**Supplemental information**

Differential effects of the second SARS-CoV-2

mRNA vaccine dose on T cell immunity

in naive and COVID-19 recovered individuals

Daniel Lozano-Ojalvo, Carmen Camara, Eduardo Lopez-Granados, Pilar Nozal, Lucía del Pino-Molina, Luz Yadira Bravo-Gallego, Estela Paz-Artal, Marjorie Pion, Rafael Correa-Rocha, Alberto Ortiz, Marcos Lopez-Hoyos, Marta Erro Iribarren, Jose Portoles, Maria Pilar Rojo-Portoles, Gloria Ojeda, Isabel Cervera, Maria Gonzalez-Perez, Irene Bodega-Mayor, Maria Montes-Casado, Pilar Portoles, Mayte Perez-Olmeda, Jesus Oteo, Rodrigo Sanchez-Tarjuelo, Venu Pothula, Megan Schwarz, Manisha Brahmachary, Anthony Tanoto Tan, Nina Le Bert, Cecilia Berin, Antonio Bertoletti, Ernesto Guccione, and Jordi Ochando

Figure S1
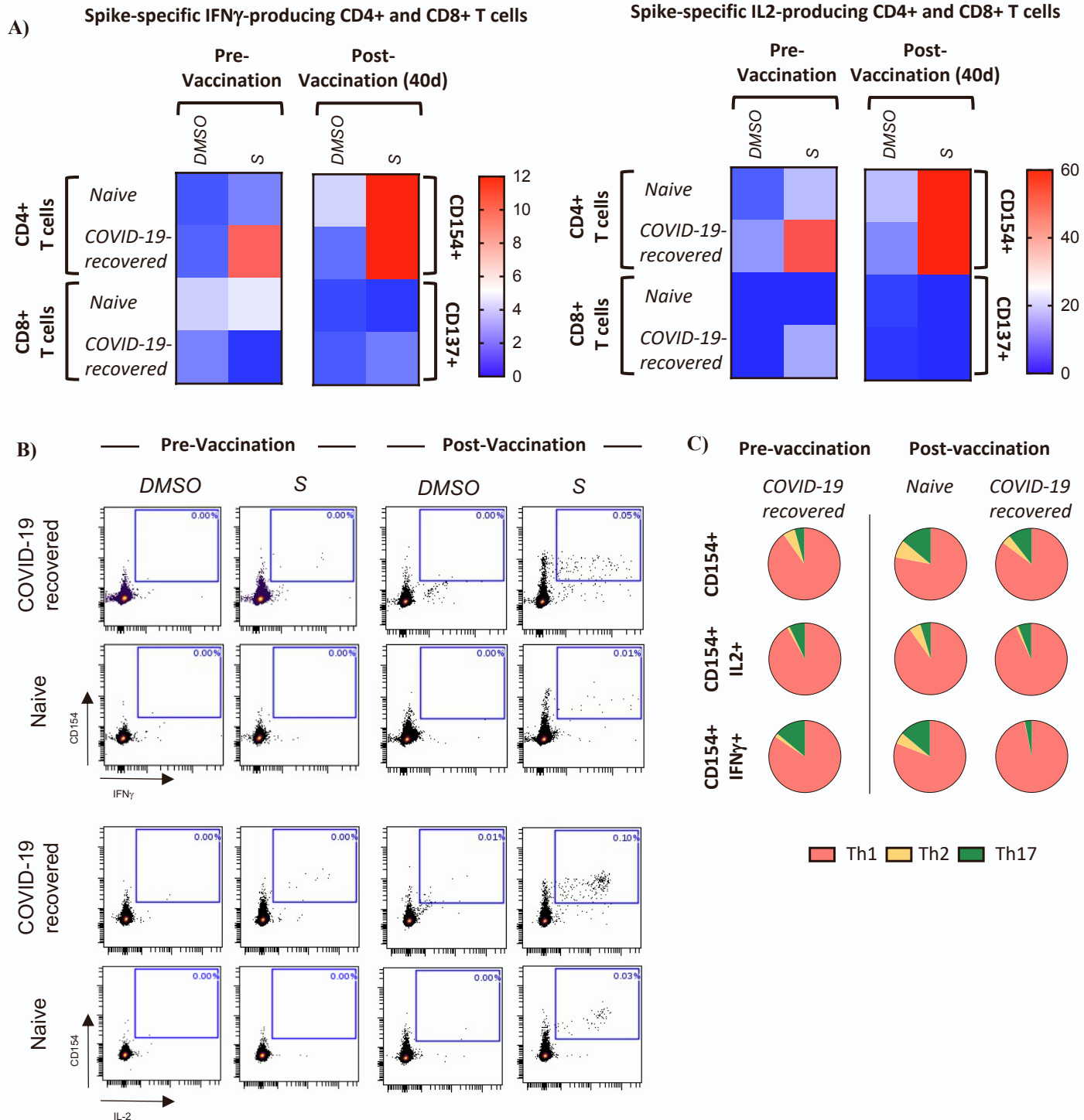
Figure S1. Flow cytometric characterization of spike-specific CD4+ T cells. Related to Figure 1.
Flow cytometry determination and identity of SARS-CoV-2 spike-specific T cells in peripheral blood mononuclear cells obtained (PBMCs) from naïve and COVID-19 recovered individuals before and 20 days after the second dose of the BNT162b2 vaccine (40d) (n=5 individuals per group and time-point). PBMCs were unstimulated (DMSO) or stimulated with SARS-CoV-2 spike (S) peptide pool. **a)** Heatmaps show the percentage of expression of the activation markers CD154 and CD137 in IFN-γ and IL-2 producing CD4+ and CD8+ T cells, respectively. **b)** Representative flow cytometric plots of IFN-γ and IL-2 expression by antigen-specific (CD154^{+}) memory (CD45RA^{-}CCR7^{-}) CD4^{+} T cells. **c)** Frequency of the CD4 T cell subsets Th1 (CCR6^{-}CCR4^{-}CXCR3^{+}), Th2 (CCR6^{-}CCR4^{+}CXCR3^{-}) and Th17 (CCR6^{+}CCR4^{-}CXCR3^{+}) in total CD154^{+} memory CD4^{+} T cells as well as IFN-γ and IL-2 producing spike-specific (CD154^{+}) memory CD4^{+} T cells (n=5 individuals per group and time-point).
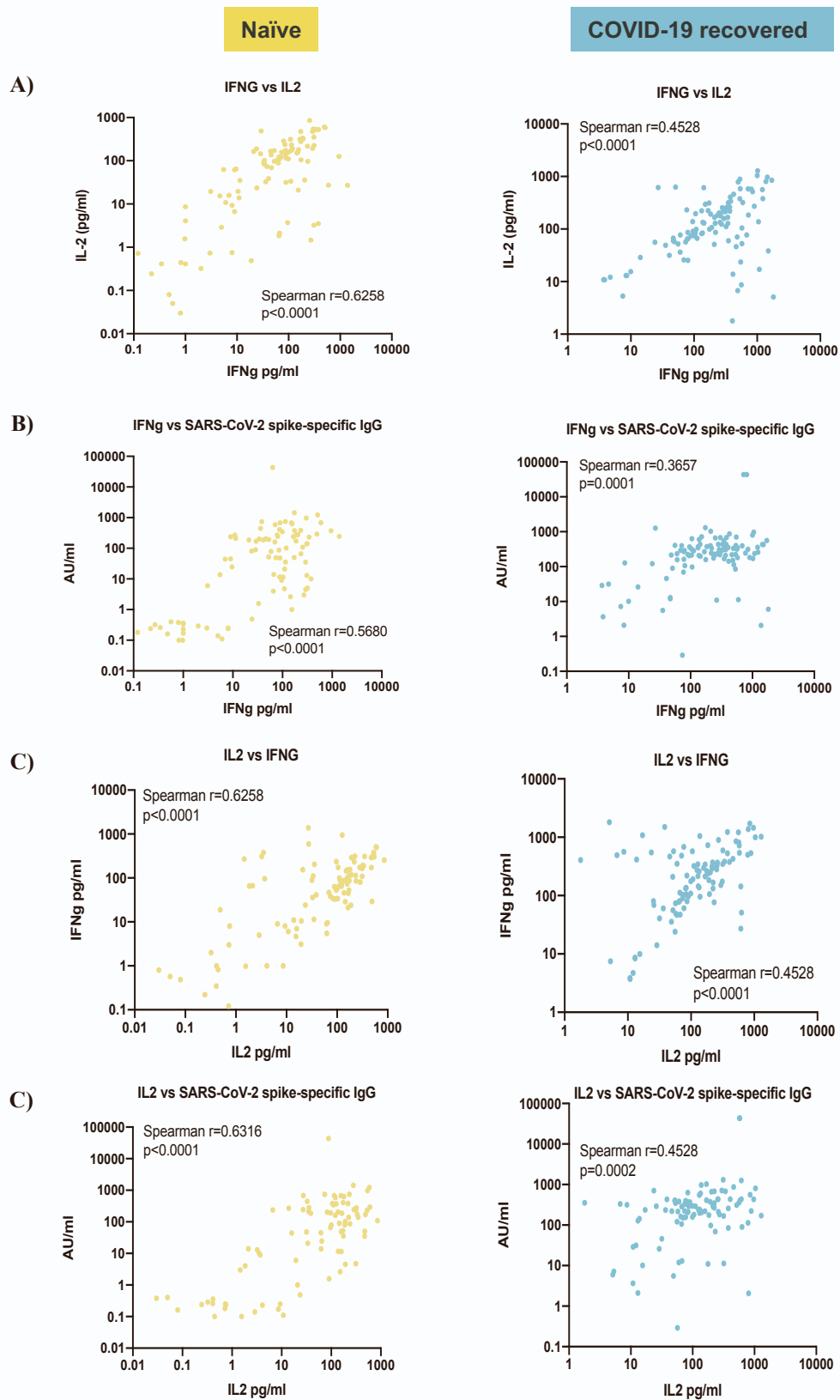
Figure S2

Figure S2. Humoral and Cellular immunity correlation in naïve and COVID-19 recovered. Related to Figure 1.
a) Correlation between IFNg and IL-2 levels in naïve (top) and COVID-19 recovered (bottom). **b)** Correlation between IFNg and SARS-CoV-2 spike-specific IgG levels in naïve (top) and COVID-19 recovered (bottom). **c)** Correlation between IL-2 and SARS-CoV-2 spike-specific IgG levels in naïve (top) and COVID-19 recovered (bottom). Correlations were estimated by nonparametric Spearman r correlation coefficient and significance was calculated by two tailed p value (indicated in each plot).