**Supplementary Information for “OpenCyto: An Open Source Infrastructure for Scalable, Robust, Reproducible, and Automated, End-to-End Flow Cytometry Data Analysis”**

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**Supporting Text S1**

*Model Description*

In order to identify cell subsets that exhibit antigen-specific changes upon vaccination, we fit a linear mixed effects model to the background subtracted proportions of cytokine positive cells within each population and gating method. Without loss of generality, for a fixed cell subset and stimulation condition, we let

be the proportions of cytokine positive cells in the antigen stimulated () and non-stimulated () sample from subject, visit , and vaccine regimen , and be the background subtracted proportion of cytokine positive cells. We model:

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where is the intercept for the cell subset, is a subject-level random effect with , is the effect of visit , is the effect of vaccine regimen , and is the interaction between visit and vaccine regimen , and . We want to test whether (i.e., whether there is an increase in the proportion of cytokine positive cells at the post-vaccine time-point compared to the pre-vaccine time-point) for each vaccine regimen. We fit the model with the R’s *lmer* function from the *lme4* package, and perform linear hypothesis tests using the *glht* and *contrasts* packages.