**Supplemental Table 1: Demographic characteristics of participants**

|  | All Participants | COVID-19 + | COVID-19 - |
|---|---|---|---|
|  | n=110 | n=24 | n=86 |
| **Demographics** |  |  |  |
| **Mean Age (years) (±SD)** | 6.84 (±4.4) | 6.22 (±3.2) | 7.01 (±4.7) |
| **Sex** |  |  |  |
| Male | 61 (55.5%) | 13 (54.2%) | 48 (55.8%) |
| Female | 49 (44.5%) | 11 (45.8%) | 38 (44.2%) |
| **Race/Ethnicity** |  |  |  |
| Black | 27 (24.5%) | 3 (12.5%) | 24 (27.9%) |
| Hispanic | 42 (38.2%) | 18 (75.0%) | 24 (27.9%) |
| White | 26 (23.6%) | 0 (0.0%) | 26 (30.2%) |
| Other/Mixed | 15 (13.6%) | 3 (12.5%) | 12 (14.0%) |
| **Clinical Characteristics** |  |  |  |
| **Primary Diagnosis** |  |  |  |
| Sleep disordered breathing | 60 (54.5%) | 14 (58.3%) | 46 (53.5%) |
| Mild OSA, AHI >1 and < 5 | 14 (12.7%) | 6 (25.0%) | 8 (9.3%) |
| Moderate OSA, AHI 5 to 10 | 6 (5.5%) | 1 (4.2%) | 5 (5.8%) |
| Severe OSA, AHI >10 | 14 (12.7%) | 2 (8.3%) | 12 (14.0%) |
| Eustachian tube dysfunction | 1 (0.9%) | 1 (4.2%) | 0 (0.0%) |
| PFAPA | 7 (6.4%) | 0 (0.0%) | 7 (8.1%) |
| Recurrent tonsillitis | 3 (2.7%) | 0 (0.0%) | 3 (3.5%) |
| Chronic tonsillitis/tonsil stones | 5 (4.5%) | 0 (0.0%) | 5 (5.8%) |
| **Medications (within 2 weeks prior to surgery)** |  |  |  |
| Inhaled or nasal corticosteroid | 14 (12.7%) | 4 (16.7%) | 10 (11.6%) |
| Oral corticosteroid | 7 (6.4%) | 0 (0.0%) | 7 (8.1%) |
| Loratadine (Claritin) | 10 (9.1%) | 1 (4.2%) | 9 (10.5%) |
| Montelukast (Singulair) | 6 (5.5%) | 0 (0.0%) | 6 (7.0%) |
| Cetirizine (Zyrtec) | 5 (4.5%) | 0 (0.0%) | 5 (5.8%) |
| **Prior COVID-19 infection by serology/flow cytometry** |  |  |  |
| Negative | 86 (78.2%) | 0 (0.0%) | 86 (100%) |
| Positive | 24 (21.8%) | 24 (100%) | 0 (0.0%) |

OSA = Obstructive sleep apnea

AHI = Apnea hypopnea index obtained by polysomnography

PFAPA = periodic fever, aphthous stomatitis, pharyngitis, adenitis syndrome

**Supplementary Table 3: Characteristics of Participants with Prior COVID-19**

| Patient ID | Age (years) | Sex | Reason for tonsillectomy/ adenoidectomy | Medication | Co-morbid conditions | Prior positive SARS-CoV-2 PCR/Ag test | Days from positive PCR/Ag to surgery | Symptomatic at time of positive PCR/Ag test? | Symptoms at time of positive PCR/Ag test |
|---|---|---|---|---|---|---|---|---|---|
| CNMC 001 | 2.6 | F | SDB | | | No | | | |
| CNMC 005 | 7.5 | F | SDB | | Asthma | No | | | |
| CNMC 008 | 3.4 | M | Mild OSA | | | No | | | |
| CNMC 011 | 8 | M | Mild OSA | | Asthma | Yes | 142 | Symptomatic | Headache, sore throat, diarrhea, myalgia |
| CNMC 016 | 8.9 | F | SDB | | | No | | | |
| CNMC 022 | 4.1 | M | Mild OSA | | | Yes | 108 | Symptomatic | Cough, myalgia |
| CNMC 029 | 3.1 | M | SDB | | | No | | | |
| CNMC 032 | 3.6 | F | Severe OSA | | | Yes | 26 | Asymptomatic | |
| CNMC 041 | 6 | F | Severe OSA | | | No | | | |
| CNMC 046 | 7.2 | M | SDB | | | No | | | |
| CNMC 050 | 2.9 | F | SDB | Inh Steroid | | Yes | 31 | Asymptomatic | |
| CNMC 069 | 3.4 | F | SDB | Inh Steroid | | Yes | 201 | Symptomatic | Shortness of breath, myalgia, dizziness, GI symptoms |
| CNMC 070 | 16.4 | M | Mild OSA | | | Yes | 98 | Symptomatic | Fever, cough, anosmia, myalgia, GI symptoms, chest pain |
| CNMC 071 | 6.2 | F | SDB | | | Yes | 71 | Symptomatic | Fever, cough, shortness of breath, anosmia, myalgia, GI symptoms |
| CNMC 087 | 6 | M | SDB | Loratadine | | No | | | |
| CNMC 089 | 12.1 | F | SDB | | | No | | | |
| CNMC 091 | 9.2 | M | Moderate OSA | | | No | | | |
| CNMC 100 | 4.7 | F | Mild OSA | Inh Steroid | | Yes | 25 | Asymptomatic | |
| CNMC 101 | 4.6 | M | SDB | | | No | | | |
| CNMC 102 | 5.1 | M | SDB | | | Yes | 35 | Symptomatic | Fever, myalgia |
| CNMC 103 | 8.1 | M | Mild OSA | | | Yes | 303 | Asymptomatic | |
| CNMC 104 | 6.8 | M | SDB | Inh Steroid | | No | | | |
| CNMC 108 | 4.2 | F | ETD | | | Yes | 84 | Symptomatic | Cough, diarrhea, nasal congestion |
| CNMC 109 | 5.1 | M | SDB | | | No | | | |

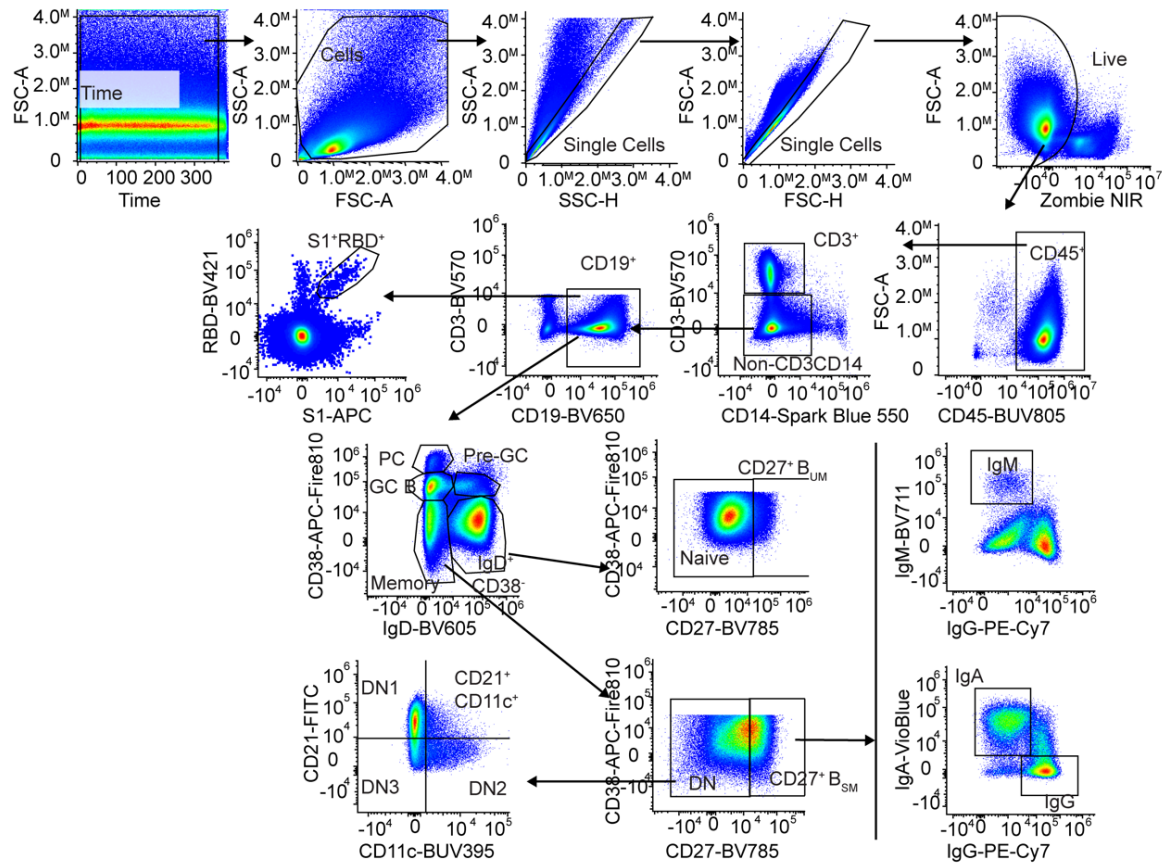SDB: sleep disordered breathing
OSA: obstructive sleep apnea
ETD: eustachian tube dysfunction
Ag: antigen    Inh Steroid: inhaled steroid      GI: gastrointestinal
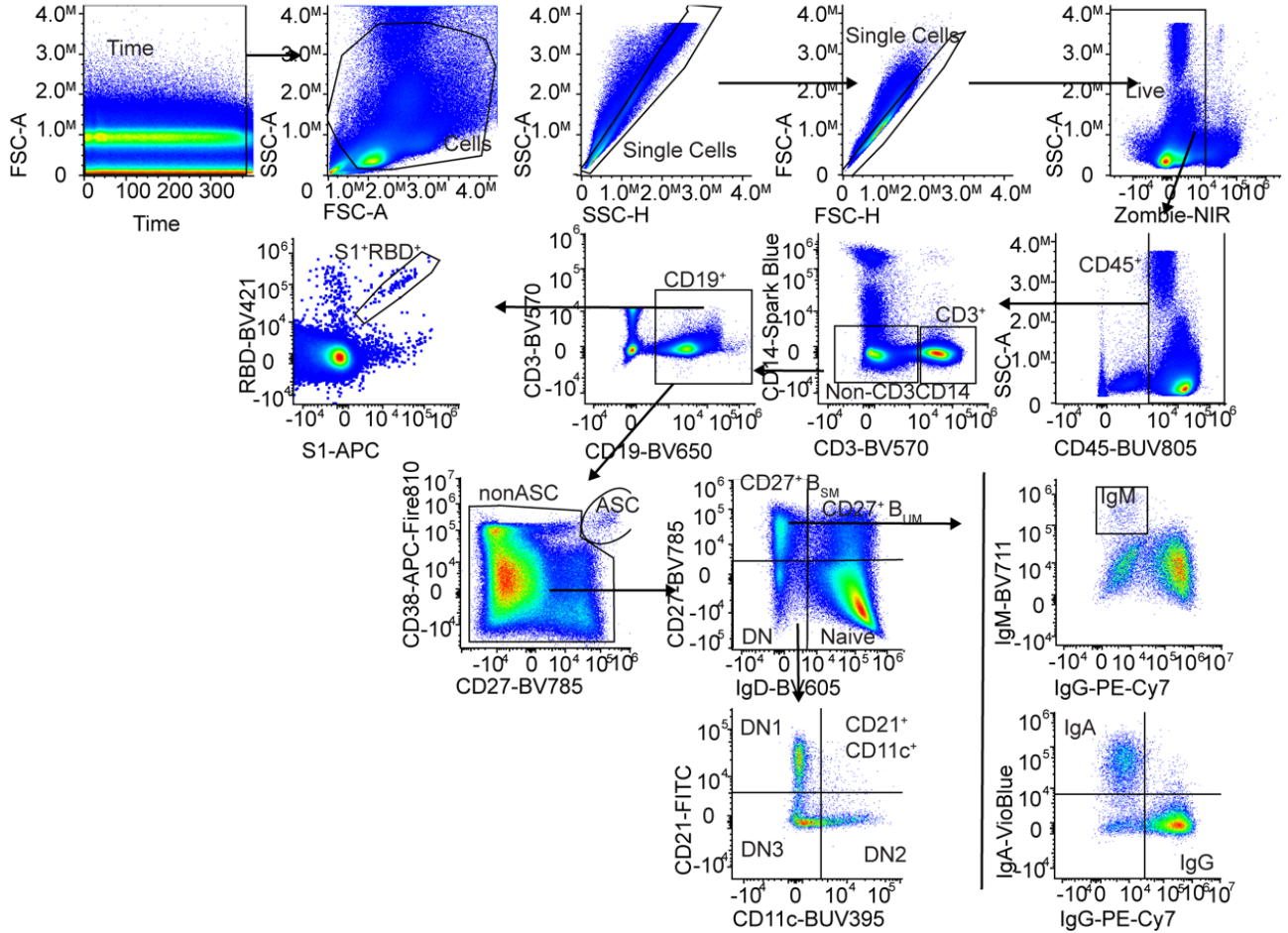F: female      M: male

**Supplementary Figure 1**



**Supplementary Figure 1. Gating strategy of major CD19⁺ B cell populations and S1⁺RBD⁺ B cells in pharyngeal tissues**

Representative flow cytometry plots of major B cell populations and S1⁺RBD⁺ B cell gating strategy in tonsils and adenoids, including PC (plasma cells), GC (germinal center) B cells, pre-GC (pre-germinal center B cells), naïve (naïve B cells), DN (double negative B cells), CD27⁺ B$_{SM}$ (CD27⁺ switched memory B cells), CD27⁺ B$_{UM}$ (CD27⁺ unswitched memory cells). Gating for DN subsets is also shown.
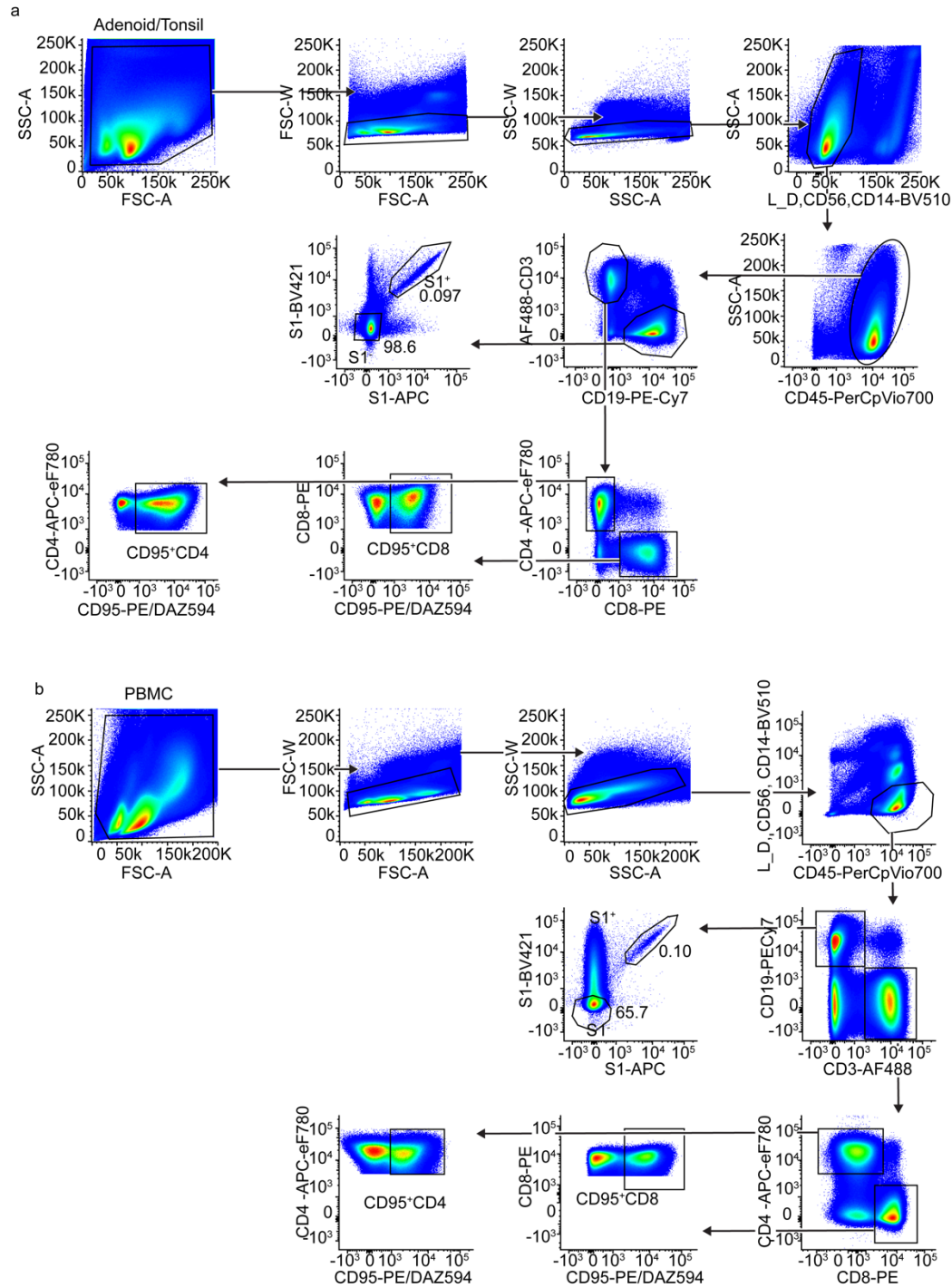
**Supplementary Figure 2**



**Supplementary Figure 2. Gating strategy of major CD19$^+$ B cell populations and S1$^+$RBD$^+$ B cells in peripheral blood**

Representative flow cytometry plots of major B cell populations and S1$^+$RBD$^+$ B cell gating strategy in PBMCs, including ASC (antibody secreting cells, equivalent to plasma cells and plasmablasts), naïve (naïve B cells), DN (double negative B cells), CD27$^+$ B$_{SM}$ (CD27$^+$ switched memory B cells), CD27$^+$ B$_{UM}$ (CD27$^+$ unswitched memory B cells). Gating for DN subsets is also shown.

**Supplementary Figure 3**



**Supplementary Figure 3. Gating strategy for sorting B and T cells used in CITE-seq**

Flow cytometry plots showing sorting strategy for or S1 binding (S1$^+$) B cells (S1-BV421 and S1-APC double positive), S1$^-$ B cells, CD95$^+$ CD4$^+$ T cells, and CD95$^+$ CD8$^+$ T cells in (a) tonsils and adenoids cells and (b) PBMCs. Sorted cells were analyzed with CITE-seq.
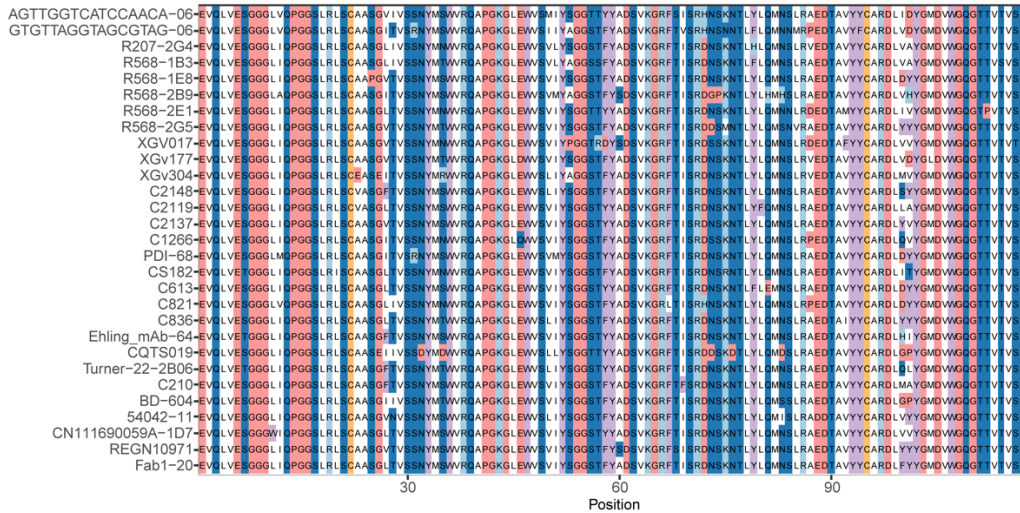
## Supplementary Figure 4
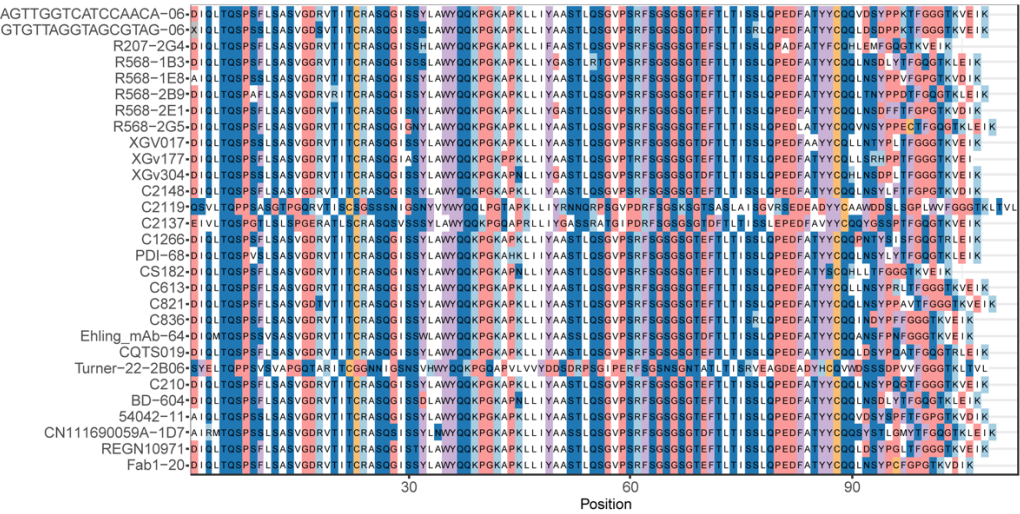


**Supplementary Figure 4. S1⁺ B cell clone closely matching publically reported SARS-CoV-2-specific BCR**

One S1⁺ B cell clone (**a**) found in both the tonsil and adenoid of CNMC 71 (post-COV participant) was highly similar to 37 BCR heavy chain sequences in the CoV-AbDab database of SARS-CoV-2 specific antibody sequences. Similarity of both the heavy chain (**b**) and light chain (**c**) sequences of this clone (noted in first two lines of each panel) to convergent sequences in CoV-AbDab are shown. Only sequence matches from CoV-AbDab with full length sequences available are shown.

**Supplementary Figure 5**
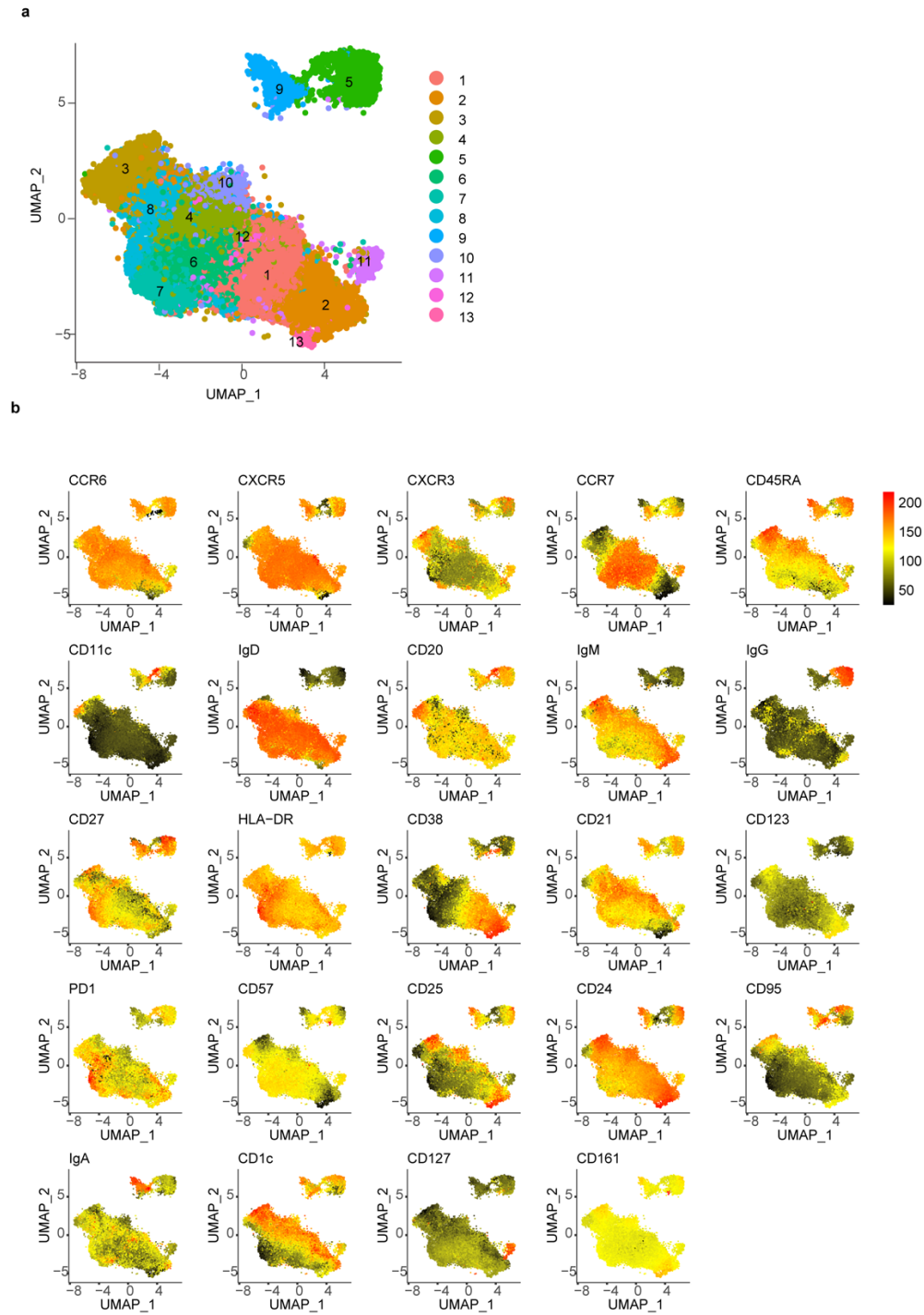


**Supplementary Figure 5. UMAP of unsupervised clustering of B cells from PBMC**

a.  UMAP of unsupervised clustering of surface markers from flow cytometric analysis of CD19+ B cells from PBMC.

b.  Heatmaps of marker/antibody expression overlayed on UMAP.

**Supplementary Figure 6**

-
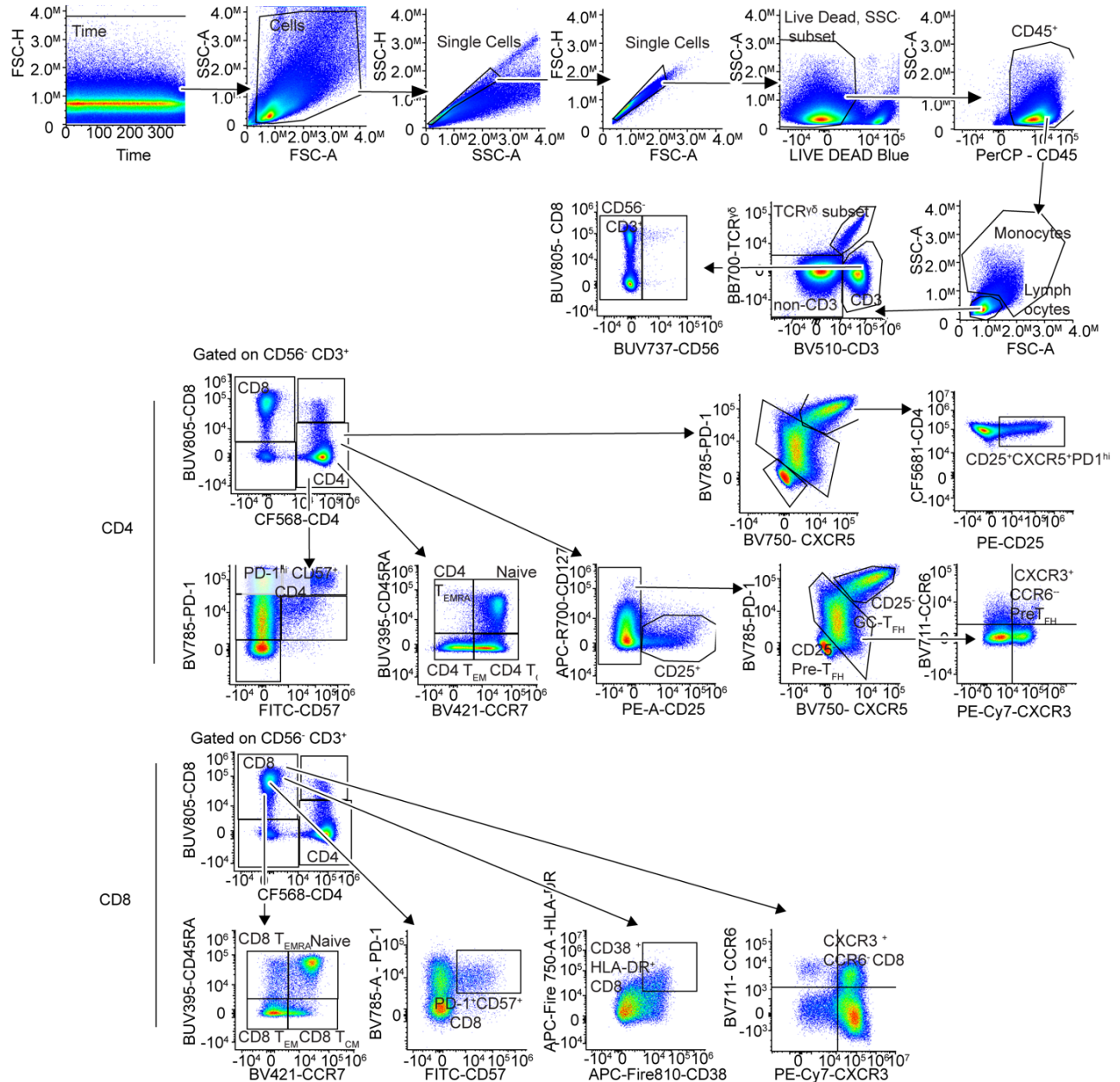


**Supplementary Figure 6. Gating strategy of major CD4⁺ and CD8⁺ T cell populations in tonsils and adenoids**

Representative flow cytometry plots are shown. $T_{EM}$ are effector memory T cells, $T_{CM}$ are central memory T cells, $T_{EMRA}$ are terminally differentiated effector memory T cells, and $T_{FH}$ are T follicular helper cells.

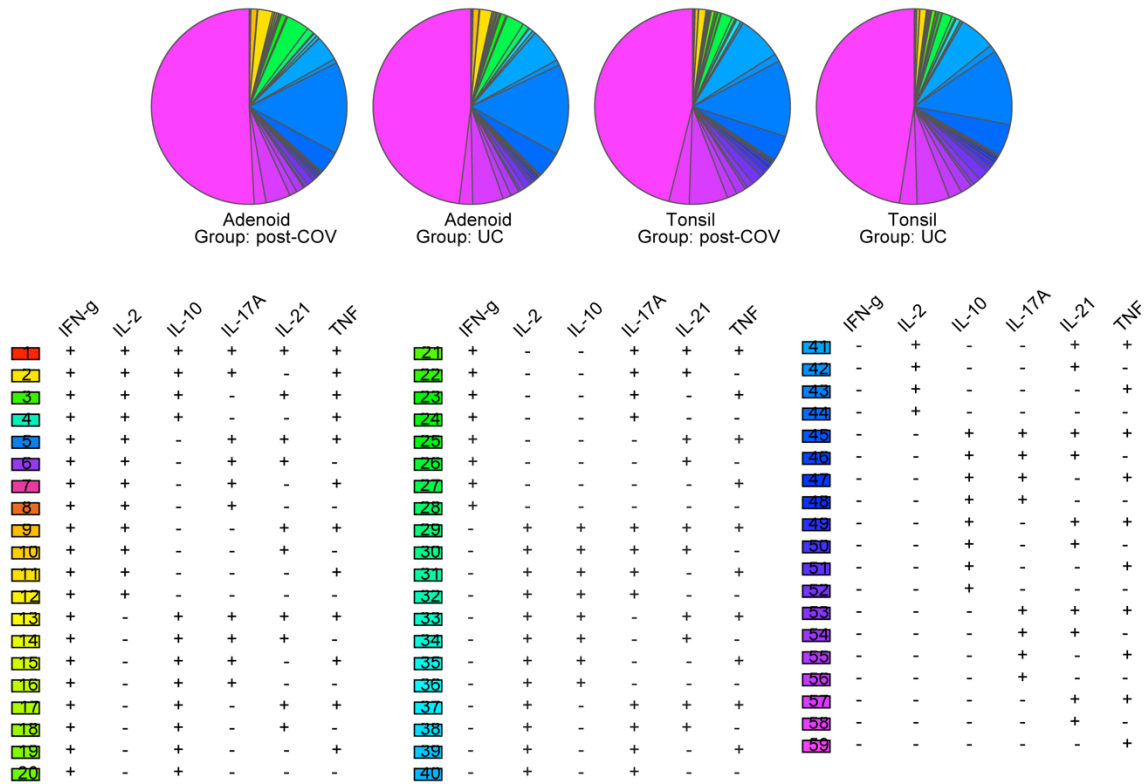| | IFN-g | IL-2 | IL-10 | IL-17A | IL-21 | TNF |
|---|---|---|---|---|---|---|
| 1 | + | + | + | + | + | + |
| 2 | + | + | + | + | - | + |
| 3 | + | + | + | - | + | + |
| 4 | + | + | + | - | - | + |
| 5 | + | + | - | + | + | + |
| 6 | + | + | - | + | + | - |
| 7 | + | + | - | + | - | + |
| 8 | + | + | - | + | - | - |
| 9 | + | + | - | - | + | + |
| 10 | + | + | - | - | + | - |
| 11 | + | + | - | - | - | + |
| 12 | + | + | - | - | - | - |
| 13 | + | - | + | + | + | + |
| 14 | + | - | + | + | + | - |
| 15 | + | - | + | + | - | + |
| 16 | + | - | + | + | - | - |
| 17 | + | - | + | - | + | + |
| 18 | + | - | + | - | + | - |
| 19 | + | - | + | - | - | + |
| 20 | + | - | + | - | - | - |

| | IFN-g | IL-2 | IL-10 | IL-17A | IL-21 | TNF |
|---|---|---|---|---|---|---|
| 21 | + | - | - | + | + | + |
| 22 | + | - | - | + | + | - |
| 23 | + | - | - | + | - | + |
| 24 | + | - | - | + | - | - |
| 25 | + | - | - | - | + | + |
| 26 | + | - | - | - | + | - |
| 27 | + | - | - | - | - | + |
| 28 | + | - | - | - | - | - |
| 29 | - | + | + | + | + | + |
| 30 | - | + | + | + | + | - |
| 31 | - | + | + | + | - | + |
| 32 | - | + | + | + | - | - |
| 33 | - | + | + | - | + | + |
| 34 | - | + | + | - | + | - |
| 35 | - | + | + | - | - | + |
| 36 | - | + | + | - | - | - |
| 37 | - | + | - | + | + | + |
| 38 | - | + | - | + | + | - |
| 39 | - | + | - | + | - | + |
| 40 | - | + | - | + | - | - |

| | IFN-g | IL-2 | IL-10 | IL-17A | IL-21 | TNF |
|---|---|---|---|---|---|---|
| 41 | - | + | - | - | + | + |
| 42 | - | + | - | - | + | - |
| 43 | - | + | - | - | - | + |
| 44 | - | + | - | - | - | - |
| 45 | - | - | + | + | + | + |
| 46 | - | - | + | + | + | - |
| 47 | - | - | + | + | - | + |
| 48 | - | - | + | + | - | - |
| 49 | - | - | + | - | + | + |
| 50 | - | - | + | - | + | - |
| 51 | - | - | + | - | - | + |
| 52 | - | - | + | - | - | - |
| 53 | - | - | - | + | + | + |
| 54 | - | - | - | + | + | - |
| 55 | - | - | - | + | - | + |
| 56 | - | - | - | + | - | - |
| 57 | - | - | - | - | + | + |
| 58 | - | - | - | - | + | - |
| 59 | - | - | - | - | - | + |

**Supplementary Figure 7. SPICE analysis of CD4+ T cells from tonsils and adenoids**

Pie charts show the proportion of responding CD4+ T cells from adenoids and tonsils producing 59 different combinations of 6 cytokines (IFN-$\gamma$, IL-2, IL-10, IL-17A, IL-21 and TNF) after PMA and ionomycin stimulation from post-COV and UC donors (adenoid post-COV n=13, UC n=13; tonsil post-COV n=13, UC n=13). Frequencies were determined by Boolean combination gates in FlowJo and analyzed with SPICE. Combinations with frequencies below 0.01 from each donor were excluded from the analysis (categories IFN$\gamma$+IL2+IL10+IL17A+IL21+TNF-, IFN$\gamma$+IL2+IL10+IL17A+IL21-TNF-, IFN$\gamma$+IL2+IL10+IL17A-IL21+TNF- and IFN$\gamma$+IL2+IL10+IL17A-IL21-TNF- were excluded). Significance calculated using two-sided Mann-Whitney U test. Combinations with significant differences (p<0.05) in post-COV and UC are highlighted in red.
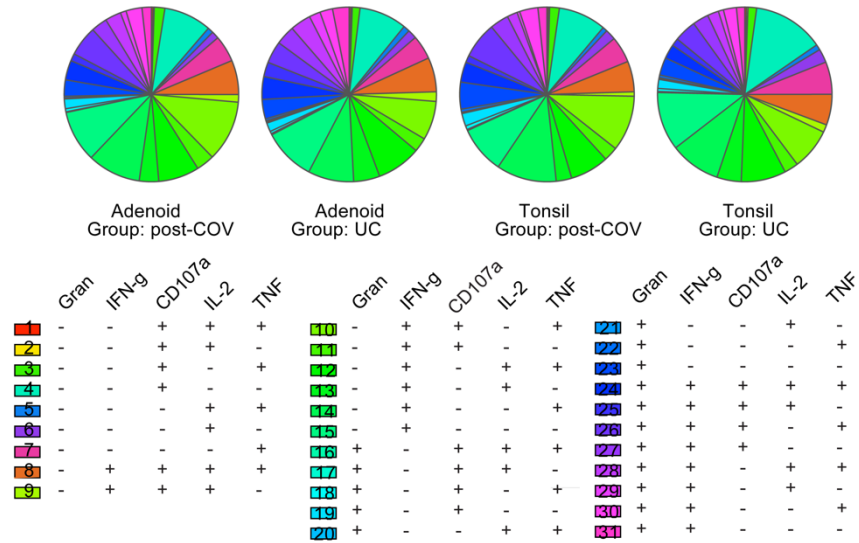
**Supplementary Figure 8**
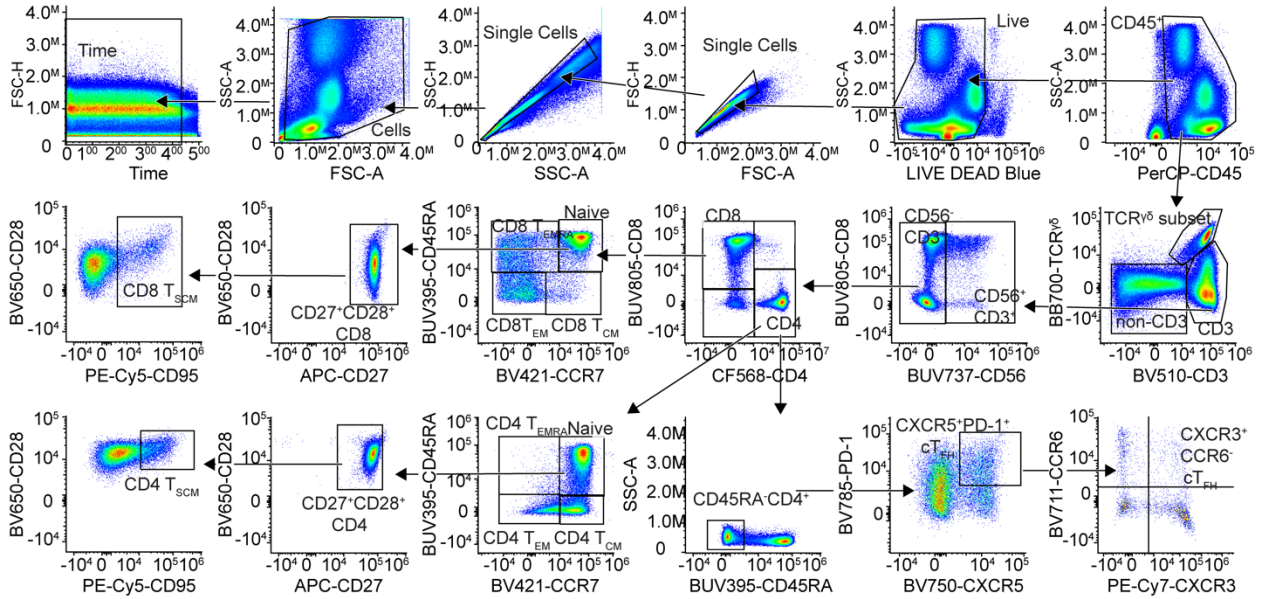


**Supplementary Figure 8. Gating strategy of major CD4$^+$ and CD8$^+$ T cell populations in PBMCs**

Representative flow cytometry plots are shown. $T_{SCM}$ are T stem cell-like memory cells, $T_{EM}$ are effector memory T cells, $T_{CM}$ are central memory T cells, $T_{EMRA}$ are terminally differentiated effector memory T cells, and $cT_{FH}$ are circulating T follicular helper cells.
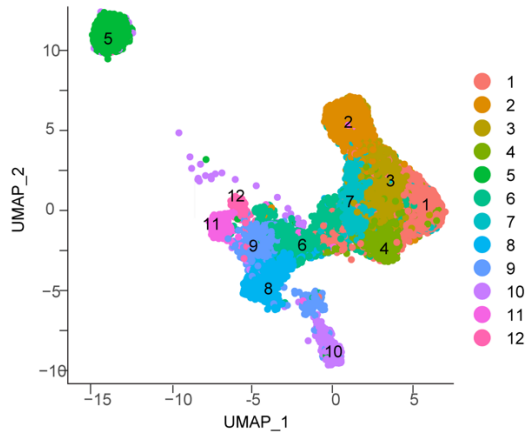
**Supplementary Figure 9. SPICE analysis of CD8$^+$ T cells from tonsil and adenoid**

| | Gran | IFN-g | CD107a | IL-2 | TNF |
|---|---|---|---|---|---|
| 1 | - | - | + | + | + |
| 2 | - | - | + | + | - |
| 3 | - | - | + | - | + |
| 4 | - | - | + | - | - |
| 5 | - | - | - | + | + |
| 6 | - | - | - | + | - |
| 7 | - | - | - | - | + |
| 8 | - | + | + | + | + |
| 9 | - | + | + | + | - |

| | Gran | IFN-g | CD107a | IL-2 | TNF |
|---|---|---|---|---|---|
| 10 | - | + | + | - | + |
| 11 | - | + | + | - | - |
| 12 | - | + | - | + | + |
| 13 | - | + | - | + | - |
| 14 | - | + | - | - | + |
| 15 | - | + | - | - | - |
| 16 | + | - | + | + | + |
| 17 | + | - | + | + | - |
| 18 | + | - | + | - | + |
| 19 | + | - | + | - | - |
| 20 | + | - | - | + | + |

| | Gran | IFN-g | CD107a | IL-2 | TNF |
|---|---|---|---|---|---|
| 21 | + | - | - | + | - |
| 22 | + | - | - | - | + |
| 23 | + | - | - | - | - |
| 24 | + | + | + | + | + |
| 25 | + | + | + | + | - |
| 26 | + | + | + | - | + |
| 27 | + | + | + | - | - |
| 28 | + | + | - | + | + |
| 29 | + | + | - | + | - |
| 30 | + | + | - | - | + |
| 31 | + | + | - | - | - |

Pie charts show the proportion of responding CD8$^+$ T cells from adenoids and tonsils producing 31 different combinations of 6 cytokines (granzyme B, IFN-γ, CD107a, IL-2 and TNF) after PMA and ionomycin stimulation from post-COV and UC donors (adenoid post-COV n = 13, UC n = 13; tonsil post-COV n = 13, UC n = 13). Frequencies were determined by Boolean combination gates in FlowJo and analyzed with SPICE software. Combinations with frequencies < 0.01 from each donor were excluded from the analysis. Combinations with significant differences in post-COV and UC are highlighted in red. Significance calculated using two-sided Mann-Whitney U test. Combinations with significant differences (p<0.05) in post-COV and UC are highlighted in red.

**Supplementary Figure 10**
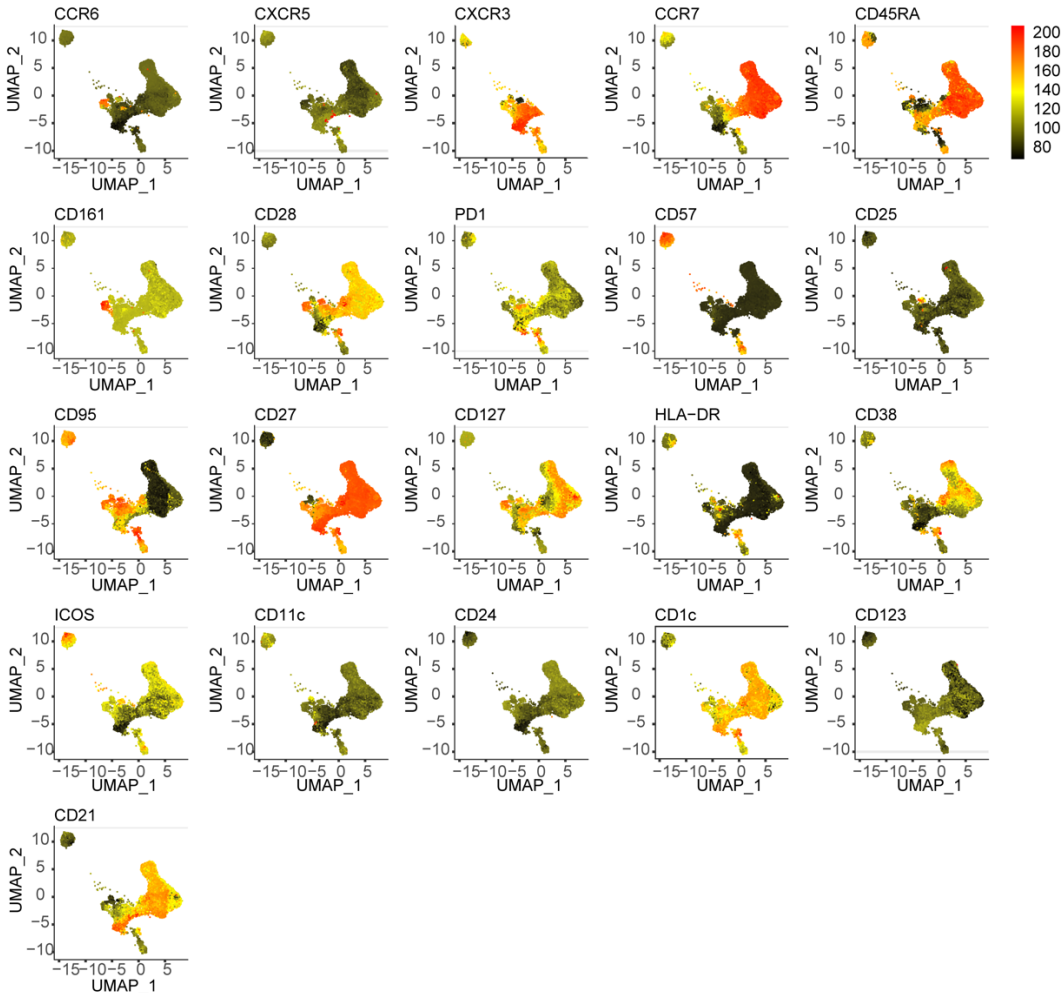


**Supplementary Figure 10. UMAP of unsupervised clustering of CD8+ T cells from PBMCs**

a. UMAP of unsupervised clustering of surface markers from flow cytometric analysis of CD8+ T cells from PBMCs.

b. Heatmaps of marker/antibody expression overlayed on UMAP.