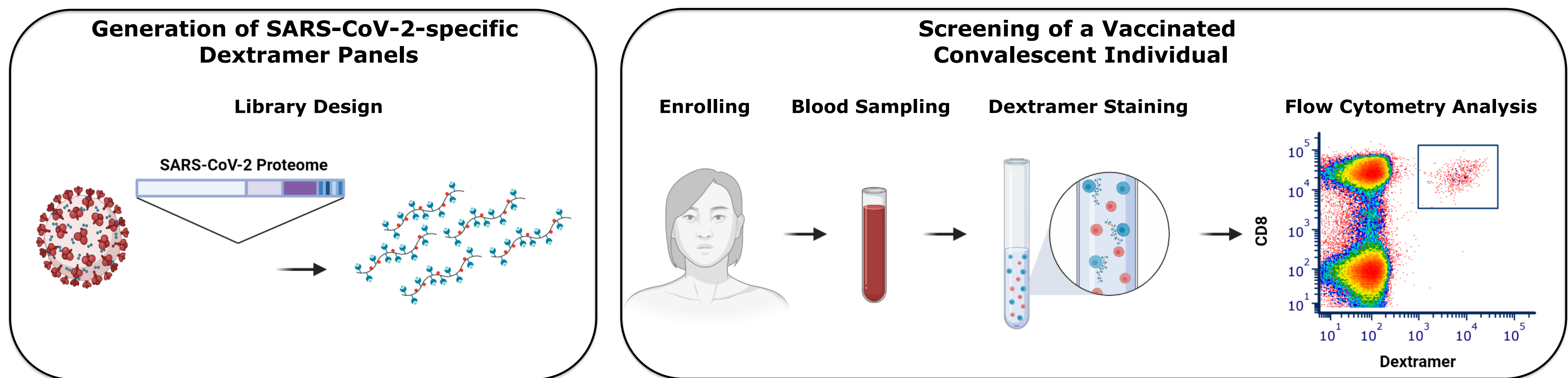## Detection of the longitudinal T-cell responses in SARS-CoV-2 by Dextramer assay

Figure 1. Schematic overview of the experimental workflow to Dextramer staining of the CD8+ T-cell response to SARS-CoV-2. To detect SARS-CoV-2-specific CD8+ T cells in peripheral blood mononuclear cells (PBMCs), a SARS-CoV-2 MHC Dextramer assay was developed. The assay was designed to cover eight of the most common class I human leukocyte antigen (HLA) alleles including A*01:01, A*02:01, A*03:01, A*11:01, A*24:02, B*07:02, B*35:01 and B*44:02 complexed to epitopes from Spike and Non-Spike (Nucleocapsid, ORF1ab and ORF3a) proteins of SARS-CoV-2. To evaluate the clinical performance of the assay, consecutive PBMC samples were collected from a convalescent vaccinated individual after infection and after 1st and 2nd vaccination. A PBMC sample from the same individual collected before the pandemic was used as a reference. The PBMCs were evaluated in the Dextramer assay to determine the frequency of SARS-CoV-2-reactive CD8+ T cells. Briefly, PBMCs were stained in a tube containing Dextramer reagents, spanning the HLA alleles A*01:01, B*07:02 and B*44:02 displaying epitopes from Spike (PE labelled) and Non-Spike (APC labelled) proteins. In a second tube cells were labelled with Dextramer reagents consisting of same HLA alleles displaying influenza, EBV or CMV epitopes (PE labelled) as well as negative control Dextramer reagents (APC labelled). Samples were analyzed using flow cytometry. Images were created with BioRender.com.

Dextramer assay reveals changes in magnitude and kinetics of SARS-COV-2 specific CD8+ T-cell immunity
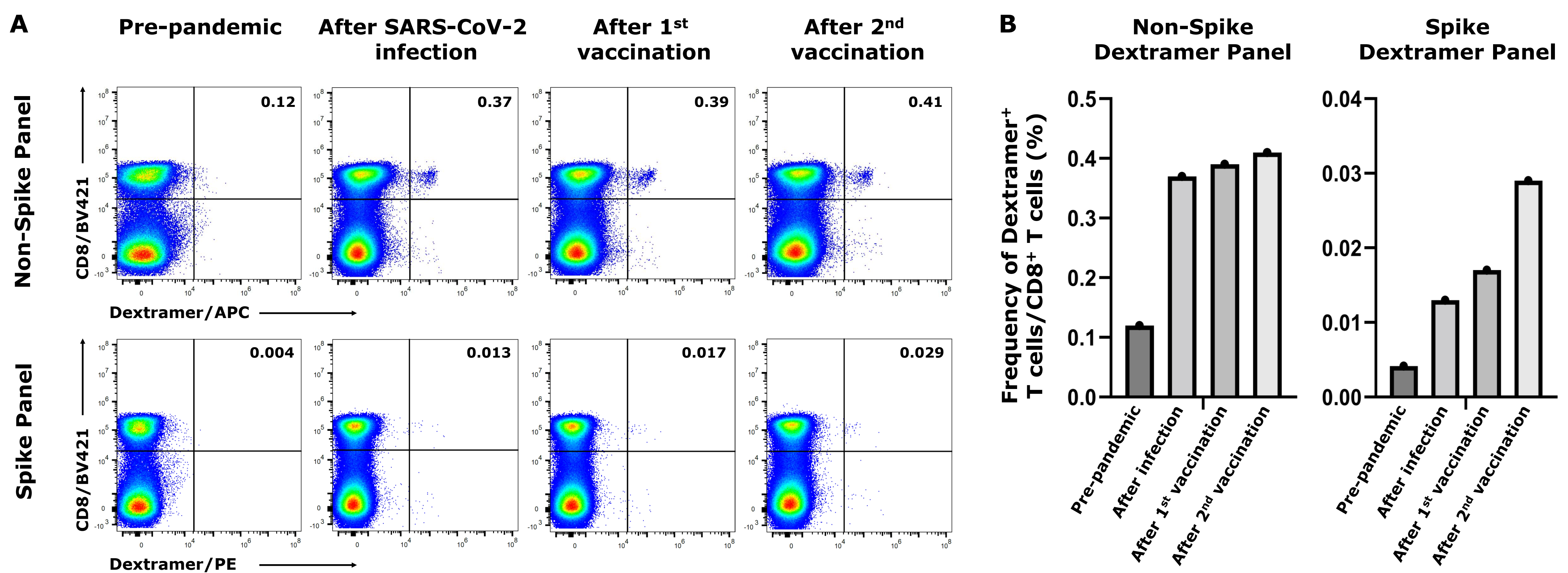

Figure 2. The Dextramer assay detects SARS-CoV-2-specific CD8+ T-cell responses displaying different magnitude and kinetics. (A) Flow cytometry plots showing SARS-CoV-2-specific CD8+ T cells reactive with Spike and Non-Spike epitopes detected in PBMCs from a vaccinated convalescent donor. The frequencies are here defined as % frequency of SARS-COV-2-specific CD8+ T cells of total count of CD8+ T cells. A CD8+ T-cell response was detected for all time points after infection and vaccination. There was no detectable response against either Spike or Non-Spike in the pre-pandemic sample. **(B)** Bar plots summarizing the change in the frequencies of Spike- and Non-Spike-specific CD8+ T cells across time points. Data showed a persistent longitudinal increase in the levels of Spike-specific CD8+ T cells from naturally acquired infection to the 1st and 2nd vaccination. The Non-Spike-specific CD8+ T-cell response appeared after infection and the response was persistent over time.

Conclusion

Here we developed a Dextramer assay to monitor the dynamics of CD8+ T-cell responses against Spike and Non-Spike proteins of SARS-CoV-2. The SARS-CoV-2-specific Dextramer assay was evaluated in a longitudinal case study to assess the time-dependent changes in the levels of SARS-CoV-2-specific T cells in a subject using pre-pandemic, post-infection, and post-vaccination samples. Spike and Non-Spike-specific CD8+ T-cell responses exhibited different patterns of magnitude and kinetics. These results demonstrate that the SARS-CoV-2 Dextramer assay can (1) Assess long-term immunity upon infection and vaccination, (2) Support decision-making about the need for booster doses if the immunity declines and (3) Guide future vaccine development. Currently, the SARS-CoV-2 Dextramer assay is being further evaluated in larger cohorts of vaccinated convalescent individuals.

Perspective: Intelligent monitoring of immunity by the SARS-CoV-2 Dextramer assay
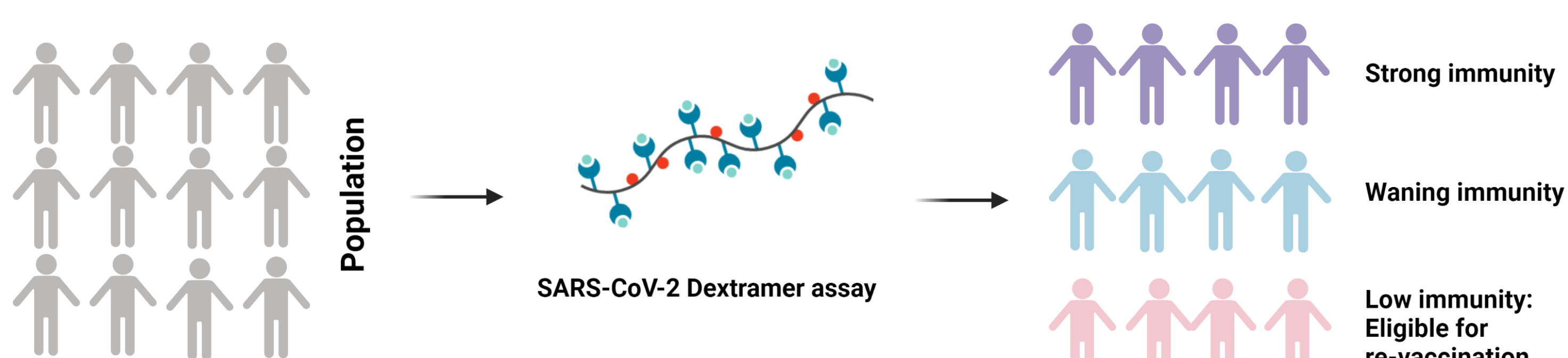

Figure 3. Proposed model for using the Dextramer assay for population stratification based on the SARS-CoV-2 immunity status. The availability of technologies like the Dextramer assay opens the possibility to use immune monitoring for population stratification and classify sub-populations eligible for re-vaccination, e.g., nursing home residents, athletes before a sporting event. Grey color symbolizes unknown status of immunity in the population. The Dextramer assay analysis of sample populations can reveal the immune status and stratify the population into sub-populations based on immunity towards SARS-CoV-2. Sub-populations showing low immunity are eligible for re-vaccination. Images created with BioRender.com.

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