

Supplementary Figure 1. Gating strategy and T cell and cell lines used in this study.

(a-c) Flow cytometry gating strategy of tetramer staining in PBMCs from donor GV16 (a), CD25⁺CD137⁺ activated T cell lines from donor GV60 (b) and tetramer staining of T cell lines from GV60 (c). (d) NF9 and QI9 stimulated-T cell lines used in this study.
(e) IFN-γ production in peptide titration of the NF9 and QI9 peptide in T cell lines from GV34 and GV60 donors. The assay was performed in triplicate, and the means are shown with the SD. (f) GFP expression of A549-ACE2-A2402-IRES-GFP cells. A549-ACE2 parental (shaded histogram) and FACS-sorted A549-ACE2-A2402-IRES-GFP cells (open histogram) are shown. (g) Western blot. Representative blots of cells expressing prototype, Omicron BA.1, and Delta spike (S) protein of three independent experiments. Source data are provided as a Source Data file.



Supplementary Figure 2. Cell lines used in TCR-sensitivity assay.

(a) TCR sensitivity of GV34 #5-3 toward cells expressing spike protein and peptide-pulsed target cells.
(b) Representative western blots of cells expressing various spike (S) proteins of three independent experiments.
Source data are provided as a Source Data file. (c) The level of peptide/HLA complexes was evaluated following treatment with a proteasome inhibitor, MG-132 (10 μM). The effect of the inhibitor was evaluated as reporter activity and shown as a fold change in the level of peptide/HLA on target cells expressing Omicron BA.1 spike protein with the S446G reversion (Omicron BA.1/S446G). Statistical analysis versus DMSO alone was determined by an unpaired two-tailed Student' s t-test. Data are expressed as a median. ns, no statistical significance. a, c The assay was performed in triplicate, and the means are shown with the SD. Data are representative of three independent experiments.

Supplementary Table 1. Human PBMCs used in this study, related to Fig. 1

| Donor ID | Sex | Age | HLA-A24? | vaccinated? | Days after two doses of vaccination |
|----------|--------|-----|----------|-------------|-------------------------------------|
| Vku19 | Female | 36 | Positive | BNT162b2 | 21 |
| GV1 | Female | 28 | Positive | BNT162b2 | 9 |
| GV4 | Female | 18 | Negative | BNT162b2 | 20 |
| GV5 | Female | 18 | Positive | BNT162b2 | 20 |
| GV6 | Female | 79 | Positive | BNT162b2 | 21 |
| GV9 | Female | 24 | Positive | BNT162b2 | 21 |
| GV10 | Male | 23 | Positive | BNT162b2 | 22 |
| GV11 | Male | 23 | Positive | BNT162b2 | 22 |
| GV12 | Female | 28 | Negative | BNT162b2 | 23 |
| GV13 | Male | 22 | Positive | BNT162b2 | 21 |
| GV14 | Male | 23 | Positive | BNT162b2 | 22 |
| GV15 | Female | 23 | Positive | BNT162b2 | 21 |
| GV16 | Male | 22 | Positive | BNT162b2 | 22 |
| GV17 | Male | 24 | Negative | BNT162b2 | 21 |
| GV19 | Male | 24 | Positive | BNT162b2 | 22 |
| GV20 | Female | 24 | Positive | BNT162b2 | 22 |
| GV21 | Male | 22 | Positive | BNT162b2 | 21 |
| GV22 | Male | 22 | Positive | BNT162b2 | 22 |
| GV23 | Male | 25 | Positive | BNT162b2 | 22 |
| GV24 | Male | 23 | Positive | BNT162b2 | 22 |
| GV25 | Male | 24 | Negative | BNT162b2 | 22 |
| GV26 | Male | 23 | Positive | BNT162b2 | 22 |
| GV27 | Female | 23 | Negative | BNT162b2 | 21 |
| GV28 | Female | 23 | Positive | BNT162b2 | 21 |
| GV29 | Male | 23 | Positive | BNT162b2 | 21 |
| GV31 | Male | 23 | Positive | BNT162b2 | 22 |
| GV32 | Male | 56 | Positive | BNT162b2 | 27 |
| GV33 | Male | 39 | Positive | BNT162b2 | 24 |
| GV34 | Female | 38 | Positive | BNT162b2 | 24 |
| GV35 | Male | 52 | Positive | BNT162b2 | 24 |
| GV36 | Male | 39 | Positive | BNT162b2 | 24 |
| GV51 | Male | 34 | Positive | BNT162b2 | 30 |
| GV59 | Male | 37 | Positive | BNT162b2 | 126 |
| GV60 | Male | 51 | Positive | mRNA-1273 | 116 |
| GV61 | Female | 36 | Positive | mRNA-1273 | 107 |

| Specificity | ID | TRAV | TRAJ | CDR3a | TRBV | TRBJ | TRBD | CDR3β |
|-------------|-------------|-------------|-----------|-----------------|-------------|------------|----------|--------------------|
| | GV34 #2-2 | TRAV12-1*01 | TRAJ33*01 | CVVNALMDSNYQLIW | TRBV5-1*01 | TRBJ2-7*01 | TRBD1*01 | CASSLGQGYEQYF |
| NF9/A24 | GV34 #5-3 | TRAV12-1*01 | TRAJ33*01 | CVVNLFDSNYQLIW | TRBV2*01 | TRBJ2-7*01 | TRBD1*01 | CASSEGAGYEQYF |
| | VKU19 #12-3 | TRAV12-3*01 | TRAJ44*01 | CAFTGTASKLTF | TRBV7-8*01 | TRBJ2-1*01 | TRBD2*02 | CASSPELNEQFF |
| | | | | | | | | |
| | GV33 #57 | TRAV3*01 | TRAJ8*01 | CAGVLFNTGFQKLVF | TRBV20-1*02 | TRBJ2-1*01 | TRBD2*01 | CSASDRGASGSFSNEQFF |
| QI9/A24 | GV34 #43 | TRAV21*01 | TRAJ43*01 | CAAPRYNNNDMRF | TRBV2*01 | TRBJ2-2*01 | TRBD1*01 | CASSEGADAGELFF |
| | GV36 #10-2 | TRAV19*01 | TRAJ9*01 | CALSEPPSGGFKTIF | TRBV20-1*01 | TRBJ1-1*01 | TRBD1*01 | CSARGQGLNTEAFF |

Supplementary Table 2. TCR sequences isolated from NF9/A24 and QI9/A24 specific T cells by single-cell analysis, related to Fig. 3

| Product | | Primer name | Sequence (5'-to-3') |
|--------------------|-------------|-----------------|--|
| Prototype spike | N440K | N440K Fwd | CAGCAACAAGCTGGACAGCAAGGTGG |
| | | N440K Rev | CTGTCCAGCTTGTTGCTGTTCCAGGC |
| | N440K G446S | N440K G446S Fwd | GCAACAAGCTGGACAGCAAGGTGTCCGGCAACTACAAC |
| | | N440K G446S Rev | GTTGCCGGACACCTTGCTGTCCAGCTTGTTGCTGTTC |
| | G446S | G446S Fwd | CAAGGTGTCCGGCAACTACAACTACCTC |
| | | G446S Rev | GTAGTTGCCGGACACCTTGCTGTCCAG |
| Omicron BA.1 spike | S446G | S446G Fwd | GACAGCAAGGTGGGAGGCAACTACA |
| | | S446G Rev | GCCTCCCACCTTGCTGTCCAGCTTG |

Supplementary Table 3. Primers for the construction of spike derivatives, related to Fig. 3