Expanded View Figures

Figure EV1. The alternative NURF complex is required for the proliferation of acute myeloid leukemia cells.

A Overview of the CRISPRi epi-library composition.

B Scatterplot indicating log2 normalized read counts for each sgRNA in MOLM-13 cells at day 0 and day 12 of the experiment. The orange and blue dashed lines indicate 2- and 4-fold depletion in the screen, respectively. The screen was performed in two biological replicates, i.e., independent cell transductions (n = 6,184).

C Log2 fold changes for all the sgRNAs against BPTF in the screen of the MOLM-13 cells. "p1@" and "p2@" in the sgRNA name indicate that it targets either the first (p1) or second (p2) most frequently used promoter for this gene according to the FANTOM5 consortium data. The data are the mean of two biological replicates, i.e., independent cell transductions.

D Competition assay after *BPTF* KO in MOLM-13, MV4-11, SET2, U937, and OCI-AML2 wtCas9 cells. The cells were transduced with the lentiviral cassettes expressing the corresponding sgRNAs and BFP and mixed with untransduced cells. The percentage of BFP-positive cells was measured over time. The data were normalized to Day 3 for MV4-11 and SET2 cells, and to Day 0 for MOLM-13, U937 and OCI-AML2 cells. The experiment was performed at least twice with similar results.

E qRT-PCR analysis for *SMARCA5* expression in AML cell lines, values are normalized to *RPLP0* and shown as mean (n = 3 technical replicates, i.e., independent qRT-PCR reactions). The experiment was performed at least twice with similar results in a selection of cell lines.

F Western blot analysis of BPTF and SMARCA5 after the BPTF immunoprecipitation (IP) in U937, SET2, and OCI-AML2 cells.

G Competition assay after *SMARCA5* KO in MOLM-13, MV4-11, SET2, U937, and OCI-AML2 wtCas9 cells. The cells were transduced with the lentiviral cassettes expressing the corresponding sgRNAs and BFP and mixed with untransduced cells. The percentage of BFP-positive cells was measured over time. The data were normalized to Day 3 for MV4-11 and SET2 cells and to Day 0 for MOLM-13, U937, and OCI-AML2 cells. The experiment was performed at least twice with similar results.

H Correlation between the BPTF and SMARCA5 Chronos dependency scores across all the cancer cell lines in the DepMap portal (version 22Q2) (n = 1,086). The correlation statistics were calculated using the Pearson's product–moment correlation.

I Chronos scores for BAP18 in all the lineages and leukemic cell lines. In the box plot, the middle line shows the median. The lower and upper hinges represent the first and third quartiles. The upper whisker stretches from the hinge to the largest value, but only up to 1.5 times the interquartile range. The lower whisker extends from the hinge to the smallest value, but only up to 1.5 times the interquartile range. Any data beyond the whiskers are plotted separately. The dashed green line indicates the essentiality threshold defined by the DepMap (version 22Q2). Welch two sample t-test was used to assess the statistical significance. $P < 0.0001$. n = 970 for "all lineages" and n = 116 for "leukemia".

J qRT-PCR analysis for *SMARCA5* expression in THP-1 cells overexpressing wild-type (wt) or catalytic dead (cd) SMARCA5 (n = 3 technical replicates, i.e., independent qRT-PCR reactions). Data are normalized to *RPLP0* and shown as mean.

Source data are available online for this figure.

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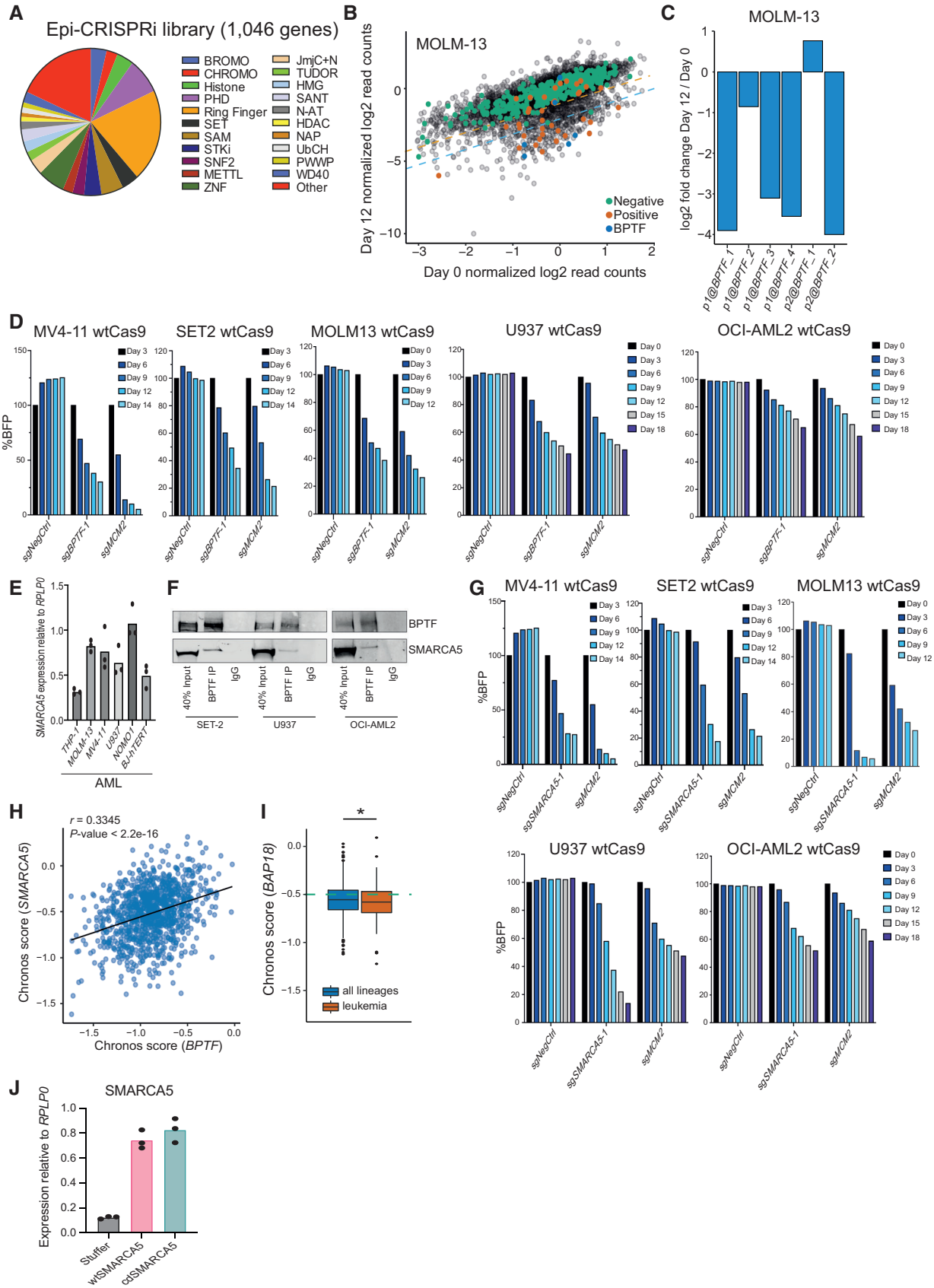

Figure EV1.

Figure EV2. BPTF and SMARCA5 sustain the expression of MYC and MYC-regulated genes.

A Flow cytometry histogram depicting CD11b levels in three cell lines: SET2, U937, and THP-1.
B The median fluorescent intensity of CD11b in THP-1, SET2, and U937 cells after knocking out either *BPTF* or *SMARCA5*. The levels were normalized to the cells that were transduced with the non-targeting sgRNA.
C Enrichment plots for MYC target genes among the downregulated genes after *BPTF* KO (GSEA categories: SCHLOSSER_MYC_TARGETS_AND_SERUM_RESPONSE_UP and ACOSTA_PROLIFERATION_INDEPENDENT_MYC_TARGETS_UP).
D Western blotting analysis of MYC protein levels (left) and their quantification (right) after *BPTF*, *SMARCA1* and *SMARCA5* KOs.
E Schematic representation of genotyping primers used to verify the successful knock-in.
F Agarose gel image after the PCR using the genotyping primers represented in (E) on either wild-type THP-1 cells or the SMARCA5 degron knock-in cells.
G KO efficiency graph generated using TIDE for the untargeted *SMARCA5* locus in the individual clones generated from the SMARCA5 degron knock-in cell pool.
H Western blotting analysis of SMARCA5 protein expression in wild-type and SMARCA5 degron knock-in cells upon treatment with dTAG-V1 (100 nM). WT stands for wild-type and KI—for knock-in.
I qRT-PCR analysis for the expression of *MYC* in THP-1 cells transduced with a non-targeting sgRNA or three different mixes of two *MYC* KD sgRNAs, as downregulation of MYC was insufficient using only one sgRNA, $n = 3$ biological replicates (independent cell transductions) for negative control and $n = 2$ biological replicates (independent cell transductions) for the remaining samples. Data are normalized to *RPLP0* and shown as mean ± SD (one-way ANOVA followed by Dunnett's multiple comparisons test was performed to assess significance, $^*P < 0.05$). The experiment was performed twice with similar results.
J Cell cycle analysis of the THP-1 cells transduced with a nontargeting sgRNA or *MYC* KD sgRNAs ($n = 3$ biological replicates, i.e., independent cell transductions). Data are shown as mean ± SD.
K Western blot for MYC and beta-actin in wild-type THP-1 cells and THP-1 cells with MYC overexpression.
L Competition assay after *BPTF* or *SMARCA5* KO in wild-type or MYC-overexpressing THP-1 cells. The cells were transduced with the lentiviral cassettes expressing the corresponding sgRNAs and BFP and mixed with untransduced cells. The percentage of BFP-positive cells was measured over time. The data were normalized to Day 1.

Source data are available online for this figure.

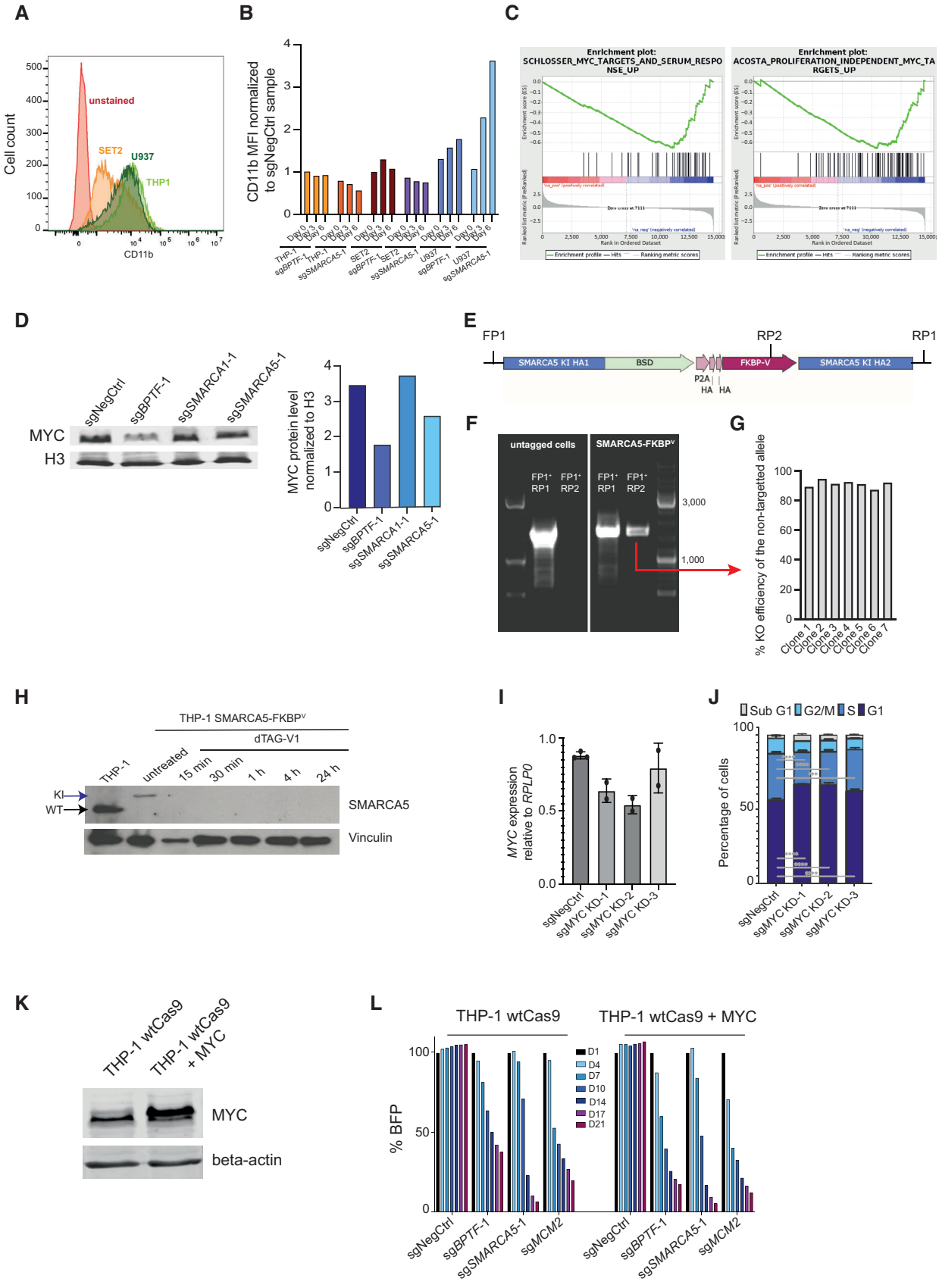

Figure EV2.

Figure EV3. BPTF and SMARCA5 bind active promoters and insulators in AML cells.

A Venn diagram of the overlap between the BPTF and HA-SMARCA5 peaks. Each peak was extended by 500 bps on each side to estimate this overlap. 701 common regions are expected by chance.

B Heatmap of the BPTF and SMARCA5 Cut&Run signal across all the insulators, active promoters, and enhancers predicted by the ChromHMM in K562 cells.

C Heatmap displaying the relative percentage of the genome represented by each state (first column) and relative fold enrichment for: CpG islands, RefSeq exons, genes, transcription end sites (TES), transcription start sites (TSS), and 2,000 base pair regions around the TSS. The darker the blue color, the higher the fold enrichment.

D Bar plots illustrating the percentage of BPTF Cut&Run peaks overlapping different chromatin domain categories defined by the ChromHMM in THP-1 cells. Some peaks may overlap several categories; hence, the total percentage does not equal 100. "trx" stands for transcription.

E, F Heatmaps of the CTCF, BPTF, and SMARCA5 Cut&Run signal across all the CTCF peaks in U937 (E) and OCI-AML2 (F) cells.

G, H Average profiles of the CTCF, BPTF, and SMARCA5 Cut&Run signal in U937 (G) and OCI-AML2 (H) cells at the insulator regions predicted by the ChromHMM in K562 cells.

I, J Examples of the common CTCF, SMARCA5, and BPTF insulator peaks in U937 (I) and OCI-AML2 (J) cells.

Data information: All the Cut&Run experiments in THP-1 cells were performed in two biological replicates, i.e., independent cell transductions or treatments.
Source data are available online for this figure.

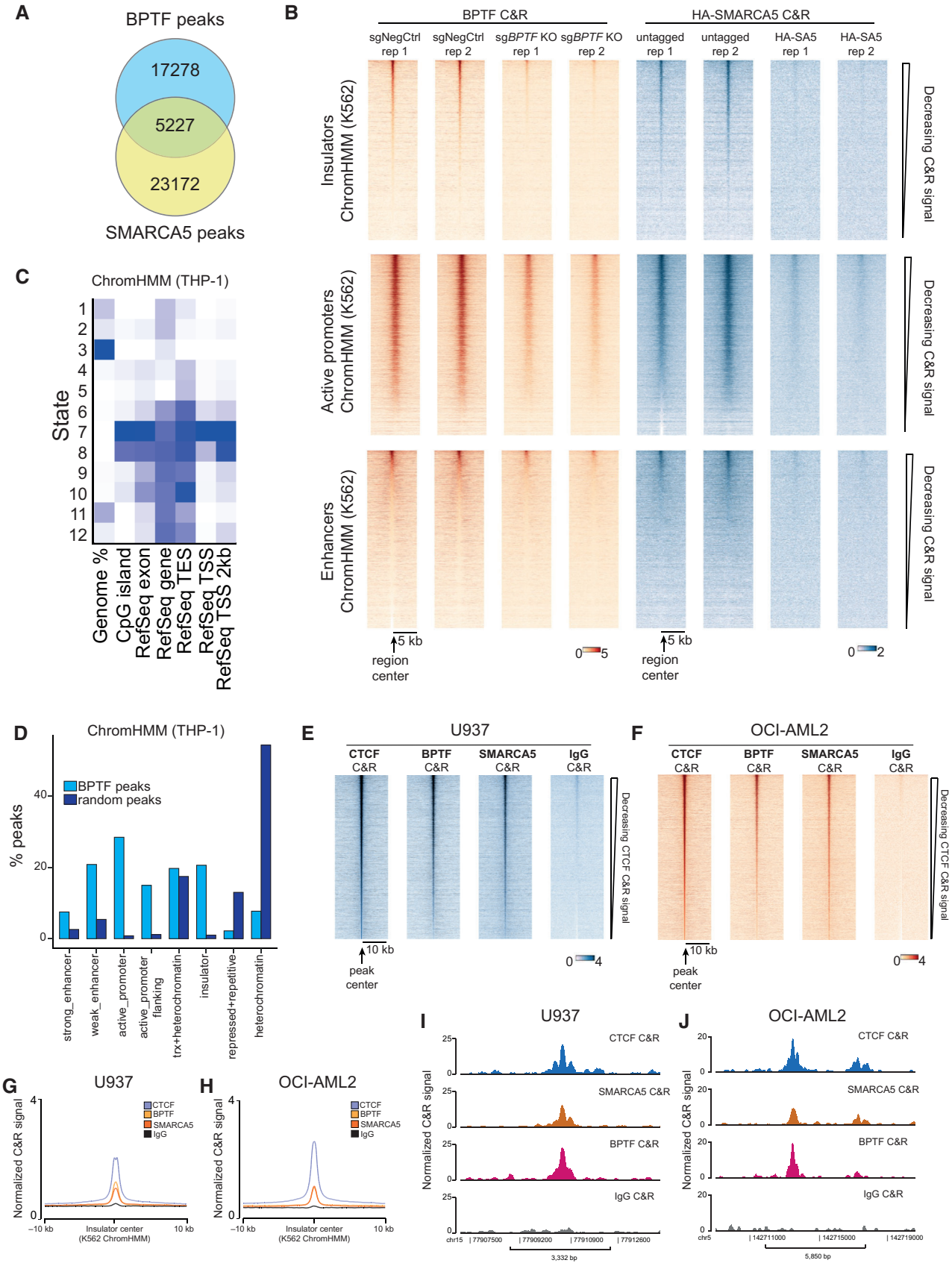

Figure EV3.

Figure EV4. The NURF complex facilitates CTCF binding and chromatin insulation.

A, B Average profile of CTCF binding at differential ATAC-seq clusters in two independent experiments.
C, D Average insulation score profiles across all the boundaries gained upon *BPTF* (C) and *SMARCA5* (D) KO.
E Average insulation score profile across the differential ATAC-seq cluster 6 regions with CTCF motifs.
F WashU Browser score at the indicated interaction points.
G Hi-C data snapshots at 25 kb resolution from negative control, *BPTF* and *SMARCA5* KO samples. Anchor points for the indicated loop (arrow) at the *HOXA* cluster are highlighted in red and blue.

Data information: The CTCF Cut&Run experiment was performed three times with similar results, each in two biological replicates. The HiC experiment was performed in two biological replicates that were pooled together at the analysis stage to increase sequencing depth.
Source data are available online for this figure.

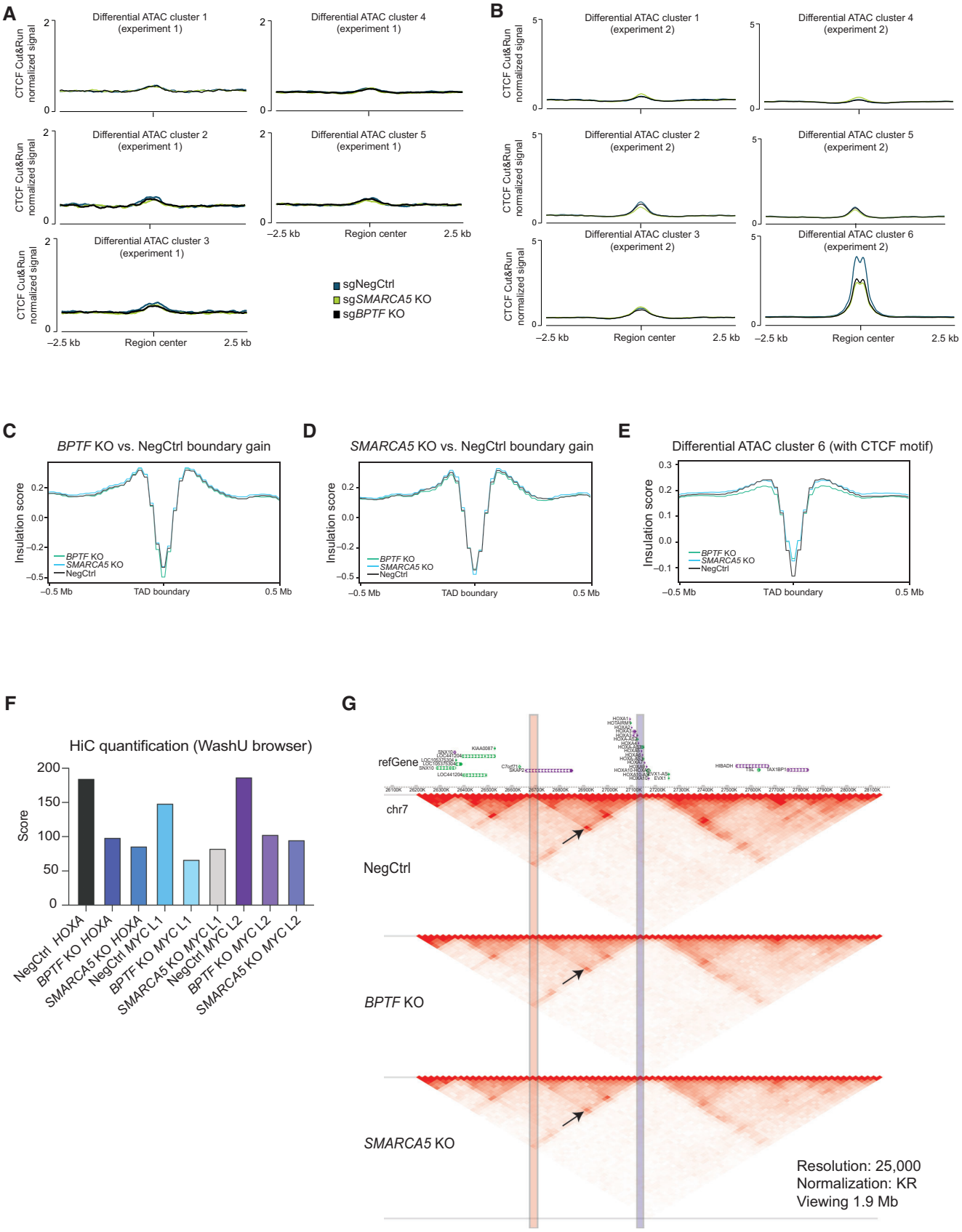

Figure EV4.

Figure EV5. Tandem PHD2 and BROMO reader domains of BPTF are not required for the proliferation of the AML cells.

A Competition assay after targeting *Bptf* with different sgRNAs in mouse MLL-AF9 wtCas9 cells. The cells were transduced with the lentiviral cassettes expressing the corresponding sgRNAs and GFP and mixed with untransduced cells. The percentage of GFP-positive cells was measured over time. The data were normalized to Day 0.

B Indel analysis of the mutations induced by the sgRNAs in the selected BPTF truncation clones.

C, D BPTF expression at the mRNA ($n = 3$ technical replicates, i.e., independent qRT-PCR reactions; the data are normalized to *RPLP0* and presented as mean) (C) and protein (D) levels in the wild-type and BPTF truncation clones.

E BPTF binding in wild-type and BPTF truncation clones across all the active promoter regions defined by the ChromHMM in K562 cells. The Cut&Run was performed in two biological replicates.

F BPTF binding in wild-type and BPTF truncation clones at a representative promoter region.

G Relative cell viability of the THP-1 cells after treatment with increasing concentrations of either a BPTF BROMO domain inhibitor or a structurally related negative control compound. Data are shown as mean ± SD, $n = 3$ biological replicates, i.e., independent cell treatments.

H Box plot indicating SMARCA5 DepMap KO score in cancer cell lines expressing and not expressing SMARCA1. In the box plot, the middle line shows the median. The lower and upper hinges represent the first and third quartiles. The upper whisker stretches from the hinge to the largest value, but only up to 1.5 times the interquartile range. The lower whisker extends from the hinge to the smallest value, but only up to 1.5 times the interquartile range. Any data beyond the whiskers are plotted separately. The dashed line indicates the essentiality threshold defined by the DepMap (version 22Q2). $P < 0.0001$ based on Wilcoxon rank sum test with continuity correction. $n = 833$ for "expressed" and $n = 172$ for "not expressed".

Source data are available online for this figure.

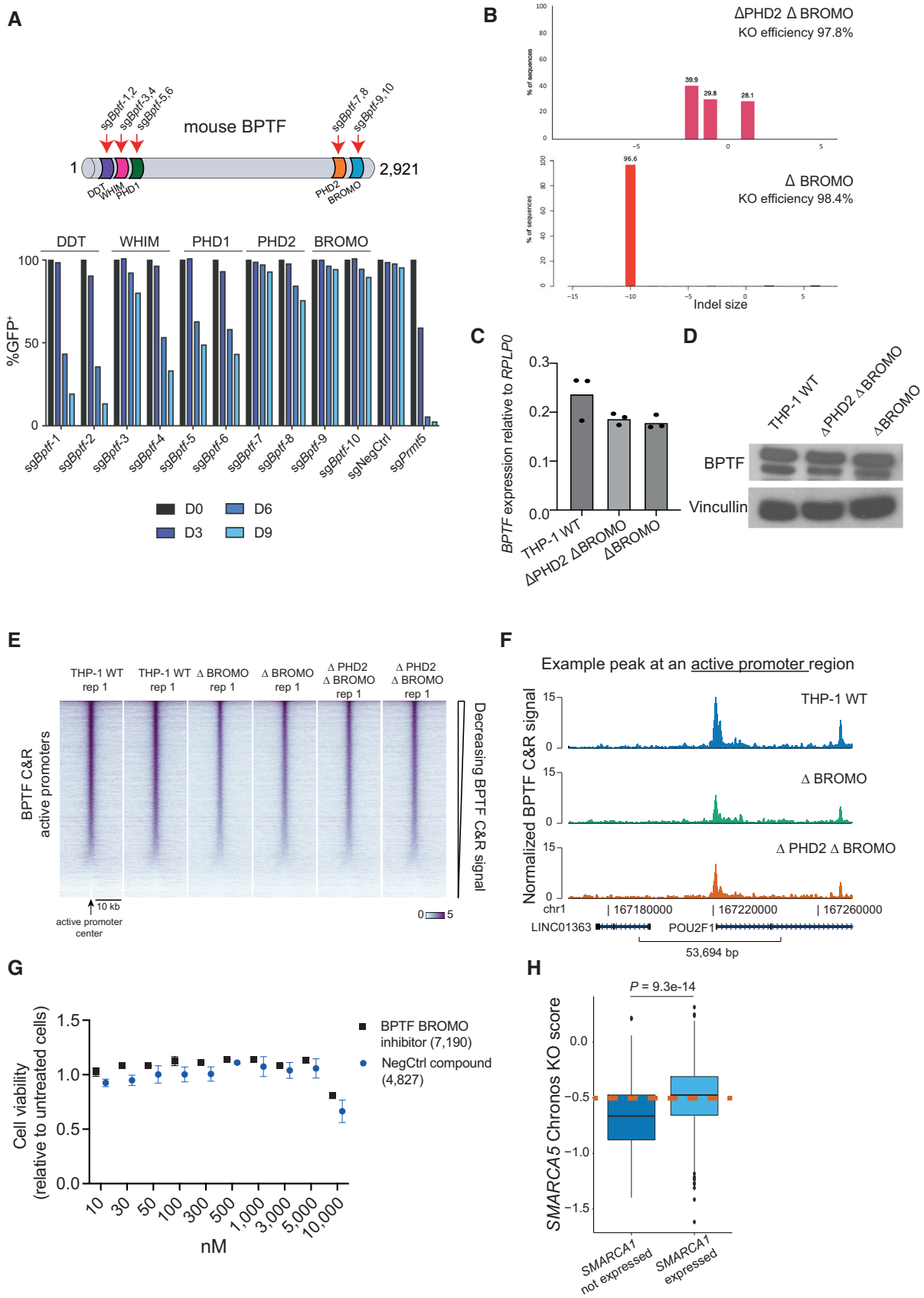

Figure EV5.