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Supplemental information

Intradermal-delivered DNA vaccine induces durable immunity mediating a reduction in viral load in a rhesus macaque SARS-CoV-2 challenge model

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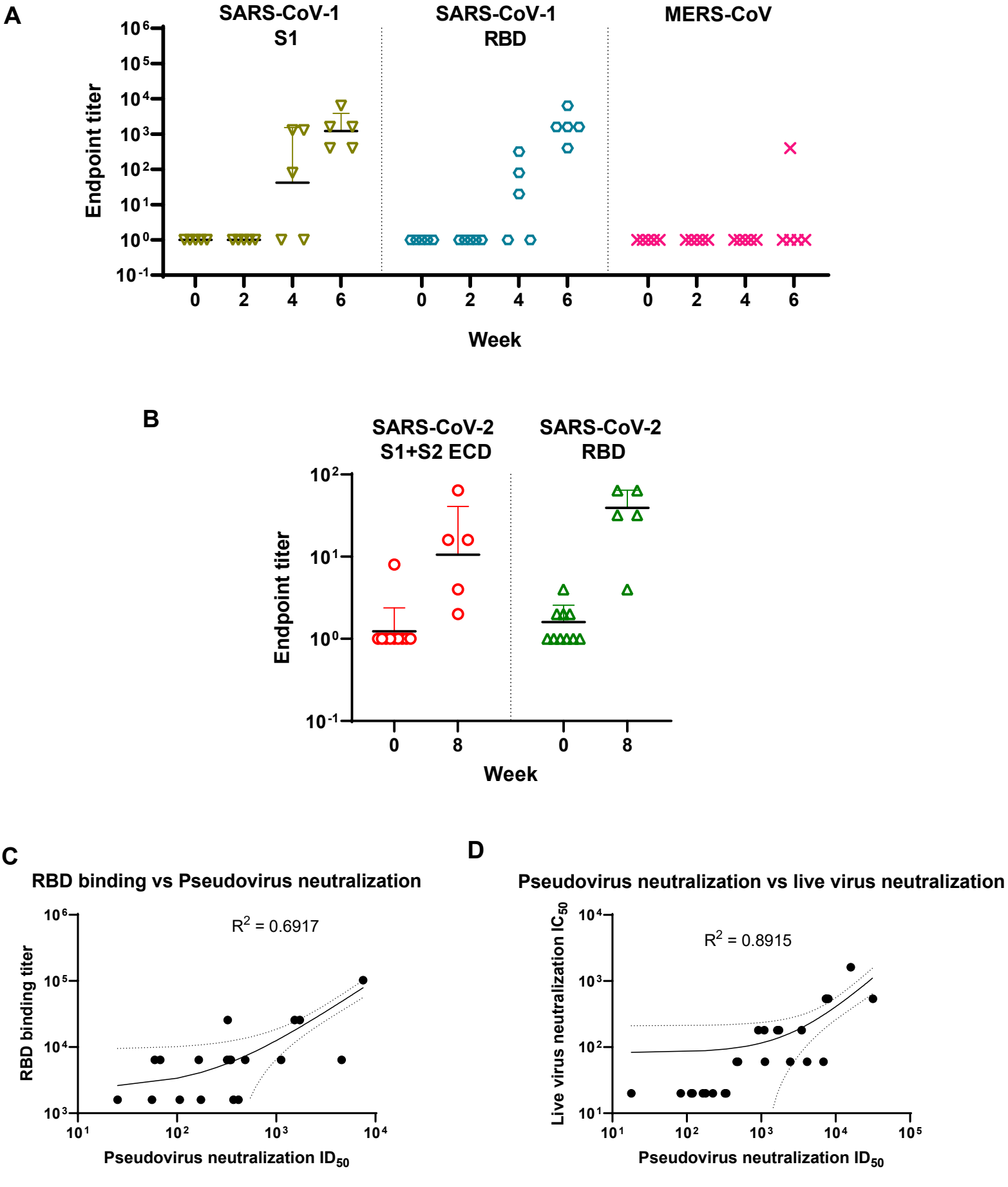

Figure S1. Immune Responses in INO-4800 vaccinated rhesus macaques, Related to Figure 1 **A)** Serum IgG cross-reactivity to SARS-CoV and MERS-CoV spike protein. IgG binding was measured in sera from INO-4800 vaccinated rhesus macaques to SARS-CoV S1 and MERS-CoV S1 protein antigen. **B)** Bronchoalveolar lavage (BAL) IgG reactive to SARS-CoV-2 S protein antigens. BAL samples collected from vaccinated animals were assessed for SARS-CoV-2 reactive IgG binding to the full-length SARS-CoV-2 spike protein and the RBD domain. **C and D)** Simple linear regression analysis comparing total IgG antibodies against (**C**) the RBD and (**D**) neutralizing antibodies raised in DNA-vaccinated macaques

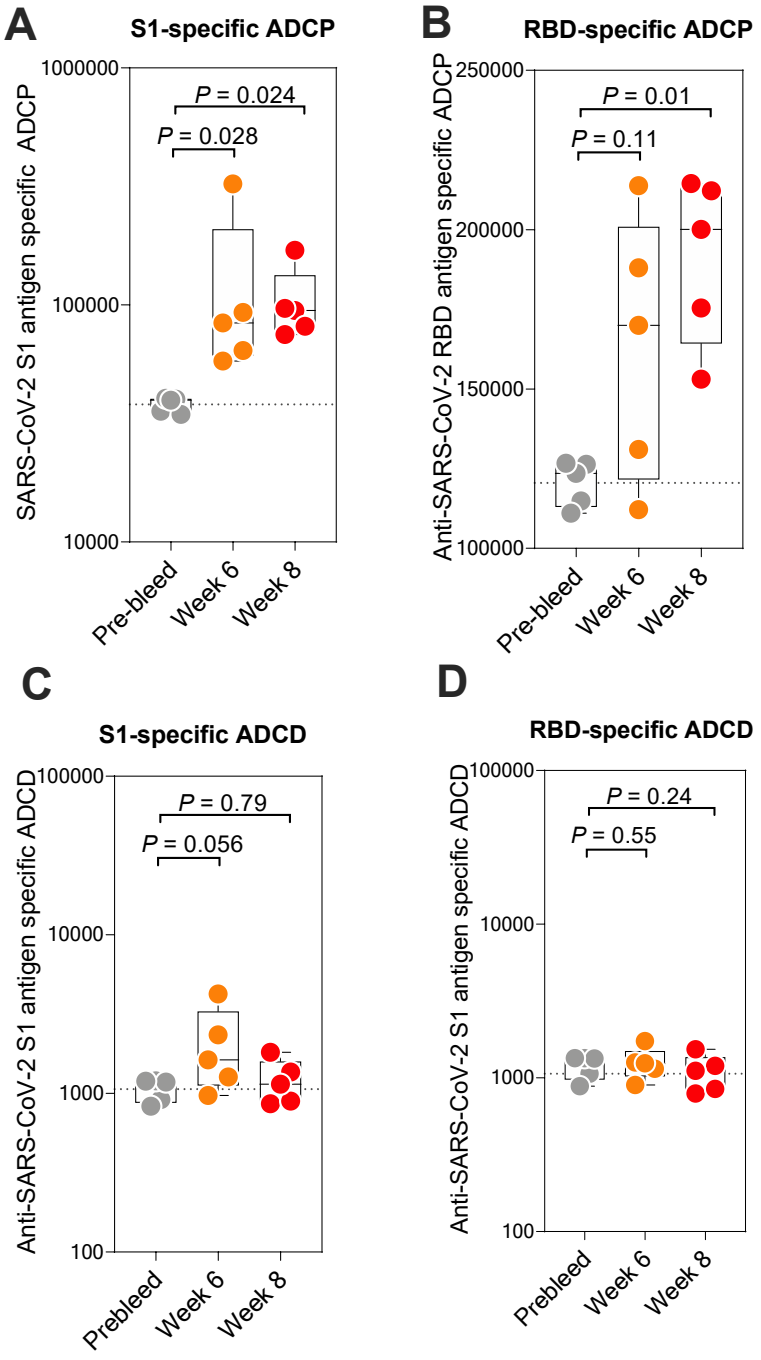

Figure S2. Antibody-dependent cellular phagocytosis (ADCP) and antibody-dependent complement deposition (ADCD) activity of NHP sera at peak immune response time points post-vaccination, Related to Figure 1. To measure ADCP activity, sera from immunized macaques were combined with SARS-CoV-2 A) S1 or B) RBD-conjugated fluorescent beads, followed by overnight incubation with THP-1 cells. The phagoscore was determined by calculating the percentage of fluorescent / bead+ cells and the median fluorescence intensity of the THP-1 cells. To measure ADCD activity, ACE2-CHO cells were pulsed with biotinylated SARS-CoV-2 C) S1 and D) RBD proteins and subsequently combined with NHP sera. Freshly diluted guinea pig complement was added to the cells and following incubation, complement deposition was detected with a goat anti-guinea pig C3-FITC conjugated antibody and analyzed by flow cytometry. ADCD is reported as MFI of FITC+ cells.

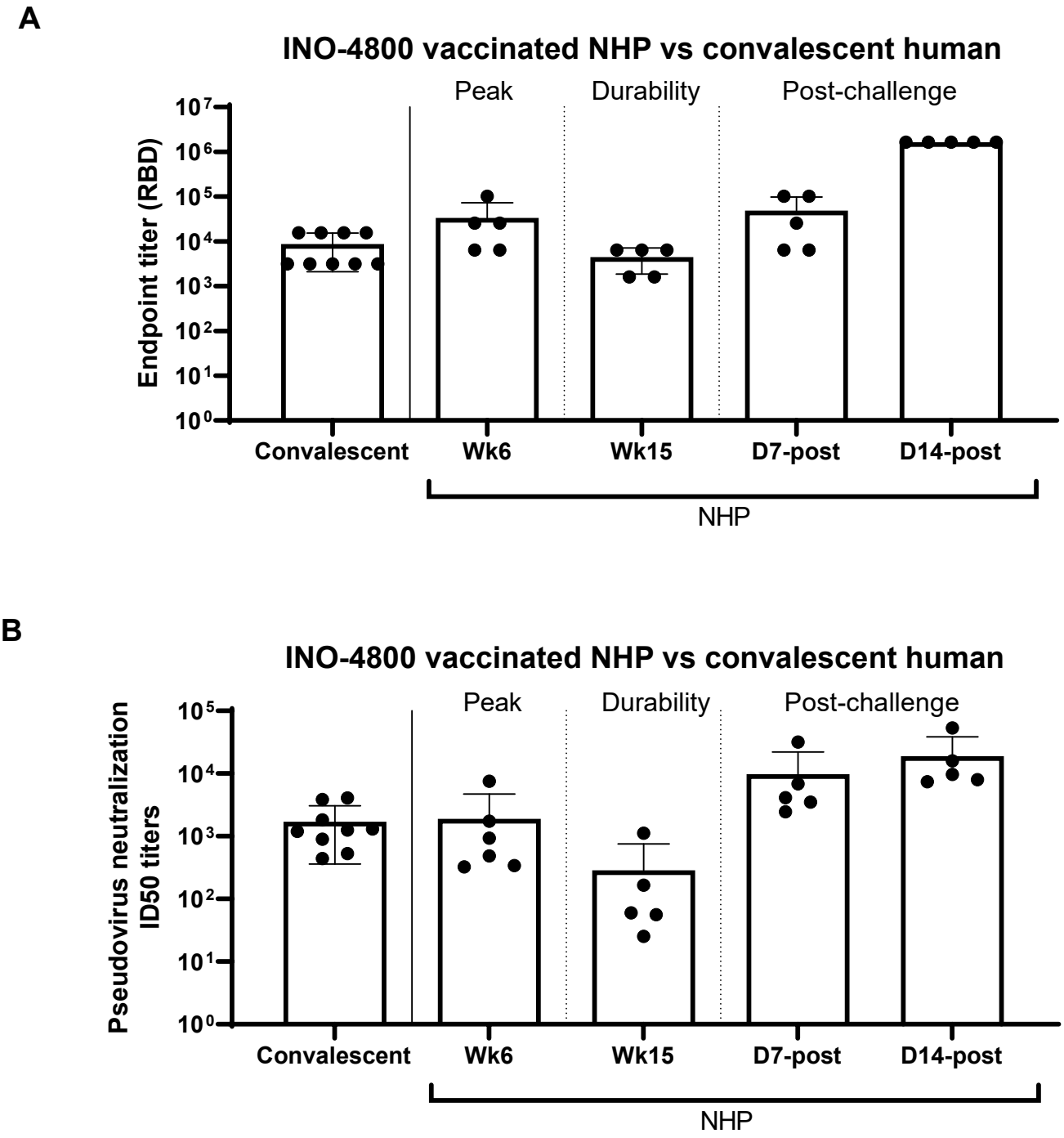

Figure S3. Comparison of NHP endpoint binding antibody and pseudoneutralization titers with SARS-CoV-2 convalescent human donors, Related to Figures 1 and 3. (**A**) ELISA endpoint titer comparison (RBD) and (**B**) Pseudoneutralization assay (D614) comparing INO-4800 vaccinated NHP peak, durability, and post-challenge pseudoneutralization ID50 titers with SARS-CoV-2 convalescent human sera titers.

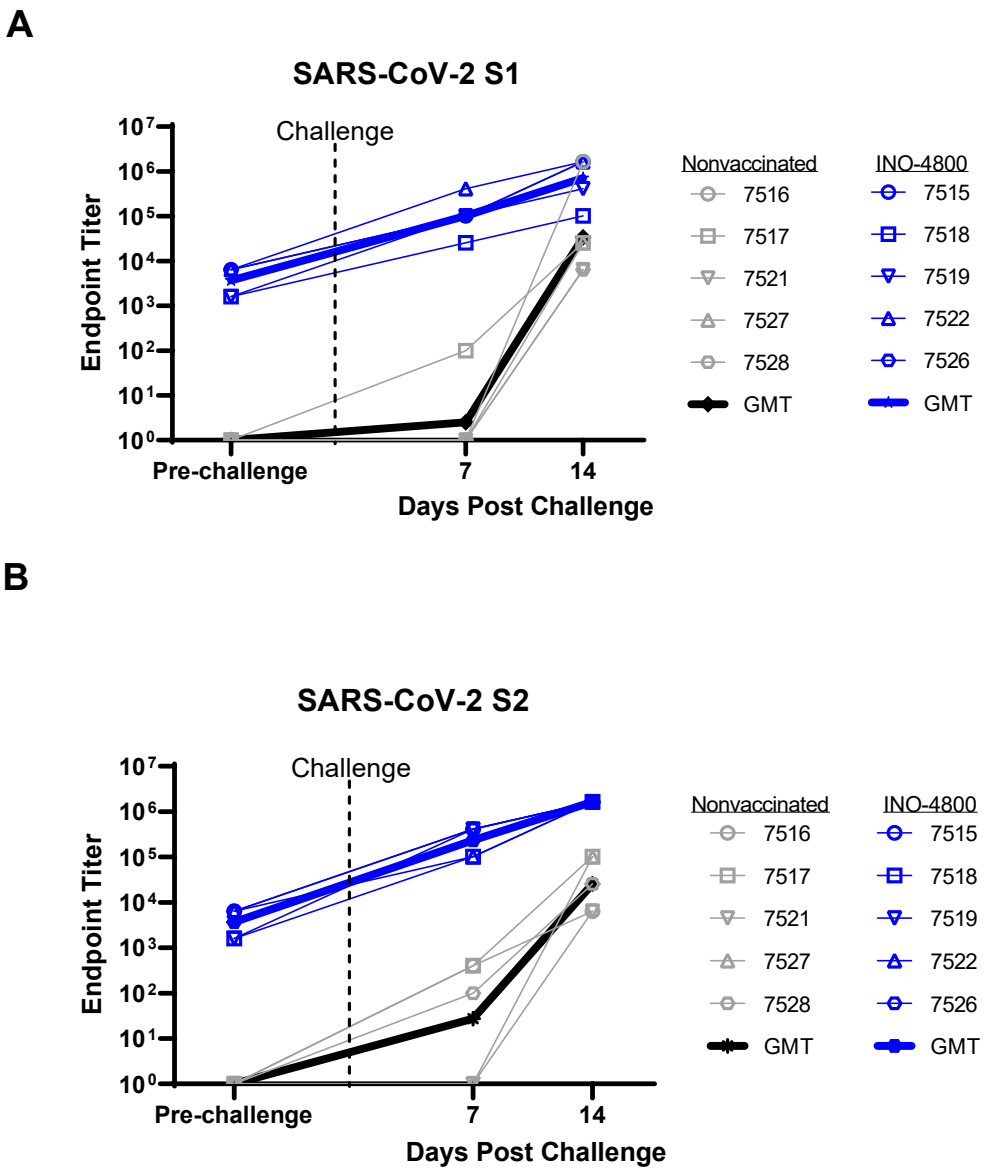

Figure S4. **Recall of humoral immune responses after viral challenge, Related to Figure 3.** (**A**) SARS-CoV-2 S1 protein and (**B**) SARS-CoV-2 S2 protein antigen binding of IgG in diluted NHP sera collected prior to challenge, during challenge and post challenge.