Different human vaccine adjuvants promote distinct antigen-independent immunological signatures tailored to different pathogens

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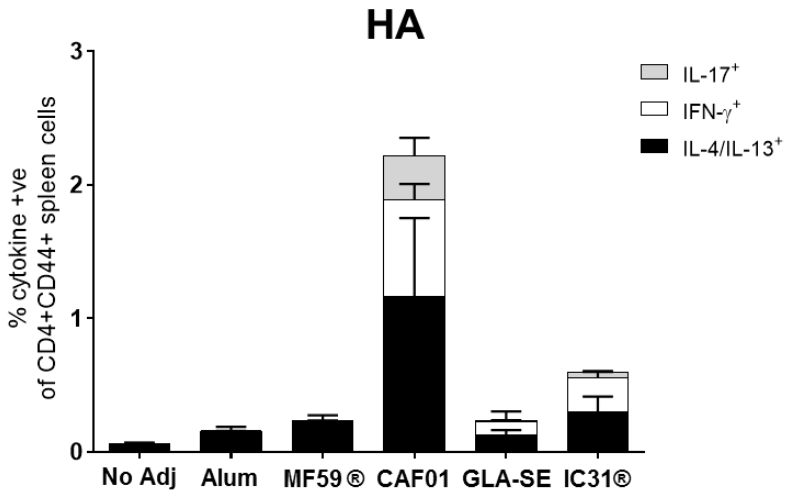
Figure 1S. icFACS analysis of vaccine responses measured by cytokine expression in HA specific CD4 T-cells. Mice were vaccinated 3 times s.c. with 1 μg of HA formulated in Alum, MF59® CAF01, GLA-SE or IC31® with 3 week intervals. Splenocytes from four individual mice, isolated two weeks after the third immunization, were stimulated with 1 μg/ml of recombinant HA O/N + 4h in the presence of BFA. Cells were gated as follows: live > singlets > lymphocytes > $CD3^+$ > $CD4^+$ versus $CD8^+$. Cytokine-producing cells (IFN-γ, IL-17 or IL-4/IL-13) within the $CD4^+CD44^{high}$ population were measured. Bar charts show the frequency of each cytokine subset being $CD44^{high}$ out of the total CD4 T cell population. Bars represent the mean + SEM of four individual mice. The background from non-stimulated samples have been subtracted.

Figure 2S. An icFACS analysis of vaccine responses measured by cytokine co-expression in H56 or MOMP-specific CD4 T-cells. Mice were vaccinated 3 times s.c. with 5 μg of H56 or MOMP formulated in Alum, MF59® CAF01, GLA-SE or IC31® with 3 week intervals. Splenocytes from four individual mice, isolated two weeks after the third immunization, were stimulated with 2 μg/ml of recombinant H56 or MOMP for 1h + 5h in the presence of BFA. Cells were gated as follows: singlets > lymphocytes > $CD4^+$ versus $CD8^+$. Cytokine-producing cells (IFN-γ, TNF-α, and IL-2) within the $CD4^+CD44^{high}$ population were divided into seven distinct subpopulations based on Boolean gating and are shown as bar and pie charts. Bar charts show the frequency of each cytokine subset being $CD44^{high}$ out of the total CD4 T cell population. Bars represent the mean + SEM of four individual mice. Pie charts show the relative contribution of each cytokine subpopulation to the Ag-responsive CD4 T cell population. The background from non-stimulated samples have been subtracted. The experiment was repeated with similar results.

Supplementary Figure S1



Supplementary Figure S2