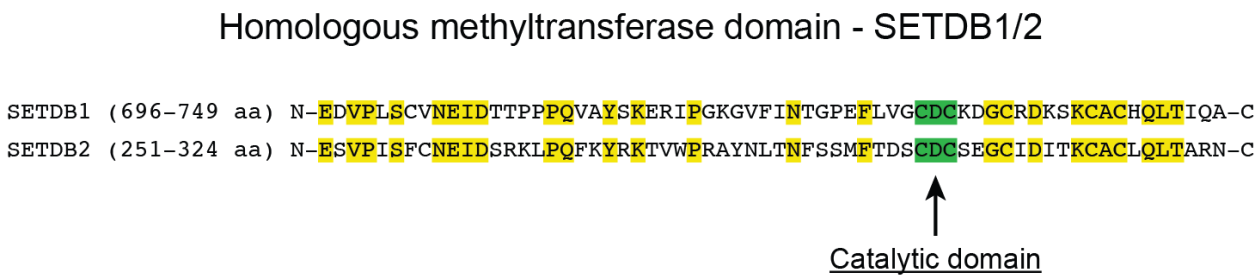


Fig. S1.

Diagram of catalytic domain homology between SETDB1 and SETDB2. Homologous regions in SETDB1 and SETDB2 were aligned against each other. The region highlighted in yellow denotes identical amino acids, while the region highlighted in green shows the amino acids that were replaced to generate the mutated variant (C293L/C295P).

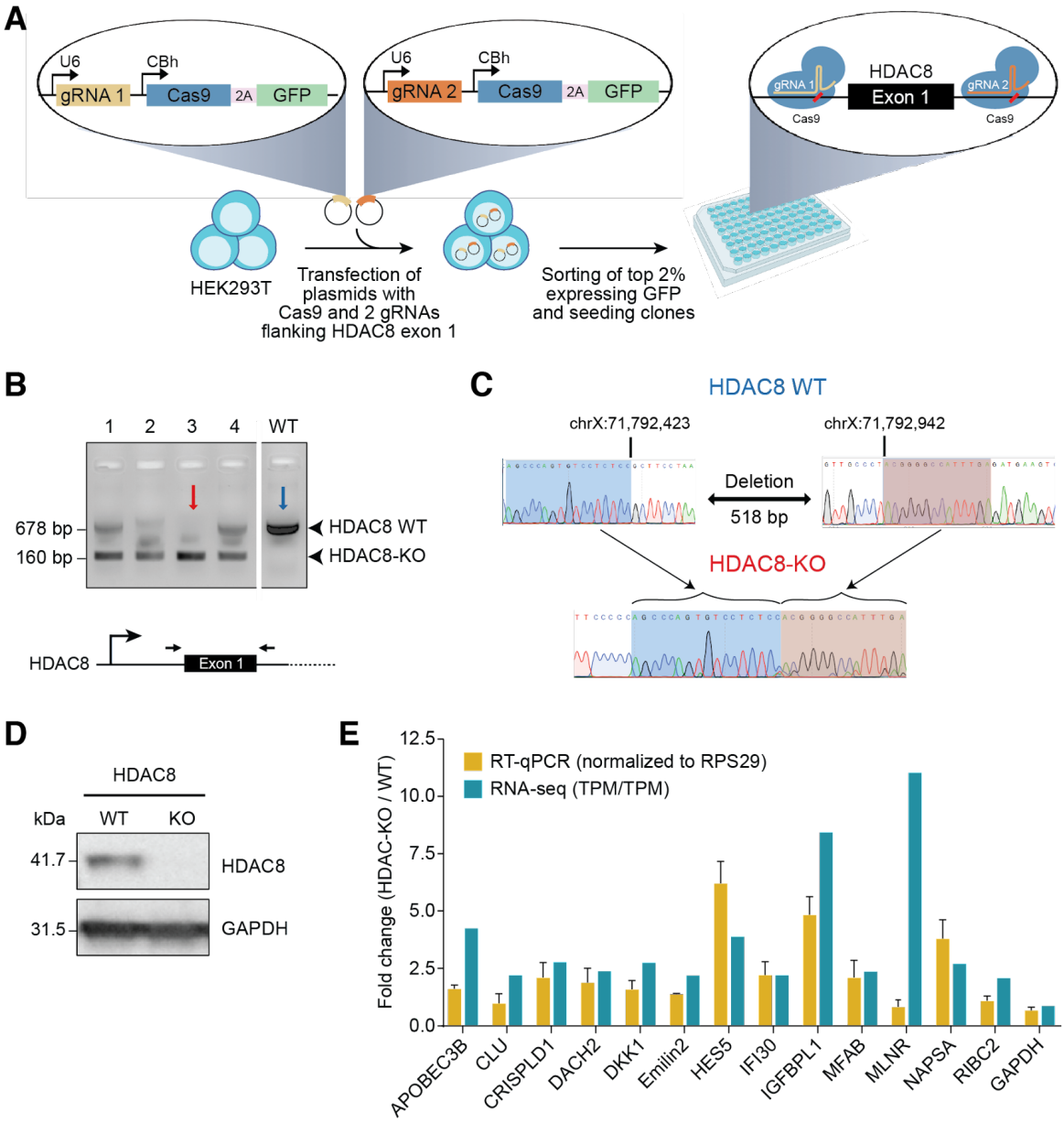


Fig. S2. Generation of a HDAC8 knock-out cell-line to identify potential responsive target genes for our dCas9-GEMs. (A) A workflow for the HDAC8 knock-out generation. Cas9-2A-GFP expressing plasmids that include gRNA cassettes flanking the first exon of HDAC8 were co-transfected in HEK293T cells, and top 2% GFP-expressing clones were further analyzed to generate a HDAC8 knock-out. (B) Gel electrophoresis of HDAC8-KO clones 1 to 4. Genomic DNA was amplified with PCR primers surrounding exon 1 of HDAC8, visibly showing

HDAC8 deletion band. Red arrow: the colony HDAC8-KO #3 shows a homozygous deletion band of HDAC8. Blue arrow: wt band of HDAC8. (C) Sanger sequencing results of wildtype HDAC8 band that was extracted from wildtype HEK293T, compared with HDAC8 deletion band that was extracted from HDAC8-KO #3, demonstrating homozygous removal of the first exon of HDAC8. (D) Validation of HDAC8 knockout by Western blot. Wildtype HEK293T and HDAC8-KO were stained for HDAC8 demonstrating successful deletion of HDAC8. (E) Change in expression of candidate genes for CRISPR-GEM targeting (as fold-change expression of HDAC8-KO relative to wildtype HEK293T). RT-qPCR results (normalized to RPS29; n=2 experiments) are compared to RNA-seq (as TPM levels in HDAC8-KO divided by TPM levels in wildtype HEK293T ratio; n=1).

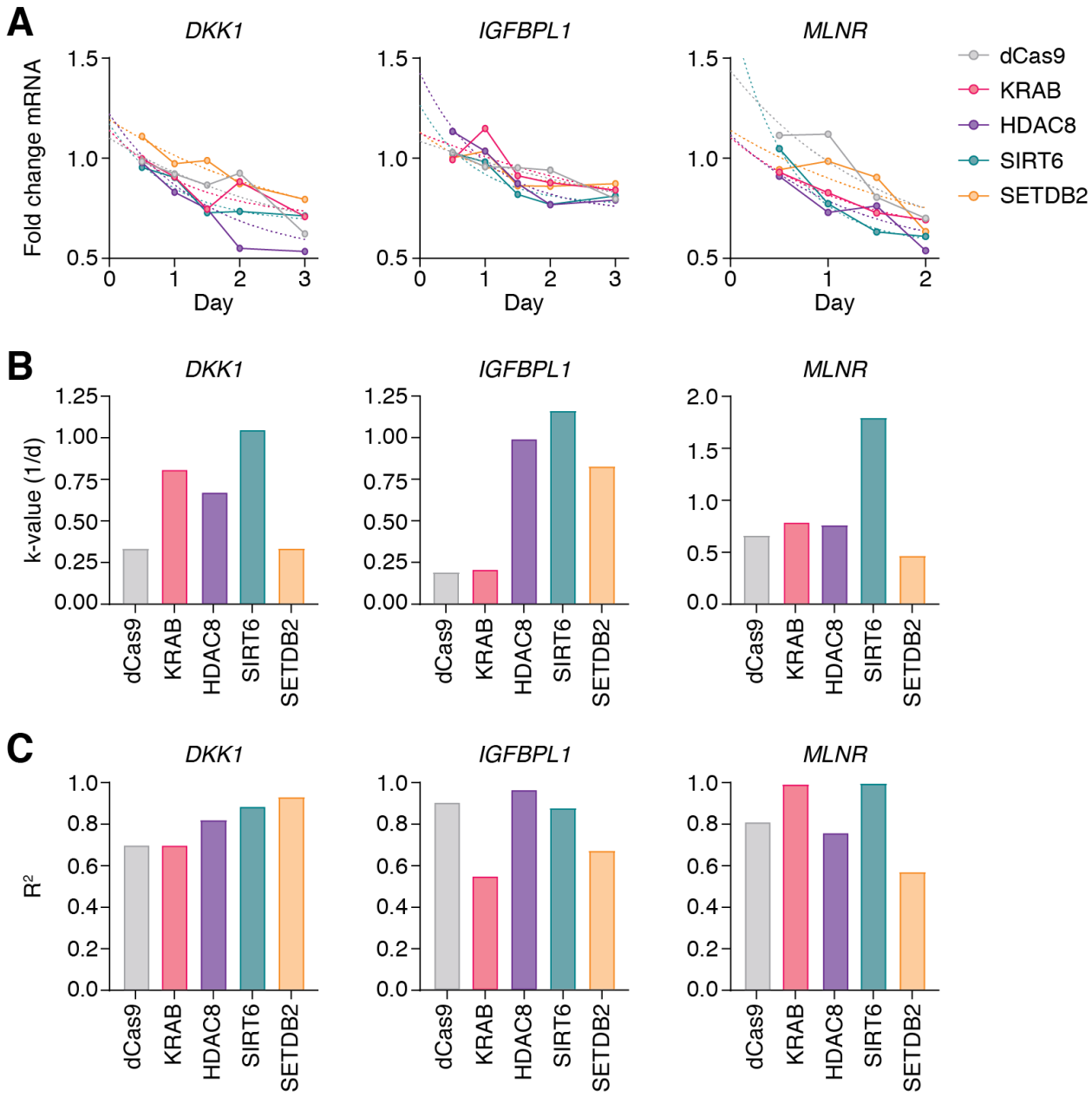
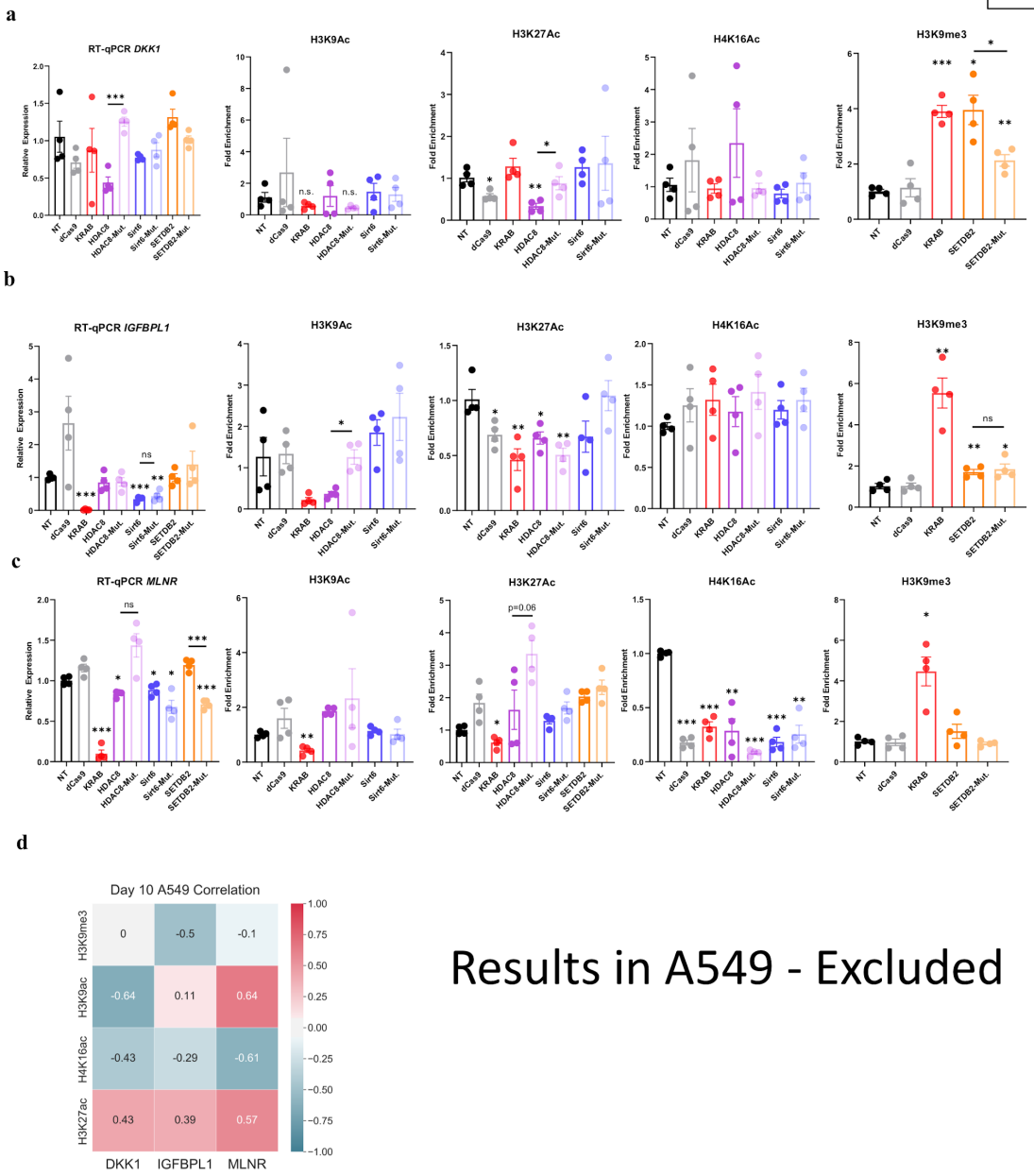


Fig. S3.

(A) Fitting of first-order logarithmic kinetic model for gene repression by CRISPR-GEMs. Fitting of a model for every gene was based on measured mRNA levels on days 0.5, 1,1.5, 2 and 3. (B) Calculated K_{exp}^{*} values for each fitted model, denoting rate of mRNA expression during the observed time period. **c.** R^2 values of each fitted model, demonstrating how well the model fits the recorded data.

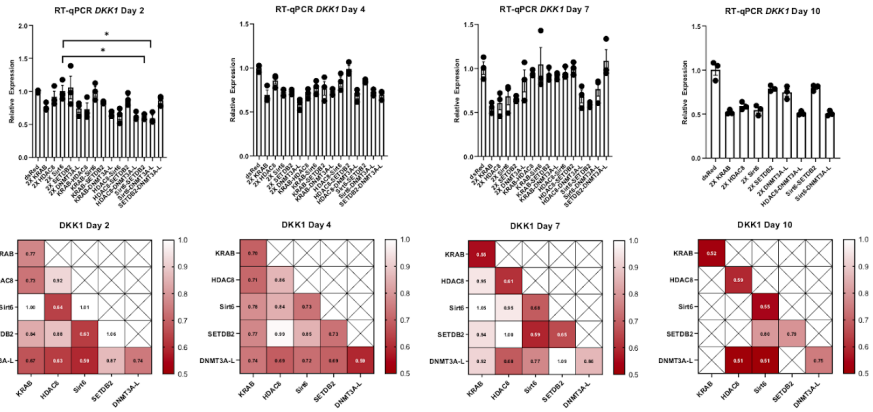
Figure depicting A549 constitutively expressing gRNAs and CRISPR-GEMs with lentiviral transduction. qRT-PCR and ChIP-qPCR at Day 15 N=4



d

Results in A549 - Excluded

a



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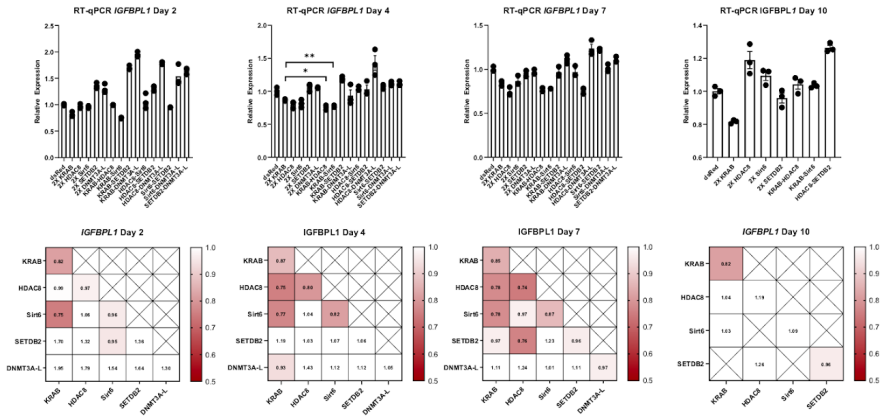
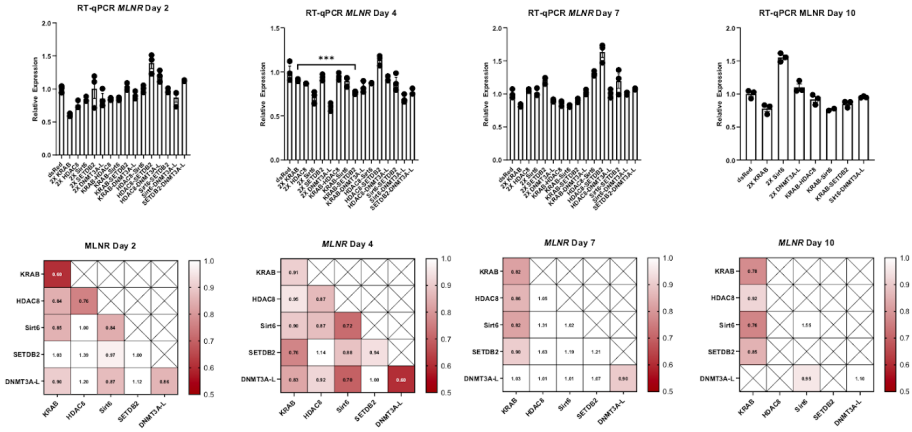


c



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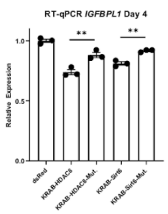

Combination data - excluded