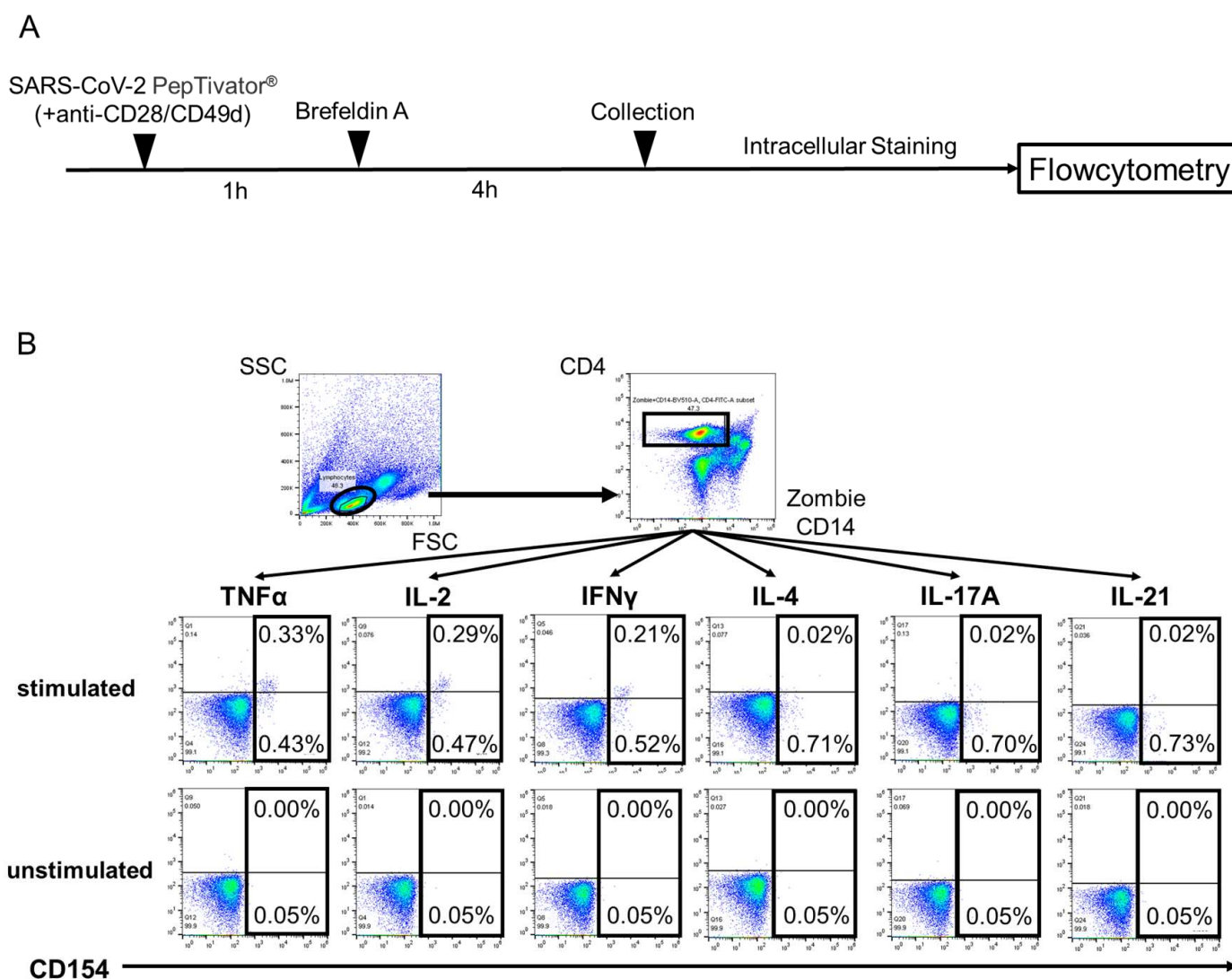


| Surface antigen | | | | Subset |
|------------------|------|----------------|-------------------------|-----------------------------------|
| Zombie- CD14- | CD4+ | CD45RA- | CXCR5- CXCR3 CCR6 | Th1-type Th2-type Th17-type |
| | | | CXCR5+ | Tfh-type |
| | | CD25+ CD127low | | Treg-type |

Supplementary figure 1

Gating strategy for flow cytometric analysis of CD4+ T cell subsets.

Zombie and CD14 were used for excluding dead cells and monocyte, respectively, in the lymphocyte gate. Representative dot plots are shown in the panels. T cell subsets defined by the pattern of surface staining were listed in the table.



Supplementary figure 2

Protocol for flow cytometric analysis of vaccine antigen-specific CD4⁺ T cells.

A. Preserved PBMC from RA patients was stimulated with a mixture of peptides from SARS-CoV-2 in the presence of anti-CD28 and CD49d mAbs, followed by an addition of brefeldin A. After stimulation, cells were firstly stained for surface antigens, and after fixation/permeabilization, they were stained for intracellular antigens.

B. Gating strategy for detecting antigen-specific (CD154⁺) CD4⁺ T cells are depicted. Upper and lower panels showed samples with and without stimulation with SARS-CoV-2 peptides. Representative dot plots are shown.