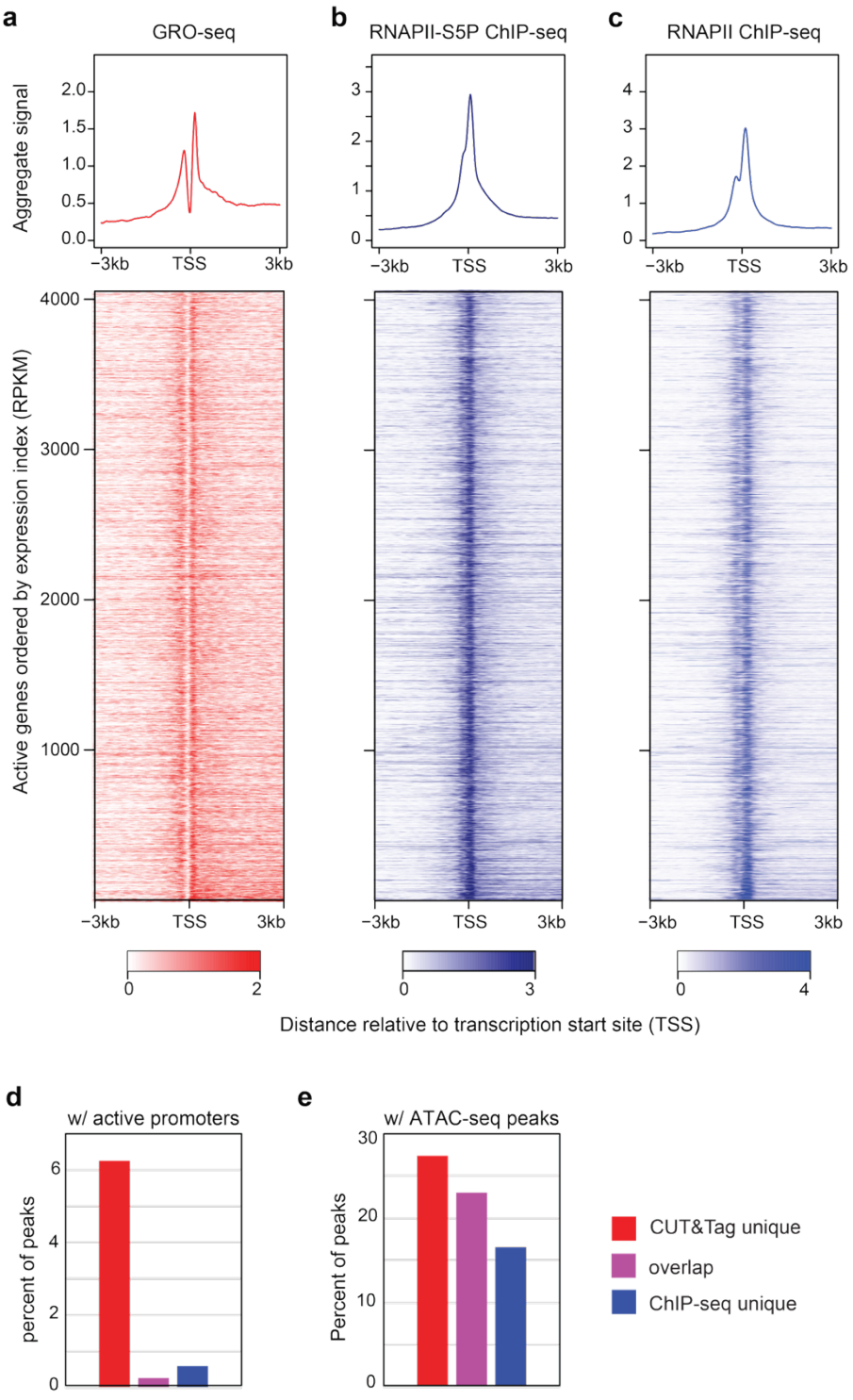

Figure S1. Enrichment of H3K27me3 CUT&Tag signal on active gene promoters. (**a**-**c**) Signal patterns across active promoter regions in K562 cells, for two GRO-seq (**a**), RNAPII-s5p ChIP-seq (**b**), and RNAPII ChIP-seq (**c**). The upper panels are the normalized aggregate signal patterns. The lower panels are heatmaps of the signal patterns at promoter regions (TSS ±3kb) of the actively transcribed genes. Rows correspond across heatmaps. (**d**, **e**) Percentage of CUT&Tag-unique (red), CUT&Tag-ChIP-overlap (purple), and ChIP-seq-unique (blue) peaks that overlap with active gene promoter regions (TSS ±1kb) (**d**) and with ATAC-seq peaks (**e**).

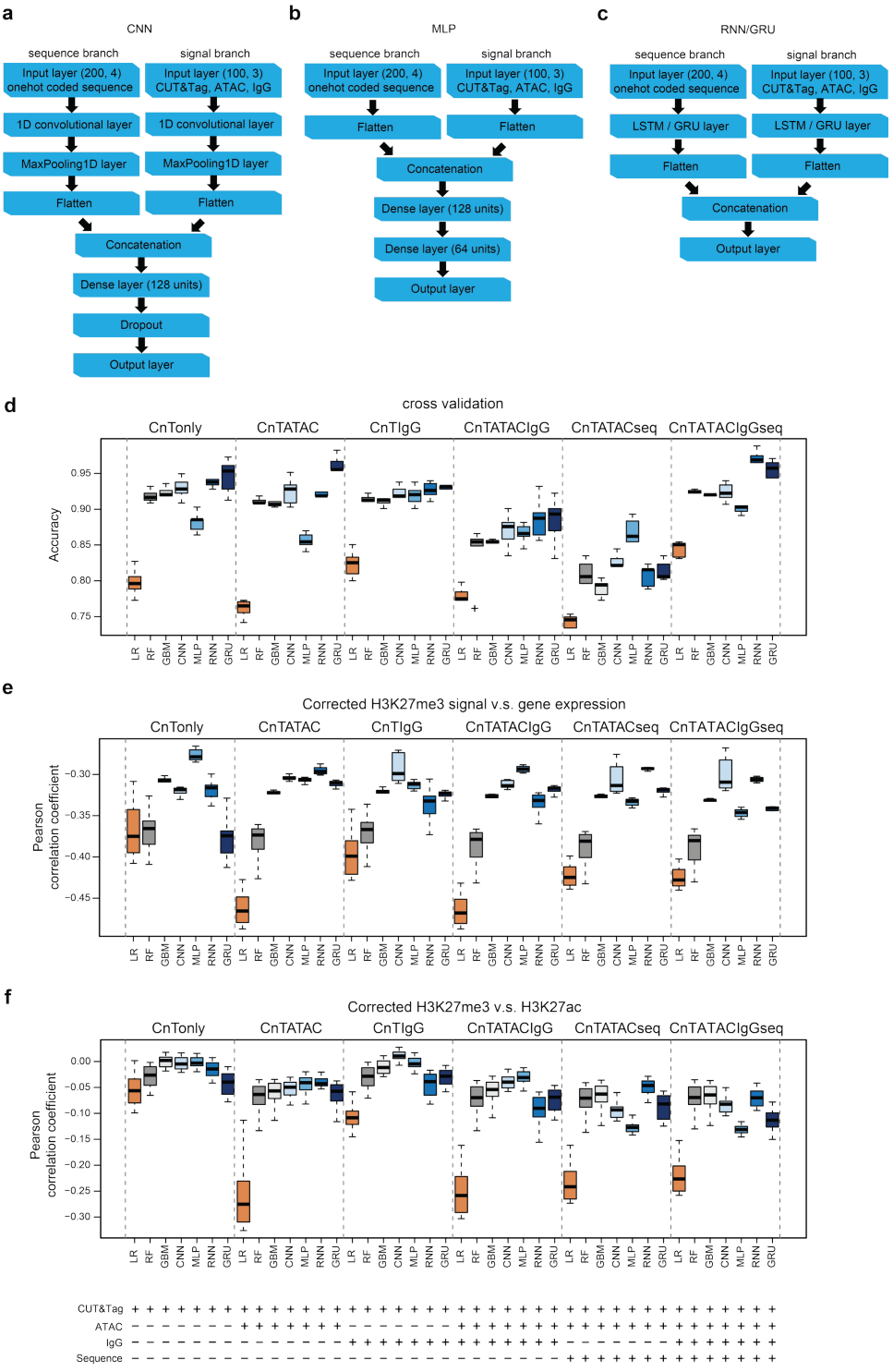

Figure S2. Performance of different models for H3K27me3 CUT&Tag. (**a-c**) Schematic of the (**a**) CNN, (**b**) MLP, (**c**) RNN, and GRU models. (**d**) The accuracy of different prediction models was evaluated using 5-fold cross-validation results. (**e**) Pearson correlation coefficient (y-axis) between the model-derived H3K27me3 score and gene expression across all gene promoter bins in the genome. (**f**) Pearson correlation between the model derived H3K27me3 CUT&Tag score and H3K27ac ChIP-seq signal level across all bins with CUT&Tag reads in the genome. Models with the same feature combination were displayed in the same block. Each data point in a boxplot represents a CUT&Tag sample.

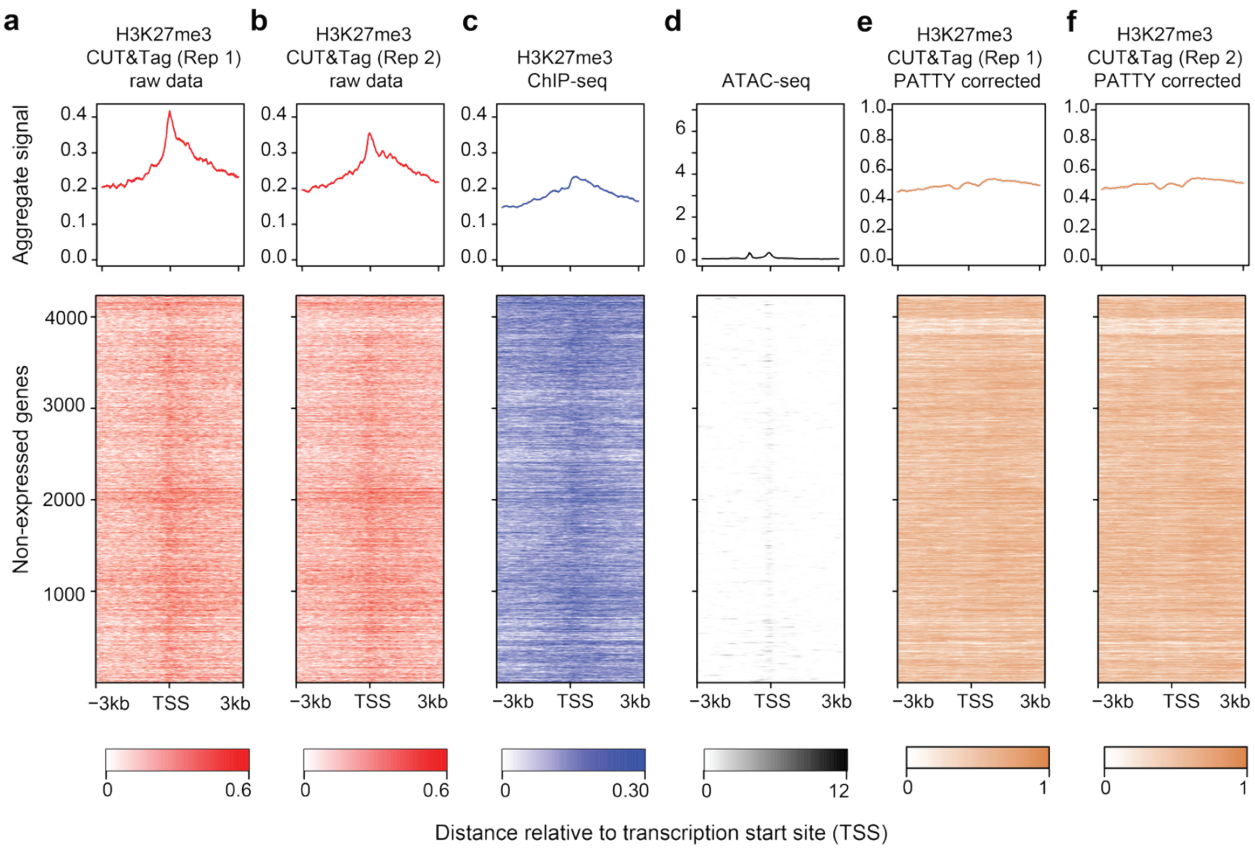

Figure S3. H3K27me3 CUT&Tag signal patterns on repressive genes before and after PATTY correction.
(**a**-**f**) H3K27me3 CUT&Tag signal patterns across repressive gene promoter regions before (**a**, **b**) and after (**e**, **f**) bias correction by PATTY. ChIP-seq (**c**) and ATAC-seq (**d**) signals across the same regions are shown for reference. The upper panels are the normalized aggregate signal patterns. The lower panels are heatmaps of the signal patterns at promoter regions (TSS ±3kb) of the repressive genes. Rows correspond across heatmaps.

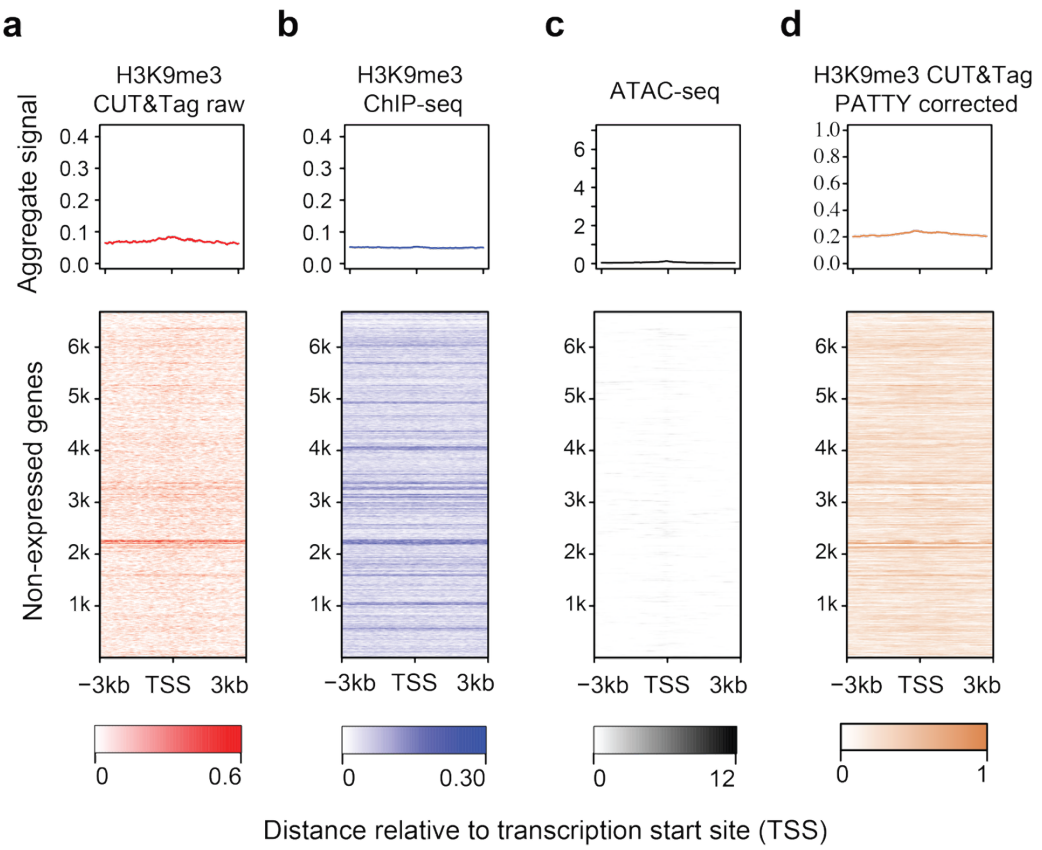

Figure S4. H3K9me3 CUT&Tag signal patterns on repressive genes before and after PATTY correction. Similar to **Figure S3** but for H3K9me3 CUT&Tag data across repressive genes in the HCT116 cell line**.**

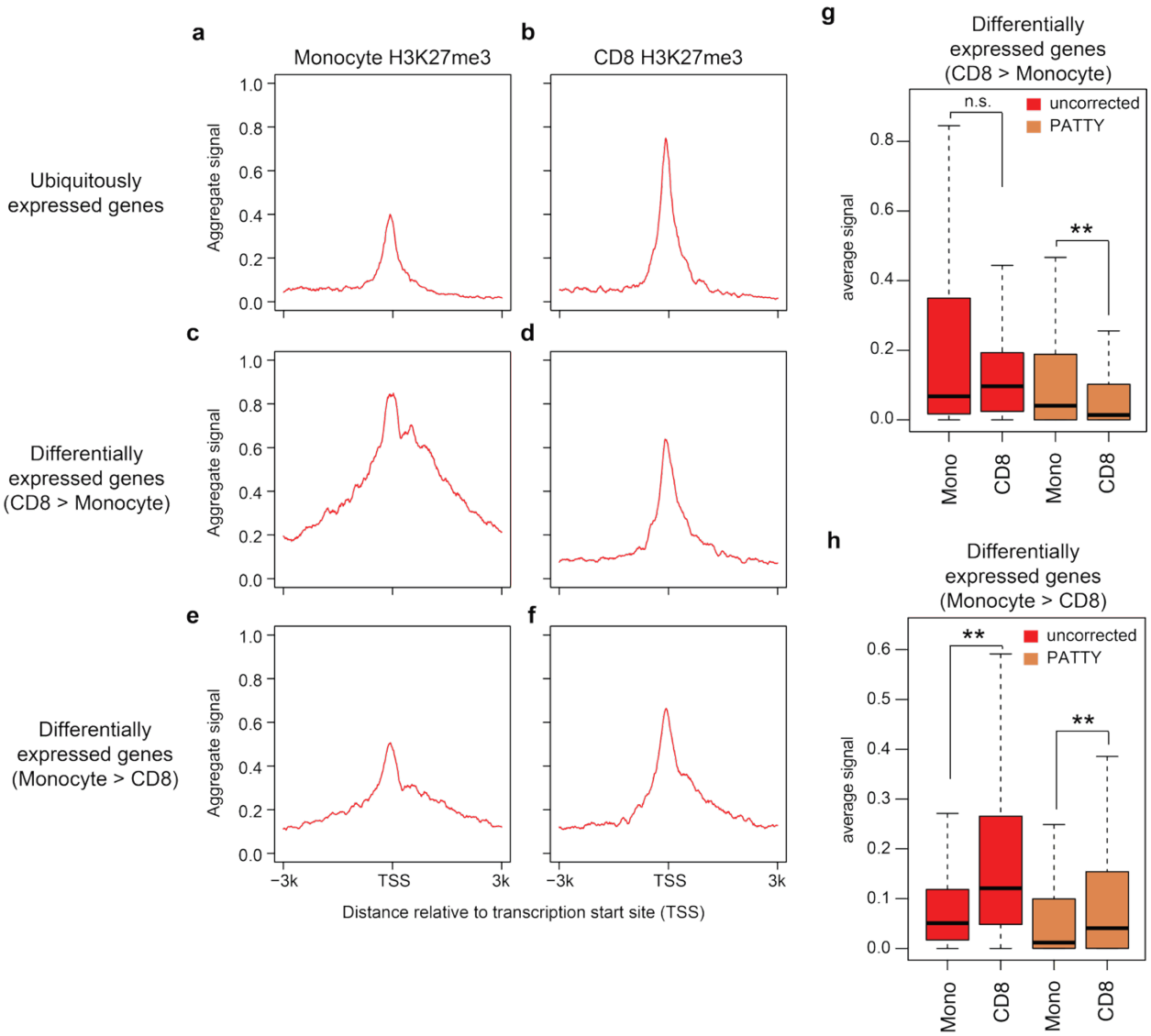

Figure S5. H3K27me3 CUT&Tag signal in scCUT&Tag-pro data. (**a**-**f**) Aggregate H3K27me3 scCUT&Tag-pro signal patterns across the promoter regions (TSS ±3kb) of PBMC-ubiquitously expressed genes (**a**, **b**), differentially expressed genes (DEGs) between Monocytes and CD8+ T cells that are higher in CD8 than Monocytes (**c**, **d**), and higher in Monocytes than CD8 (**e**, **f**), for scCUT&Tag-pro signal as pseudo-bulk in Monocytes (**a**, **c**, **e**) and in CD8+ T cells (**b**, **d**, **f**). (**g**, **h**) Average H3K27me3 scCUT&Tag-pro signal levels in a pseudo-bulk of cell group (labeled under x-axis) on the promoter regions (TSS ±3 kb) of DEGs high in CD8 (**g**) and DEGs high in Monocytes (**h**), comparing uncorrected data (red) with PATTY-corrected data (yellow). Each data point in a boxplot represents the H3K27me3 scCUT&Tag-pro signal for a gene. **, $p < 0.01$, by one-sided Wilcoxon signed-rank test.

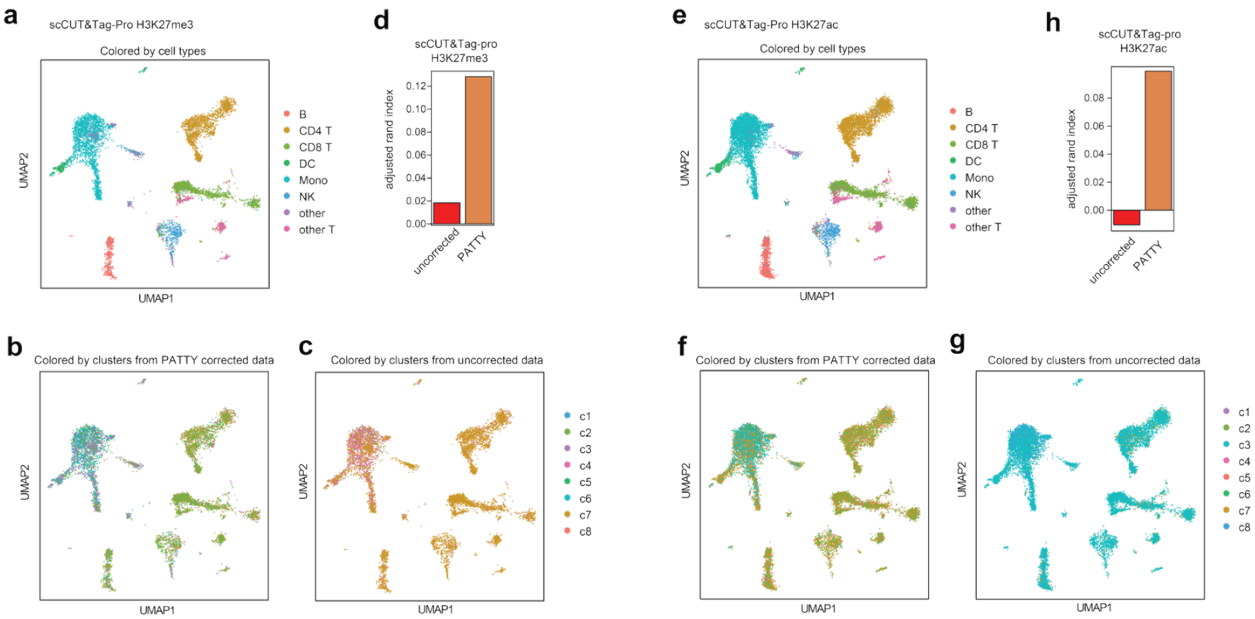

Figure S6. PATTY improves cell clustering of scCUT&Tag-pro. (**a**-**c**, **e**-**g**) UMAP visualization of H3K27me3 (**a**-**c**) and H3K27ac (**e**-**g**) scCUT&Tag-pro single cell data with cells colored by published cell type annotation as ground truth (**a**, **e**), cluster label from the PATTY corrected data (**b**, **f**), and cluster label from the raw count data (**c**, **g**). (**d**, **h**) Adjusted rand index between the clustering results and the ground truth cell type annotation for H3K27me3 (**d**) and H3K27ac (**h**) scCUT&Tag-pro data.

Supplementary Tables

Each supplementary table is a separate data sheet in the excel file.

Table S1. Metadata information for the datasets used in this study.

Table S2. The percentage of CUT&Tag peaks that are unique to CUT&Tag and not overlapped with peaks detected by ChIP-seq in the same cell line (K562). Rows are for different biological samples/datasets, and the last row is the average.

Table S3. List of true and false H3K27me3-marked regions for CUT&Tag in K562. The true and false labels are in the last column.

Table S4. Parameter tuning results for the deep neuro network models used for H3K27me3 model building. Each row represents the best hyperparameter settings for each model.

Table S5. List of true and false H3K27ac-marked regions for CUT&Tag in K562. The true and false labels are in the last column.

Table S6. List of true and false H3K9me3-marked regions for CUT&Tag in K562. The true and false labels are in the last column.