The Effect of the Omicron Variant on T-Cell Immunity

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Introduction

The COVID-19 pandemic continues to affect billions of lives worldwide. The emergence of the highly transmissible SARS-CoV-2 B.1.1.529 (Omicron) variant has once again raised concerns about the effectiveness of the vaccine-induced immunity. Whereas the durability and breadth of protection against the newly circulating SARS-CoV-2 variant remain unknown, the findings highlight the need to continuously assess and quantify the level of immunity. In particular, the Omicron variant possesses numerous mutations in the Spike protein, increasing its likelihood to evade the neutralizing antibodies induced by the most common vaccines, Pfizer/BioNTech, Moderna, AstraZeneca and Johnson & Johnson, which are all based on the Spike protein.

This has more than ever re-emphasized the importance of assessing the cellular immunity against SARS-COV-2 and variants. T-cell immunity may be less affected by the new variant than the antibody response and be an essential piece in unravelling the immunity puzzle as new variants emerge.

While several studies have demonstrated robust T-cell responses against SARS-COV-2 in convalescents and vaccinated individuals, the antibody response is still the major focus area when assessing the immunity against variants. Besides limiting the assessment of the broad immunity to SARS-CoV-2 and variants, this tendency increases the risk of a deficient understanding of SARS-CoV-2-related immunity.

Immudex provides T-cell monitoring assays for SARS-CoV-2 based on the established Dextramer® technology, enabling detection and characterization of virus-specific T-cell responses. The technology relies on displaying virus-specific epitopes on major histocompatibility complex (MHC) molecules for recognition by virus-specific T cells. In this analysis we included SARS-CoV-2 CD8+ T cell assays designed to cover eight of the most common class I human leukocyte antigen (HLA) alleles including HLA-A*01:01, HLA-A*02:01, HLA-A*03:01, HLA-A*11:01, HLA-A*24:02, HLA-B*07:02, HLA-B*35:01 and HLA-B*44:02 complexed to epitopes from Spike and Non-Spike (Nucleocapsid, ORF1ab and ORF3a) proteins of the Reference strain of SARS-CoV-2. The assays cover several literature-reported epitopes tested in several cohort studies (9, 10, 15, 19) and in-house studies (not published). Among others, Nielsen et al. (2021) reported that in a study of 106 HLA-$A2^+$ individuals, 90% showed a detectable SARS-CoV-2-specific $CD8^+$ T-cell response using the Dextramer® assay. In-house data revealed a similar frequency (96%) of SARS-CoV-2 specific $CD8^+$ T-cells in mild convalescent patients across eight HLA alleles.

Similarly, the SARS-CoV-2 CD4+ T cell Dextramer® assays included in this analysis cover the alleles HLA-DRB1*01:01, HLA-DRB1*04:01 and HLA-DRB1*07:01 coupled with epitopes from Spike and Non-Spike proteins.

Here, we examined the conservation of the T-cell epitopes used in the Dextramer® assays across the SARS-CoV-2 reference strain and Delta and Omicron variants, to underline why investigating the cellular immunity may be more prominent in relation to evaluating the vaccine protection.

Analysis

To examine the T-cell epitope conservation across the SARS-CoV-2 reference strain (Lineage B, NC_0445512), Delta (B.1.617.2) and Omicron (B.1.1.529) variants, protein sequences in FASTA format were retrieved from the National Center for Biotechnology Information (NCBI) database (**Table 1**). A multiple sequence alignment analysis was created for Spike, Nucleocapsid, ORF3a and ORF1ab using CLC Sequence Viewer 8.0 (Qiagen). The multiple alignment was overlayed with the specific epitopes included in the SARS-CoV-2 Dextramer® assays to assess whether the epitopes are affected by the mutations shown in the Delta and Omicron variants **(Tables 2-3**).

***Table 1**. NCBI accession numbers for protein sequences used for multiple alignment.*

Protein	Reference Strain	Delta	Omicron
Spike	YP_009724390.1	BCX29369.1	UFP04971.1
Nucleocapsid	YP_009724397.2	BCX29377.1	UFO69287.1
ORF3a	YP_009724391.1	BCX29370.1	UFO69280.1
ORF1ab	YP_009724389.1	BCX29367.1	UFO69277.1

***Table 2.** Conservation of Spike-specific CD8⁺ T-cell epitopes across SARS-CoV-2 reference strain, Delta and Omicron variants.*

Allele	Peptide	Antigen	Reference	Delta	Omicron	Citation
A*0101	LTDEMIAQY	S				5,8,14
A*0101	WTAGAAAYY	S				19
A*0201	YLQPRTFLL	S				1, 6, 7, 11, 12, 14, 15, 16, 17, 23
A*0201	NLNESLIDL	S				15
A*0201	FIAGLIAIV	S				15, 17, 19
A*0301	KCYGVSPTK	S				6
A*0301	GVYFASTEK	S			GVYFASIEK	14
A*1101	RLFRKSNLK	S				14
A*1101	KCYGVSPTK	S				6
A*1101	GVYFASTEK	S			GVYFASIEK	14, 19
A*2402	QYIKWPWYI	S				5, 6, 11, 14, 16, 23, 25
A*2402	NYNYLYRLF	S		NYNYRYRLF		14, 16, 23, 25
B*0702	SPRRARSVA	S		SRRRARSVA	SHRRARSVA	7, 8
B*0702	APHGVVFL	S				14
B*3501	QPTESIVRF	S				16
B*3501	LPFNDGVYF	S				16
B*3501	IPFAMQMAY	S				16

Green = no mutations in epitope, Red = mutations in epitope

Table 3. *Conservation of Non-Spike-specific CD8⁺ T-cell epitopes across Wuhan, Delta and Omicron variants.*

Allele	Peptide	Antigen	Reference	Delta	Omicron	Citation
A*0201	LLLDRLNQL	N				6, 7, 8, 14, 15, 19
A*0301	KTFPPTEPK	N				2, 6, 11, 14
A*1101	ATEGALNTPK	N				5, 6, 18
A*1101	KTFPPTEPK	N				2, 6, 14
B*0702	KPRQKRTAT	N				7, 8, 18
B*0702	SPRWYFYYL	N				2, 6, 7, 8, 13, 14
A*0101	FTSDYYQLY	ORF3a				2, 6, 8, 13, 14, 18
A*0201	LLYDANYFL	ORF3a				6, 7, 8, 14
A*2402	VYFLQSINF	ORF3a				5, 6, 14, 23, 31
A*0101	CTDDNALAYY	ORF1ab				6, 8, 11
A*0101	TTDPSFLGRY	ORF1ab		TTDLSFLGRY		5, 6, 11, 13
A*0201	ALWEIQQVV	ORF1ab				6, 14, 23
A*0301	KTIQPRVEK	ORF1ab				6, 14
A*0301	VVYRGTTTYK	ORF1ab				8, 14
A*1101	ASMPTTIAK	ORF1ab				5, 8, 23
A*2402	VYIGDPAQL	ORF1ab				5, 6, 14, 31, 32
B*0702	IPRRNVATL	ORF1ab				6, 8, 14

Green = no mutations in epitope, Red = mutations in epitope

Discussion

The multiple alignment analysis of Spike across the SARS-COV-2 variants Delta and Omicron showed a conservation of 15 and 14 out of 17 investigated CD8⁺ T-cell epitopes, respectively, corresponding to 88% and 82%. All but one of the selected Non-Spike CD8+ and CD4+ T-cell epitopes were conserved across the reference strain and both variants.

Analysis of SARS-CoV-2 specific CD4+ T-cell epitopes also showed a high degree of conservation between the Wuhan, Delta and Omicron variants (data not shown). The Delta sequence showed a conservation of 15 out of 17 Spike epitopes (88%) and 12 out of 12 Non-Spike epitopes (100%) derived from Nucleocapsid, Envelope, and ORF1ab. The Omicron sequence showed a conservation of 13 out of 17 Spike epitopes (76%) and a conservation of 12 out of 12 Non-Spike proteins (100%).

The high degree of conservation across SARS-CoV-2 T-cell epitopes in the three major variants, Wuhan, Delta and Omicron is promising for the disease course of individuals that have previously encountered SARS-COV-2 and/or been vaccinated. Based on the observations, we hypothesize that the level of T-cell responses will yield good protection against Omicron in convalescent and likely also vaccinated individuals. Moreover, the present data verify the applicability of our current SARS-CoV-2 Dextramer® assay to monitor the T-cell immunity across variants.

In future, virus strains with mutations affecting key T-cell epitopes may appear. However, our highly adaptable technology allows a rapid development of SARS CoV-2 Dextramer® assays tailored to any new variant.

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