

Samples were taken from 6 participants on days 0, 1, 2, 7, 21, 22, 28 and 42 post-vaccination, however only on day 1 post vaccination, data for 3 of the 6 participants is missing, ostensibly the data from two female participants and one male participant. I did not find any justification or explanation for this exclusion of samples in the article text, but the article is very long, and I may have missed it.

The comment is correct that data for 3 of the 6 participants is missing on day 1 post-vaccination; however, we did not exclude any data from the analysis. Rather, our study protocol involved blood collection on days 0, **1 or 2**, 7, 21, 22 or 23 and 42 post-vaccination. In **Extended Data Fig. 1a, we used one column (1/2) to indicate day 1 or 2**. We also included the statement *"We performed cellular indexing of transcriptomes and epitopes by sequencing (CITE-seq) of 45 peripheral blood mononuclear cell (PBMC) samples from 6 individuals (Extended Data Fig. 1a)"* in the description to suggest the same. That is, we have 6 subjects, with 7 time points (i.e. days 0, 2, 7, 21, 22, 28 and 42) per subject PLUS 3 of these subjects also had a day 1 time point. This adds up to 45 PBMC samples.

Of the six participants we performed CITE-seq on, all 6 participants had provided blood on day 2 post vaccination and the data are available. In addition, three participants provided blood on day 1 for which the data are available, which worked to our advantage to provide a finer kinetics.

Only samples from female participants were removed. **Incorrect. We did not "remove" any samples.**

Certain genes which are Y-linked and therefore only present in men are useful biomarkers for determining the sex of the study participants, for example ATP-dependent RNA helicase DDX3Y (DDX3Y) and histone demethylase UTY (UTY). Inspecting the raw counts for DDX3Y and UTY reveals that participants 2051 and 2052 appear to have "swapped" genders only on day 1 post vaccination.

As mentioned above, we did not remove data of any participant. We included a table below showing the age, gender, and sample availability of the 6 participants included in the CITE-seq analysis. The information is available in the phenotype file associated with our GEO submission.

Participant ID	Reported gender	Age	Day 1 availability
2047	Male	31	Yes
2049	Male	35	Yes
2051	Female	24	Yes
2052	Male	54	No
2053	Female	30	No
2055	Female	34	No

As you can see, 2052 does not have a day 1 sample. We are not sure how the reader arrived at the conclusion that "2051 and 2052 appear to have "swapped" genders only on day 1

post vaccination.” This conclusion is not accurate because 2052 did not even have a day 1 sample. Furthermore, as described below, the expression of Y-linked genes in these subjects are entirely consistent with their reported gender.

In addition, we independently examined Y-linked genes in the gene expression data and found that the gene expression data reflects gender. For e.g., the Y-linked genes including *DDX3Y* and *UTY* are expressed only in males (Fig. 1). In contrast, *XIST* is completely absent in males, as expected. We see that there may be a slightly higher expression of *DDX3Y* and *UTY* in 2051 (reported female) and *XIST* in 2049 (reported male) on day 1 but at much lower levels than in all 3 males and at other time points. In summary, the gene expression data of individuals is not “swapped”, and we did not remove participants of a particular gender.

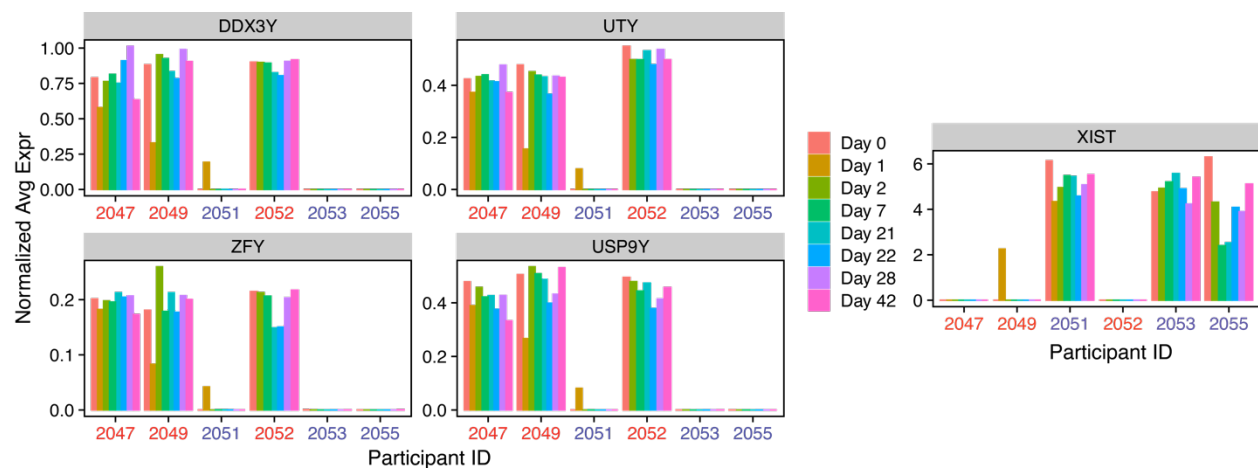


Fig. 1. Normalized expression of genes known to be expressed in somatic cells of males or females only.