Dear Dr. Weiss,

I very much appreciate the response, and I apologize for my mistake in the distinction between groups, which resolves the first concern and clears up some confusion on my part. However, the response to the second concern is very surprising, since the anomalies I've described are clearly visible in the figure provided by the authors (Response Fig. 1). Participant 2051 is reported as female, and therefore the expression of each of the shown Y-linked genes should be zero, not "much lower levels" than males. The authors' analysis of "slightly higher [than zero]" is analogous to claiming "the amount of hands this person has is slightly higher than two but much lower than four".

The same anomaly occurs in all 6 of 6 Y-linked, quantifiable genes given by (InanlooRahatloo, Liang et al. 2017), namely EIF1AY, RPS4Y1, USP9Y, ZFY, PRKY and UTY (data shown below, visualized in part by Response Fig. 1). Furthermore, the data and Response Fig. 1 show that participants 2049 and 2051 appear to be both male and female on day 1.

Day ->	0	1	2	7	21	22	28	42
-- All values for XIST--								
2047_	1	0	0	0	0	0	0	0
2049_	1	9254	0	1	0	0	0	1
2051_	15345	20030	20809	22051	22801	21231	25572	11350
2052_	0	-	0	0	0	1	0	0
2053_	20125	-	24166	23436	28426	27946	22269	24512
2055_	15280	-	148	8920	5257	14381	15190	19279

-- All values for DDX3Y--								
2047_	2628	1716	1730	3136	2445	3370	2762	2009
2049_	3284	1226	4038	3257	3867	3831	3606	3551
2051_	0	1096	0	0	2	0	1	2
2052_	3270	-	2893	3764	2803	2911	3874	3668
2053_	0	-	0	0	0	0	0	1
2055_	0	-	0	0	0	0	1	0

```
-- All values for UTY--
2047_      1318  1002  920   1645  1369  1573  1258  1130
2049_      1656  542   1908  1551  1886  1651  1506  1646
2051_      0     423   0     0     0     0     0     0
2052_      1903  -     1554  1989  1729  1641  2213  1941
2053_      0     -     0     0     0     0     0     0
2055_      0     -     0     0     0     0     0     0
```

```
-- All values for USP9Y--
2047_      1256  940   857   1357  1236  1228  1030  847
2049_      1593  819   1944  1624  1977  1486  1261  1875
2051_      0     280   0     1     0     0     0     0
2052_      1436  -     1312  1552  1429  1178  1614  1573
2053_      1     -     2     0     1     1     0     2
2055_      1     -     0     1     0     1     0     0
```

```
-- All values for RPS4Y1--
2047_      25175 14188 16638 25295 24068 22968 18114 17492
2049_      22997 10697 26873 23040 30730 24383 20662 24640
2051_      0     4165  0     0     0     0     1     5
2052_      21654 -     17296 23323 20888 18159 25540 24033
2053_      1     -     0     0     0     0     0     0
2055_      0     -     0     0     1     0     0     0
```

```
-- All values for EIF1AY--
2047_      1695  1645  1157  1976  1525  1806  1339  1644
2049_      1997  1001  2093  1765  2397  2682  1780  1962
2051_      0     495   1     1     0     0     0     1
2052_      1747  -     1449  2016  1555  1724  2004  1653
2053_      0     -     1     2     0     0     0     0
2055_      1     -     0     0     0     0     0     1
```

```
-- All values for ZFY--
2047_      697   531   463   785   773   825   613   570
2049_      713   305   1251  705   1027  831   870   881
2051_      0     230   0     1     2     1     0     0
2052_      832   -     706   855   517   565   916   938
2053_      1     -     2     0     0     1     0     1
2055_      0     -     0     0     0     0     0     3
```

-- All values for PRKY--								
2047_	1317	1183	990	1475	1157	1279	1065	1024
2049_	1760	867	1664	1622	1922	1818	1328	1738
2051_	0	350	1	0	0	2	0	0
2052_	1295	-	1256	1661	1390	1285	1308	1591
2053_	2	-	1	2	2	0	1	1
2055_	0	-	0	0	0	4	2	0

As I described in my previous email, the fact that this value is non-zero for just one Y-linked gene, at any point in time is biologically impossible. If the data supplied by the authors were to be genuine, this finding would constitute a groundbreaking discovery, i.e. that vaccination with BNT162b2 induces transient expression of Y-linked genes in women.

However, there are additional problems with the RNA-Seq dataset, which covers the same participants. As the authors have emphasized, participant 2049 is reported as male and 2051 as female, but the expression of DDX3Y shows the opposite:

Day ->	BL	1	2	7	21	22/23	28
PID2047	275	1042	373	376	191	422	876
PID2049	0	0	0	1	0	0	0
PID2051	2103	341	241	175	419	414	236
PID2052	673	-	291	440	418	450	305
PID2053	0	-	0	0	0	0	1
PID2055	427	-	296	263	236	171	339

Furthermore the data appears to be from 4 men and 2 women, not from 3 each as reported in the phenotype file. Therefore, data in both datasets appears to have been modified, calling the results of the entire study into question. I've summarized the various genders in the table below.

Participant	Reported Gender (both datasets)	Actual Gender RNA-Seq	Actual Gender CITE-seq (except Day 1)	Actual Gender CITE-Seq (Day 1)
2047	M	M	M	M
2049	M	**F**	M	**M and F**
2051	F	**M**	F	**M and F**
2052	M	M	M	
2053	F	F	F	
2055	F	**M**	F	

I remain hopeful that there is some completely benign explanation for this. In summary, gene data of individuals appears to have been not only “swapped” but consistently mislabeled across multiple data sets, and the fact that participants expressed both male- and female-specific genes on the same day strongly suggests that data may have been retroactively inserted or changed. Furthermore, the authors’ inability to recognize or address these glaring inconsistencies in their own figure is remarkable.

Sincerely,

Johannes

BIBLIOGRAPHY

InanlooRahatloo, K., G. Liang, D. Vo, A. Ebert, I. Nguyen and P. K. Nguyen (2017). "Sex-based differences in myocardial gene expression in recently deceased organ donors with no prior cardiovascular disease." PloS one **12**(8): e0183874-e0183874.