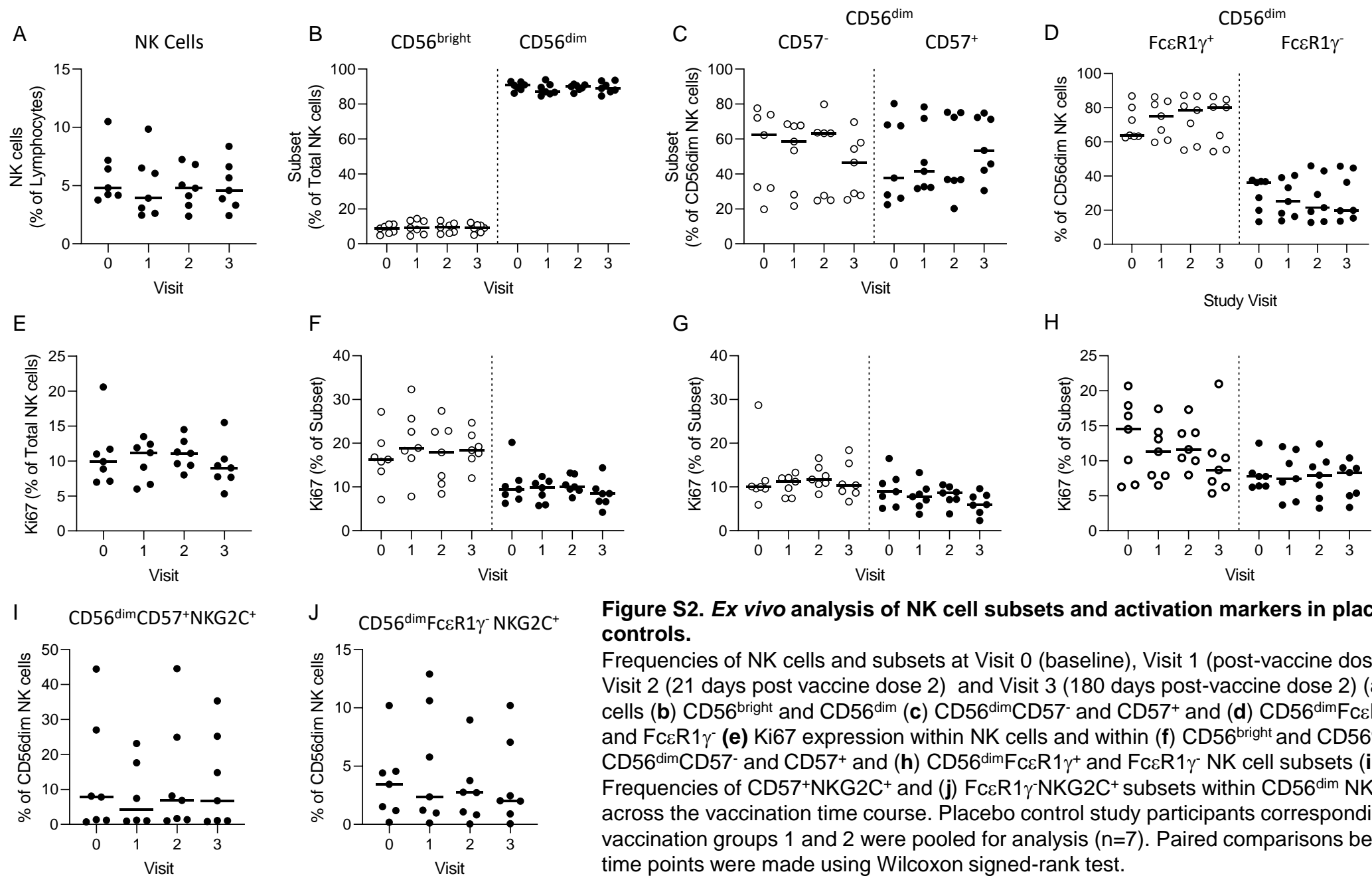


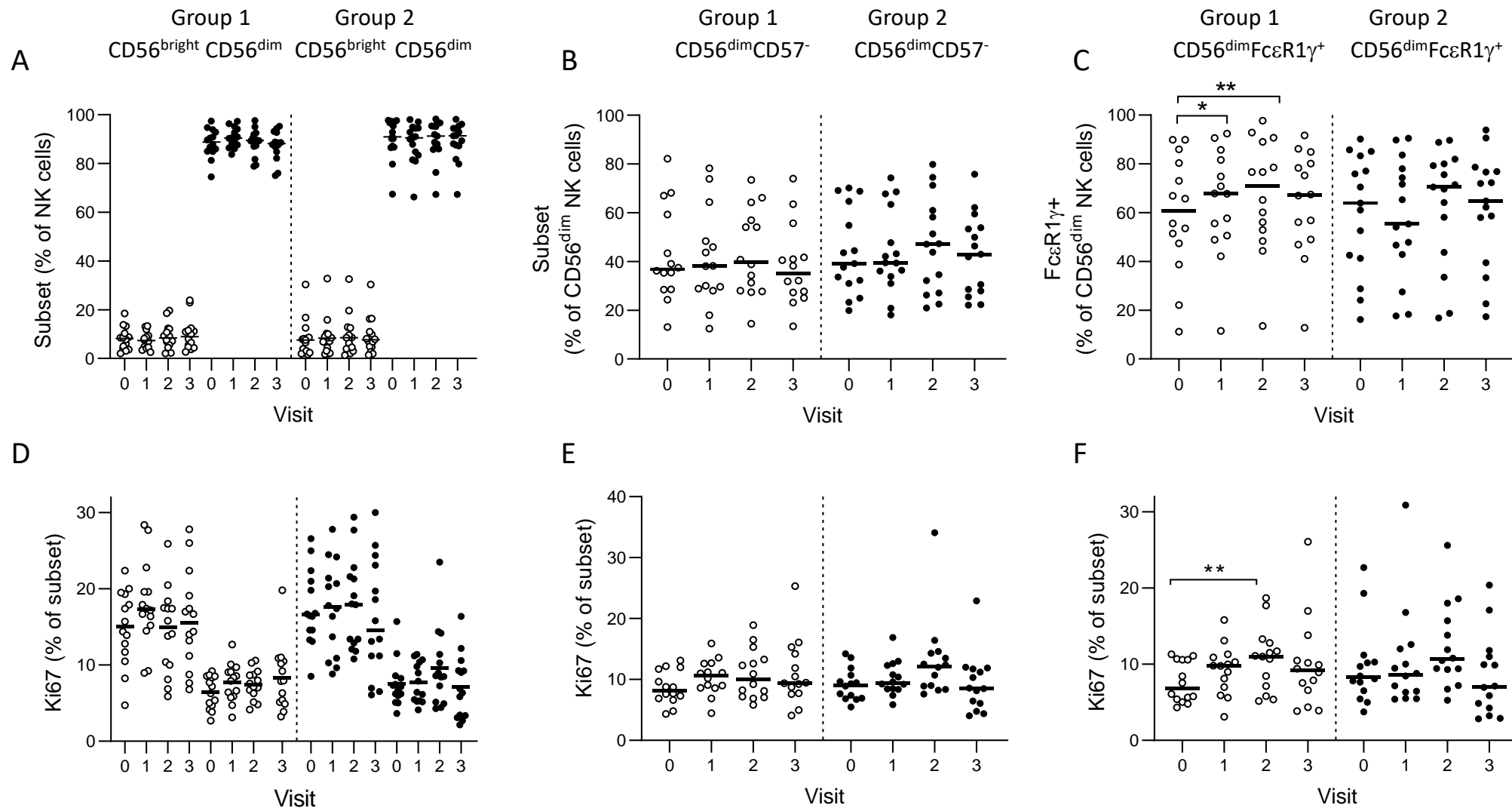
**Figure S1. Gating strategy for NK cell subsets measured ex-vivo**

For ex-vivo flow cytometric analysis, PBMC were first gated on singlet events (**a**) followed by lymphocytes (**b**) and viable lymphocytes (**c**). NK cells were defined as CD3-CD56<sup>+</sup> (**d**) and gated into CD56<sup>bright</sup> or CD56<sup>dim</sup> subpopulations according to CD57 (**e**), Ki67 (**f**) and adaptive NK cell subsets defined by FcεR1γ alone (**g**) and CD57/NKG2C (**h**) or NKG2C/FcεR1γ expression (**i**).



**Figure S2. *Ex vivo* analysis of NK cell subsets and activation markers in placebo controls.**

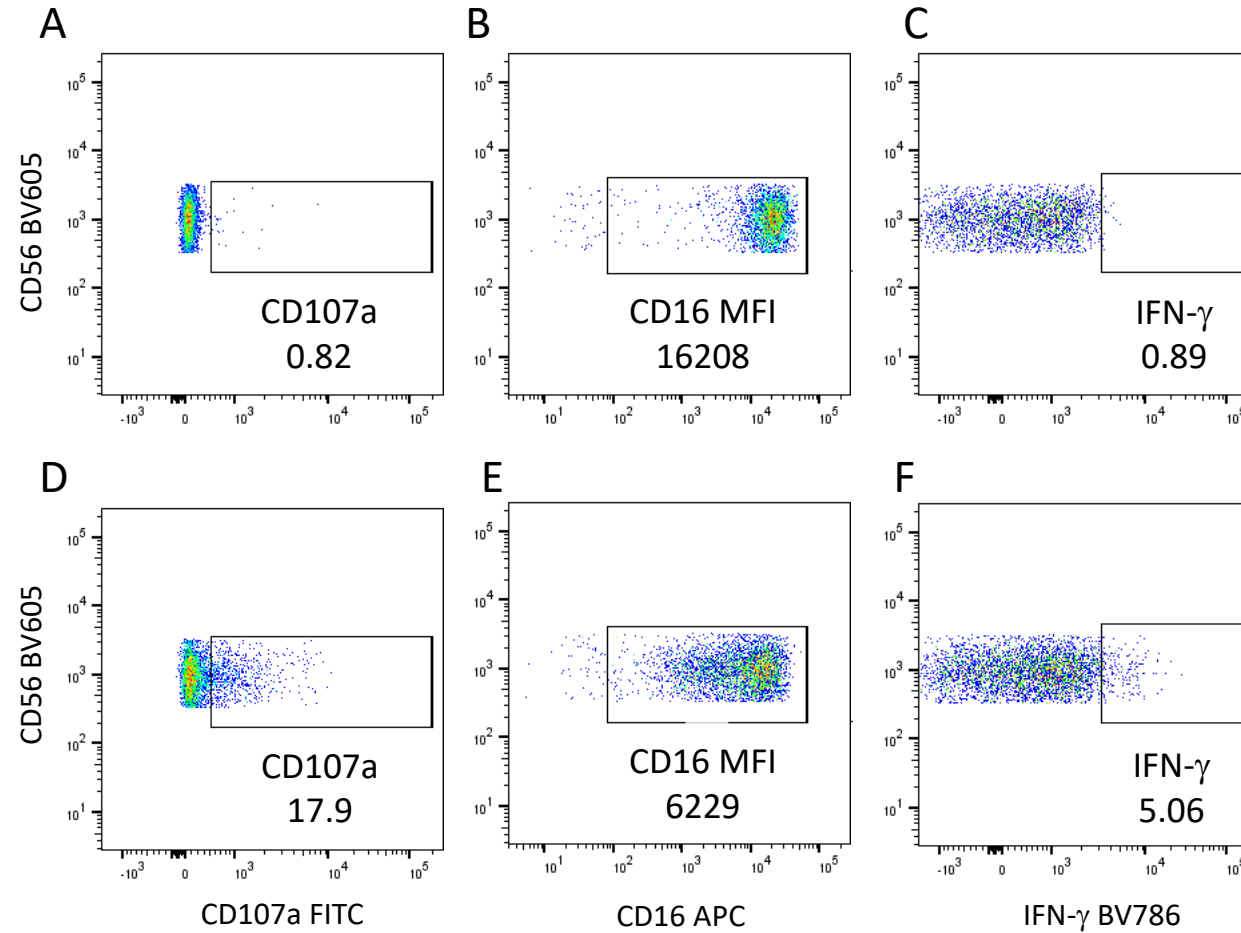
Frequencies of NK cells and subsets at Visit 0 (baseline), Visit 1 (post-vaccine dose 1), Visit 2 (21 days post vaccine dose 2) and Visit 3 (180 days post-vaccine dose 2) (a) NK cells (b) CD56<sup>bright</sup> and CD56<sup>dim</sup> (c) CD56<sup>dim</sup>CD57<sup>-</sup> and CD57<sup>+</sup> and (d) CD56<sup>dim</sup>FcεR1γ<sup>+</sup> and FcεR1γ<sup>-</sup> (e) Ki67 expression within NK cells and within (f) CD56<sup>bright</sup> and CD56<sup>dim</sup> (g) CD56<sup>dim</sup>CD57<sup>-</sup> and CD57<sup>+</sup> and (h) CD56<sup>dim</sup>FcεR1γ<sup>+</sup> and FcεR1γ<sup>-</sup> NK cell subsets (i) Frequencies of CD57<sup>+</sup>NKG2C<sup>+</sup> and (j) FcεR1γ<sup>-</sup>NKG2C<sup>+</sup> subsets within CD56<sup>dim</sup> NK cells across the vaccination time course. Placebo control study participants corresponding to vaccination groups 1 and 2 were pooled for analysis (n=7). Paired comparisons between time points were made using Wilcoxon signed-rank test.



**Figure S3. Impact of dosing interval on NK cell subset distribution *ex vivo***

*Ex vivo* flow cytometric data was analysed in actively vaccinated individuals in Group 1 (dose 2 = 28 days post dose 1; n=14) and Group 2 (dose 2 = 56 days post dose 1; n=15). Overall frequencies of CD56<sup>bright</sup> and CD56<sup>dim</sup> NK cells (a) and CD57 (b) or FcεR1γ (c) defined subsets and the frequency of Ki67 within each subset (d-f) is shown for both groups. Comparisons between paired data points within each group were made using Wilcoxon signed-rank test. \* p<0.05, \*\* p<0.01.

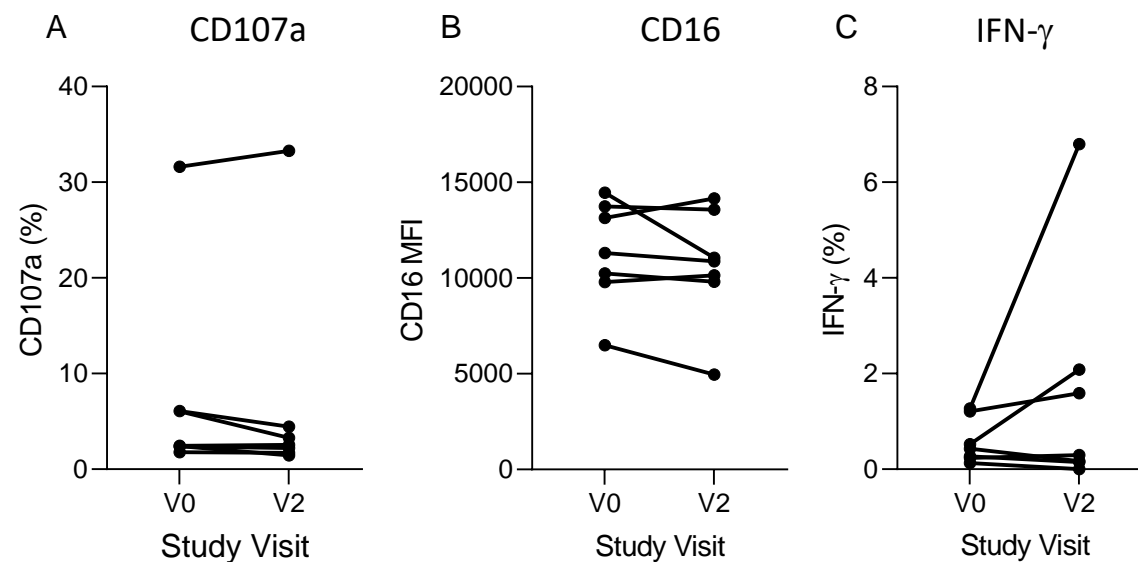
Baseline (V0)



21 days  
Post-dose 2 (V2)

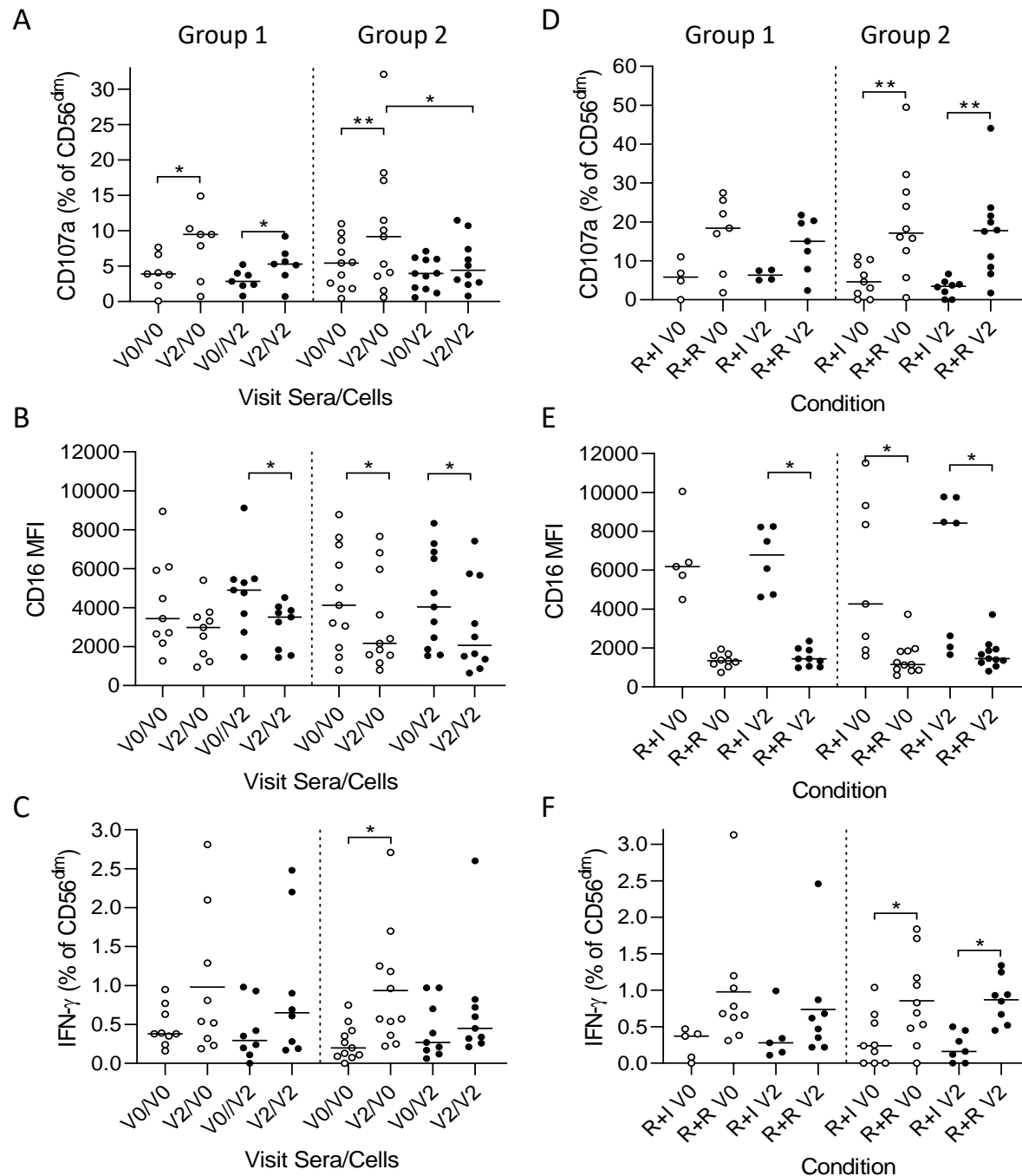
#### Figure S4. Gating strategy for in-vitro ADNKA assays

ADNKA responses were analysed after culture with recombinant Ebola glycoprotein and pre- or post-vaccination sera. Pseudocolour plots are shown for CD107a (a, d), CD16 (b, e) and IFN-γ (c, f) expression within gated CD56<sup>dim</sup> NK cells from a representative donor after culture with baseline (V0) serum (a-c) or post-dose 2 (V2) (d-f) serum.



**Figure S5. Antibody-dependent NK cell responses to immobilized EBOV GP in placebo controls.**

CD107a (a), CD16 MFI (b) and IFN-γ (c) expression within gated CD56<sup>dim</sup> NK cells from a standard PBMC readout (from a single unvaccinated donor) cultured with serum samples from individual trial donors collected pre-vaccination (Visit 0) and 21 days post-dose 2 (Visit 2). Data are presented for combined vaccination groups and plots show individual data points before after vaccination (a-c).



### Figure S6. The Impact of dosing interval on ADNKA responses of autologous NK cells

ADNKA responses were analysed in actively vaccinated individuals in Group 1 (dose 2 = 28 days post dose 1; n=8) and Group 2 (dose 2 = 56 days post dose 1; n=10). PBMC collected pre-vaccination (V0) and 21 days post-dose 2 (V2) were cultured with EBOV GP in the presence of autologous serum from the same visit or with pooled serum from Visit 2 (V2p) (**a-c**). Positive and negative control stimuli were Raji B cells + Rituximab<sup>TM</sup> (R+R) or isotype control antibody (R+I), respectively (**d-f**). Responses were analysed by flow cytometry for (**a, d**) CD107a, (**b, e**), CD16 mean fluorescence intensity (MFI) and (**c, f**) IFN- $\gamma$  expression. Individual data points are shown for 18 participants tested in this assay with a line representing median values for all tested donors. Comparisons between visits were performed using one-way ANOVA mixed effects analysis with Geisser-Greenhouse correction. \*p<0.05, \*\* p<0.01.