

Additional file 4

Supplementary Text

Alternatively to the approach used by *Flimma*, cohort batch effects could be adjusted for based on one or several first principal components. However, our results suggest that PCA-based batch effect correction with one to four principal components seems to be not effective for gene expression data (Additional file 8: Table S7).

To additionally validate the lists of differentially expressed genes produced by *Flimma* and *limma voom* with PCA-based correction on three GEO datasets, we compared them with the list obtained on TCGA-BRCA in centralized analysis. Since TCGA-BRCA is much larger than the GEO dataset (850 samples vs. 132) and seems to be unaffected by center-specific batch effects (Figure 4), we assumed that the signature discovered in this dataset by *limma voom* is reliable and, thus, used it as the second comparison partner. Again, on GEO datasets, the list of differentially expressed genes obtained by *Flimma* was more similar to the TCGA-BRCA gene list than lists produced by *limma voom* with 1-4 first principle components taken as covariates. This validates the reliability of *Flimma* regarding its ability to replicate the result obtained on a larger and more heterogeneous cohort (Additional file 8: Table S7).

Supplementary Figures

TCGA-BRCA: Lum vs Basal

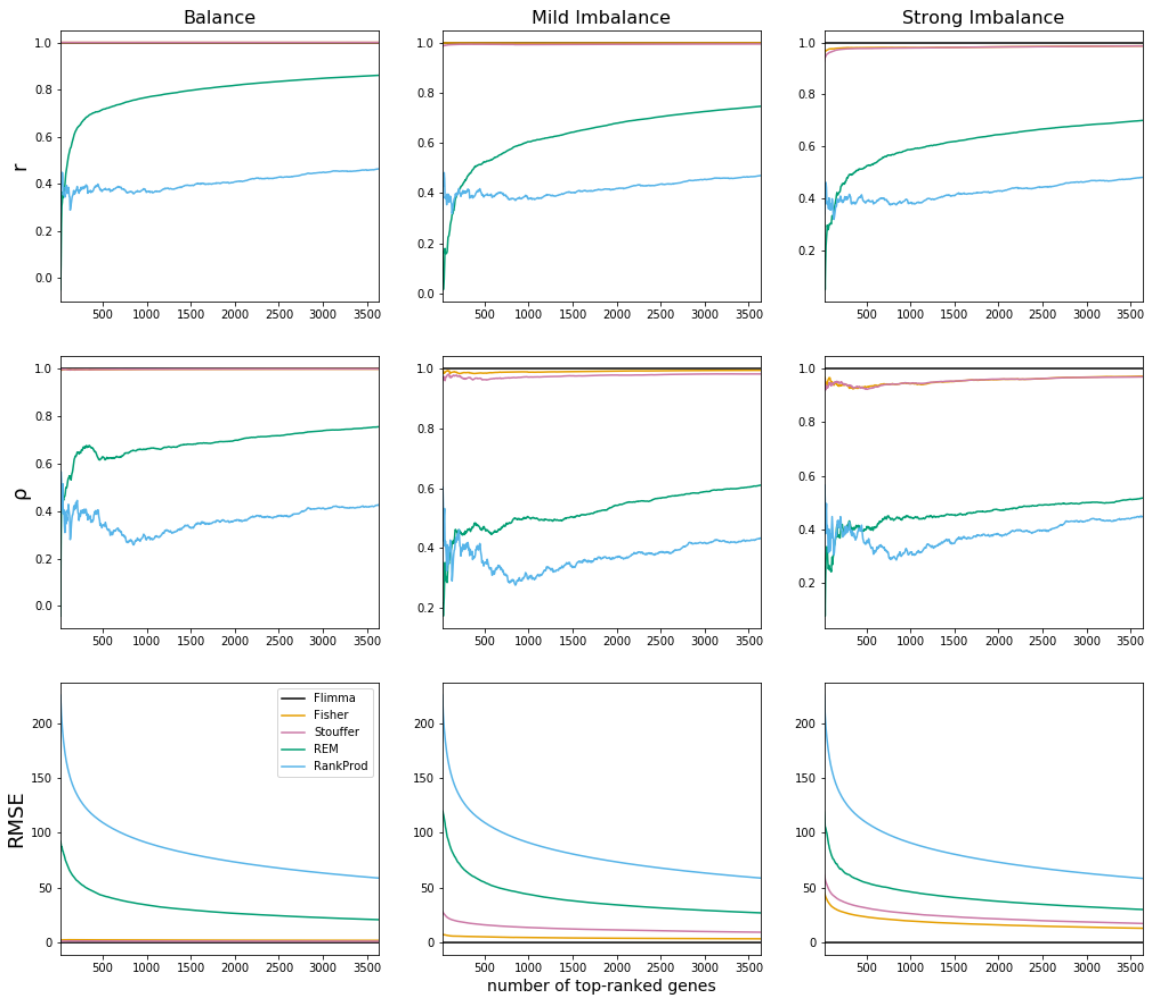


Figure S1. The dependency of Pearson's and Spearman's correlation coefficients and RMSE on the number of top-ranked genes considered to be differentially expressed in TCGA-BRCA data.

GTEx skin: sun-exposed vs not exposed

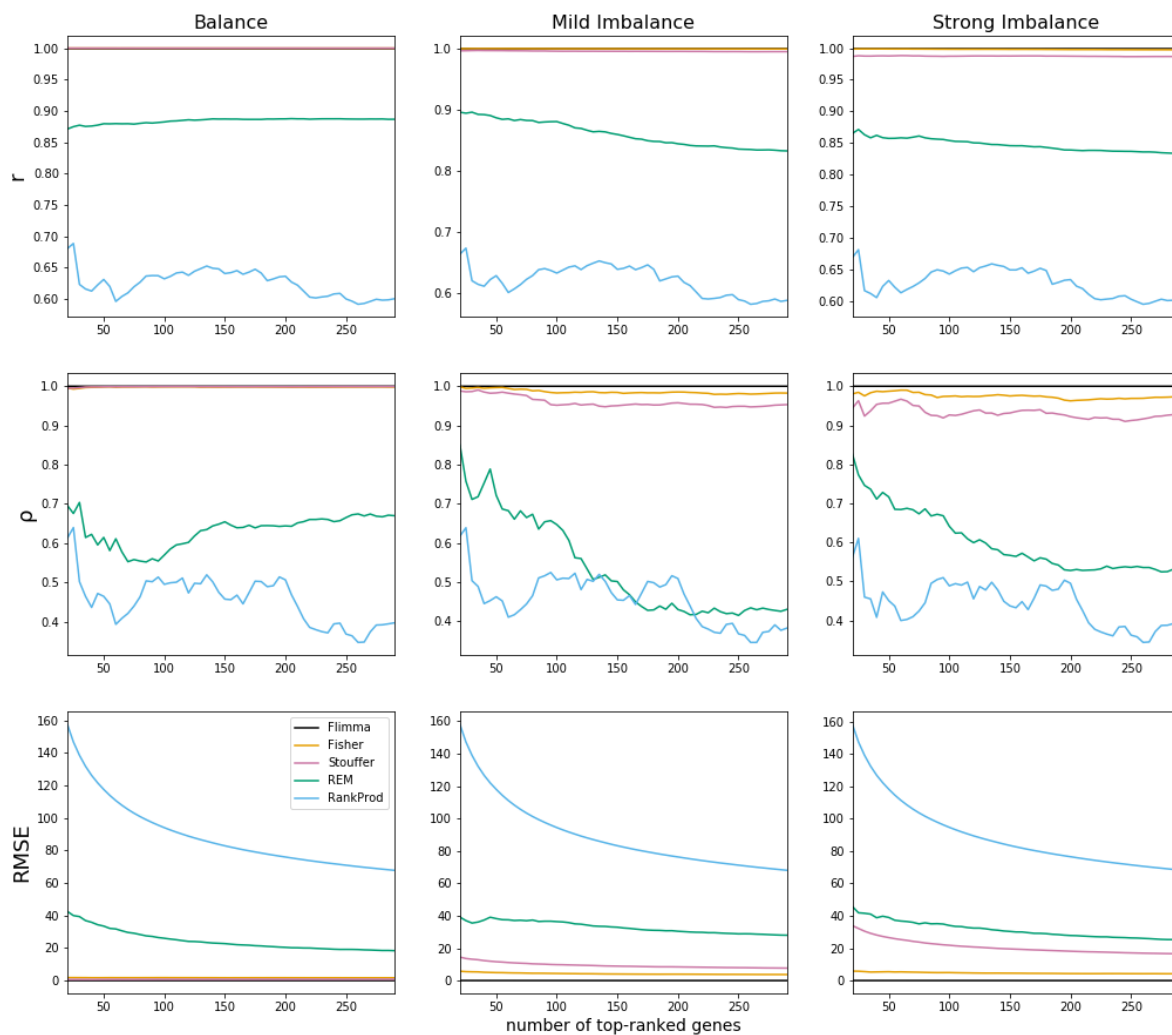


Figure S2. The dependency of Pearson's and Spearman's correlation coefficients and RMSE on the number of top-ranked genes considered to be differentially expressed in GTEx skin dataset.

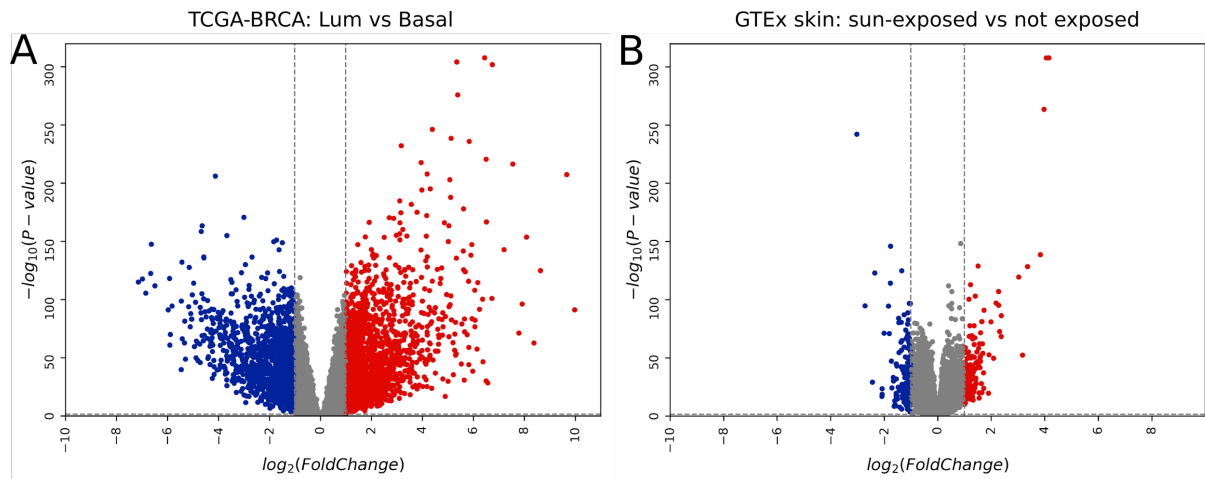


Figure S3. The results of differential expression analysis performed by *limma*-voom on the aggregated TCGA-BRCA (A) and GTEx Skin datasets (B). 3635 and 288 genes were differentially expressed with absolute log-fold change $|\log_{2}FC| > 1$ and BH-adjusted p-value < 0.05 according to *limma* voom applied on pooled TCGA-BRCA and GTEx Skin datasets respectively.