Programmable m^{6}A modification of cellular RNA with a Cas13-directed methyltransferase

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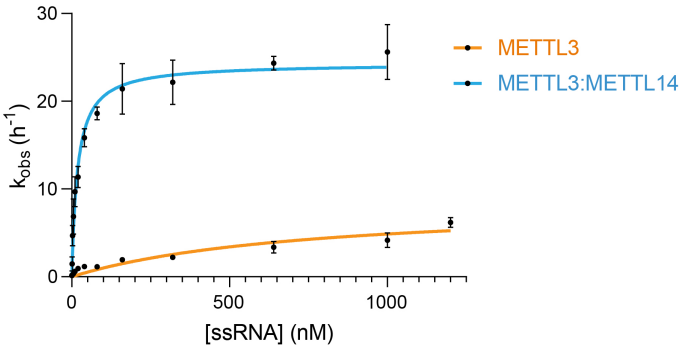
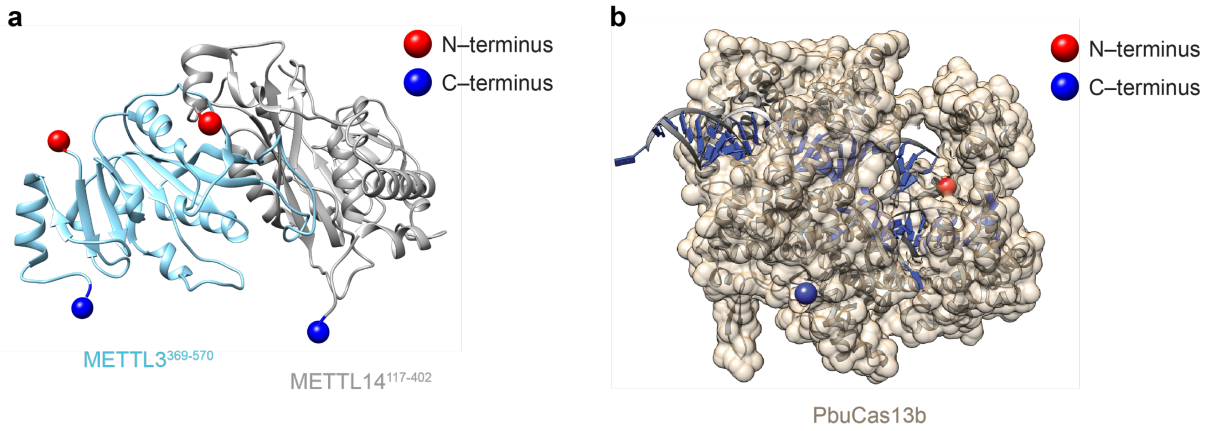
SUPPLEMENTARY INFORMATION

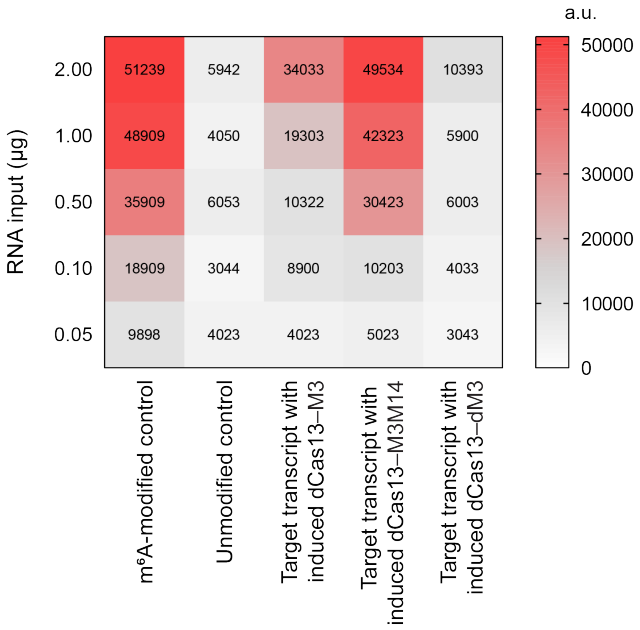
Construct	K_m (nM)	V_{max} (h^{-1})
METTL3	> 900	8.6 ± 1.11
METTL3:METTL14	18 ± 1.76	24 ± 0.50



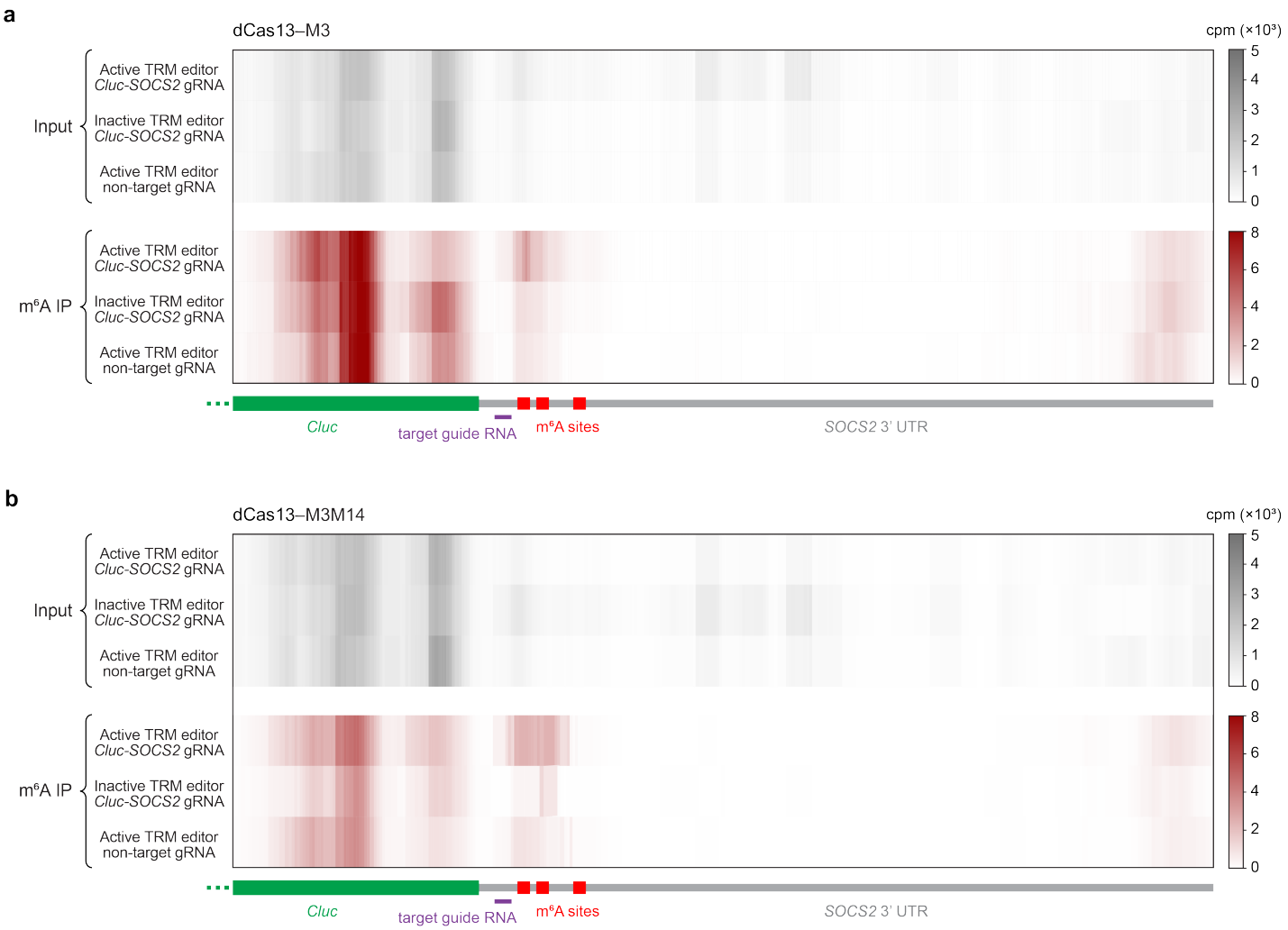
Supplementary Figure 1*. In vitro* kinetics of m^{6}A methyltransferases. Michaelis-Menten kinetics of purified METTL3 and METTL3:METTL14 activity on a ssRNA substrate. k_{obs} = observed catalytic rate. Values reflect the mean±s.e.m. of n=3 independent reactions.



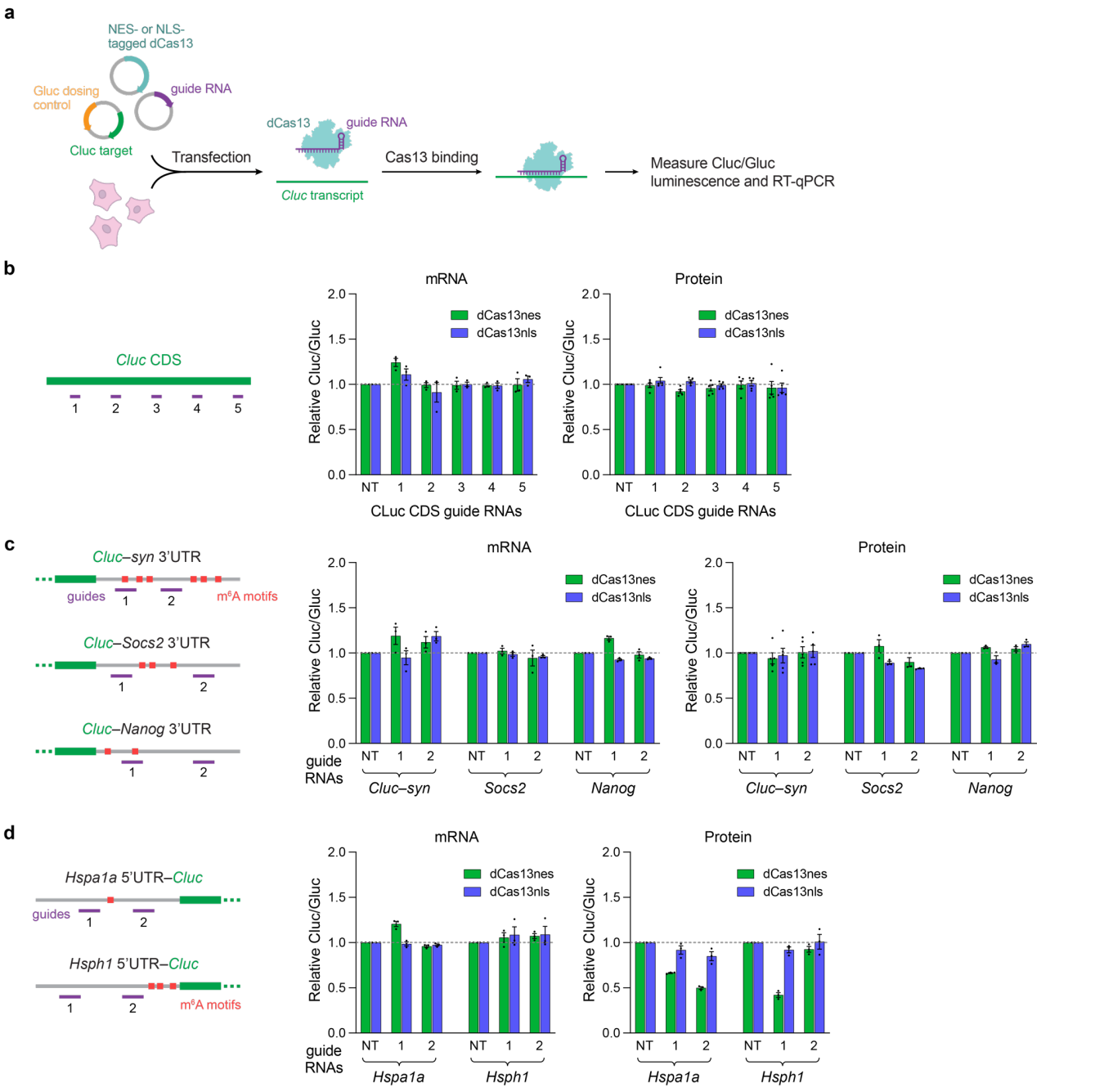
Supplementary Figure 2. Structural analysis of METTL3:METTL14 and PbuCas13b. (a) Crystal structure of the METTL3$^{369-570}$:METTL14$^{117-402}$ heterodimer (PDB 5IL1)[1]. **(c)** Co-crystal structure of *Prevotella buccae* Cas13b (PbuCas13b, brown) bound to the CRISPR RNA hairpin of a guide RNA (PDB 6DTD, grey and blue)[2]. The N-terminus (red) and C-terminus (blue) are shown for each protein.



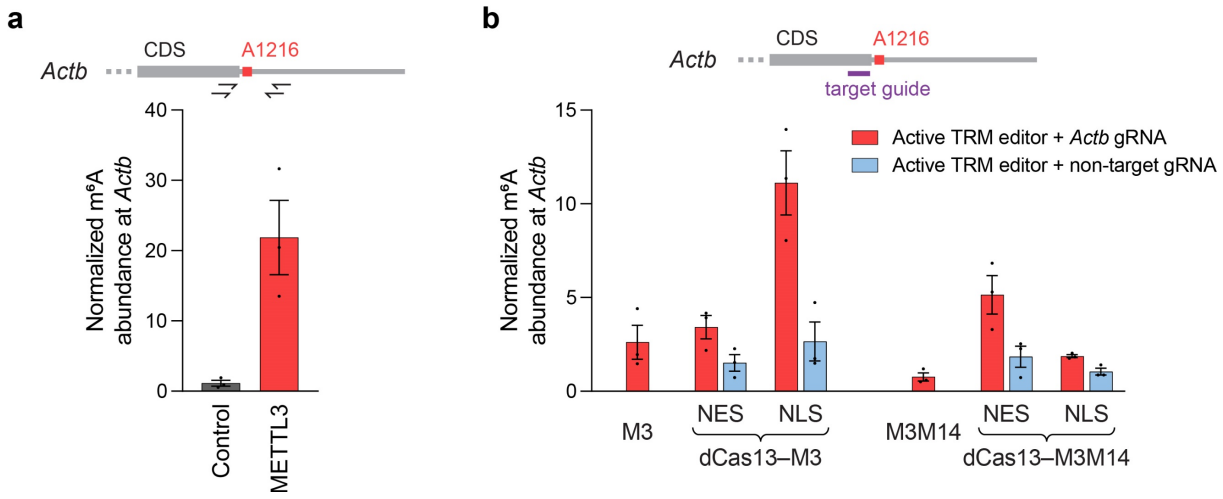
Supplementary Figure 3. Orthogonal assay for on-target RNA methylation by TRM editors in *E. coli*. Fluorescence measurements of indicated quantities of RNA captured and stained with fluorescent anti-m^{6}A antibodies to assay relative levels of m^{6}A modification. Columns 1-2: m^{6}A-modified control RNA and unmodified control RNA were commercially synthesized and served as positive and negative controls. Columns 3-5: synthetic transcript was targeted with indicated TRM editors within *E. coli*, purified from cells, selectively enriched, then quantified for m^{6}A abundance. dCas13–dM3 represents a methyltransferase-inactive dCas13–M3^{D395} control. a.u. = fluorescence in arbitrary units.



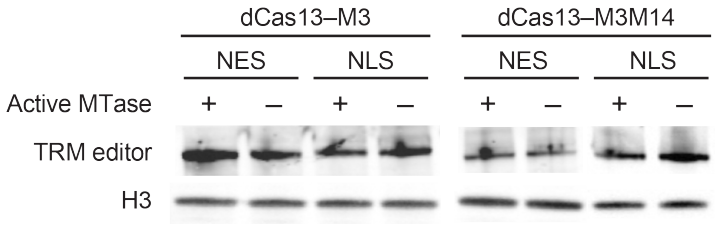
Supplementary Figure 4. MeRIP-seq of Cluc–*Socs2* targeted by TRM editors. *Cluc*–*Socs2* read coverage of input (grey) and m^{6}A-immunoprecipitated (red, m^{6}A IP) RNA from MeRIP-seq of transfected HEK293T cells. TRM constructs **(a)** dCas13–M3 and **(b)** dCas13–M3M14 targeted the *Socs2* 3' UTR under the following conditions: active editor with a target guide RNA, inactive editor with a target guide RNA, and active editor with a non-target guide RNA. Inactive TRM editors contain a methyltransferase-inactivating D395A mutation within M3. The target guide RNA (purple) and targeted m^{6}A sites are shown. Cpm = counts per million reads. MeRIP-seq analysis was performed with n=3 independent biological replicates.



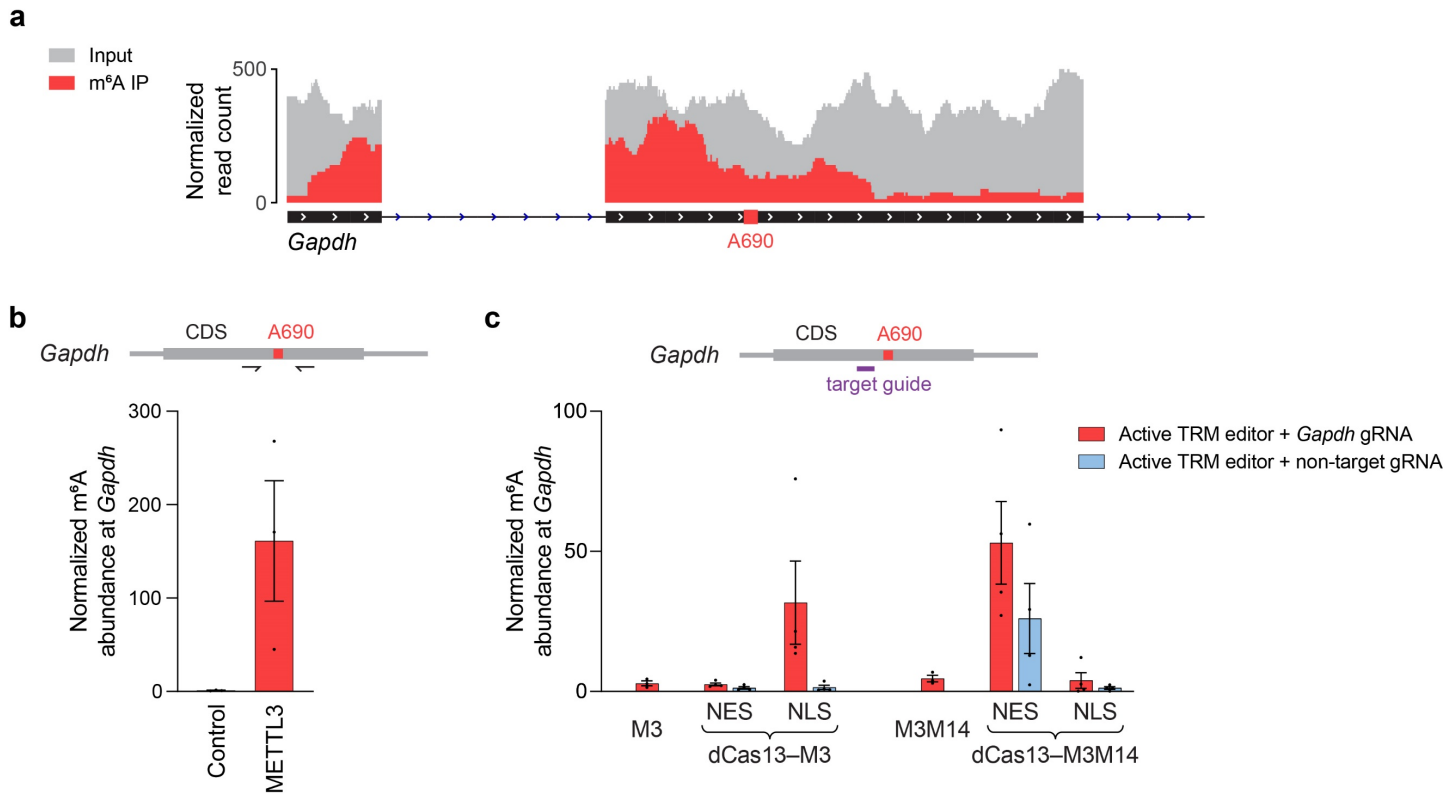
Supplementary Figure 5. Effect of dCas13 binding on transcript stability and translation. (a) Schematic of dCas13 targeting assay. Nucleus- (dCas13nes) or cytoplasm-localized dCas13 (dCas13nls), guide RNAs (purple), and a dual luciferase plasmid were transfected in HEK293T cells prior to measurement of luciferase RNA abundance and luminescence. *Cypridina* luciferase (*Cluc*, green) reporter transcripts were targeted by dCas13, while *Gaussia* luciferase (*Gluc*, orange) served as a dosing control for normalization. **(b)** dCas13 targeting *Cluc* with guide RNAs tiled across the Cluc coding sequence (CDS). **(c)** dCas13 targeting the 3' UTRs of *Cluc*–syn, and 3' UTRs of endogenous *Socs2 and Nanog* fused to a *Cluc* reporter. **(d)** dCas13 targeting the 5' UTRs of endogenous *Hspa1a and Hsph1* attached to a *Cluc* reporter. Indicated dCas13 guide RNAs (purple) were tiled across each reporter. NT = non-targeting guide RNA; NES = nuclear export signal; NLS = nuclear localization signal. For **(b-d)** mRNA and **(d)** protein, values and error bars reflect the mean±s.e.m. of n=3 independent biological replicates. For **(b-c)** protein, values and error bars reflect the mean±s.e.m. of n=5 independent biological replicates.



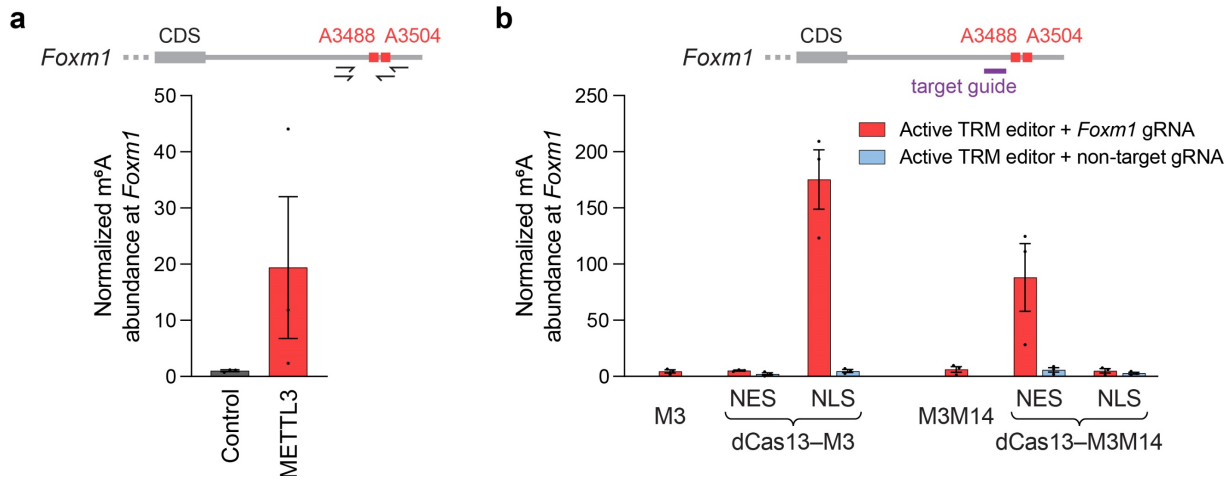
Supplementary Figure 6. Validation of targeted *Actb* methylation by TRM editors. (a) MeRIP-RT-qPCR of *Actb* A1216 from HEK293T cells transfected with a vector control or full-length METTL3. **(b)** MeRIP-RT-qPCR of indicated TRM editors or TRM methyltransferase domains with *Actb*-targeting or non-targeting guide RNAs. M3 and M3M14 refer to methyltransferase components lacking dCas13. Values and error bars reflect the mean±s.e.m. of n=3 independent biological replicates.



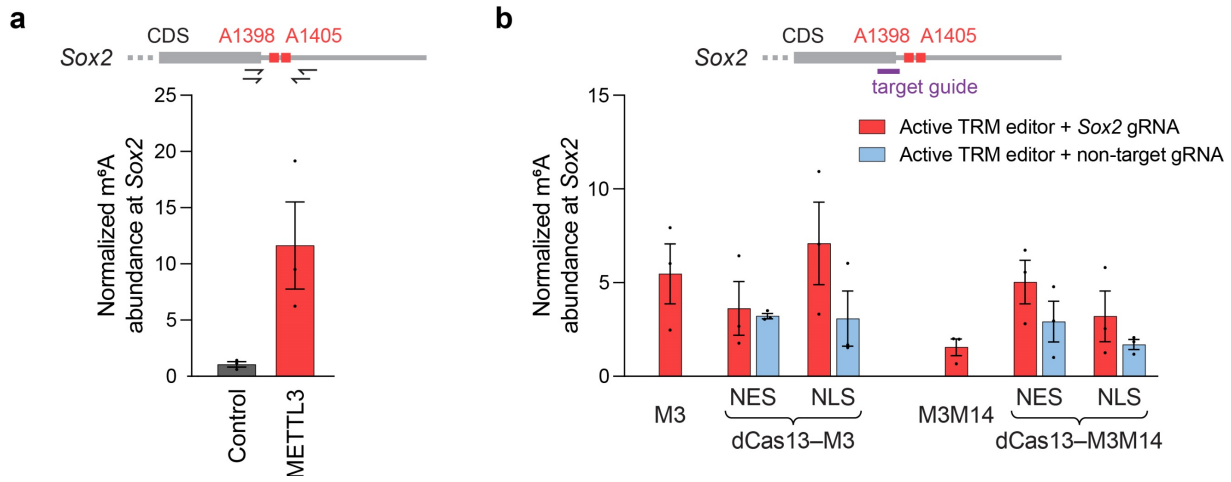
Supplementary Figure 7. Cellular protein expression of TRM editors. Representative western blots of 3×HA-tagged TRM editors compared with inactive TRM controls. HEK293T cells were co-transfected with non-targeting guide RNAs in all conditions. Editors were stained with an anti-HA antibody and histone H3 served as a loading control. Inactive TRM editors used methyltransferase-inactive $M3^{D395}$ instead of M3 and are indicated by active MTase (−). Every active and inactive pair from the same TRM construct comes from the same Western blot. Western blots were performed twice with similar results.



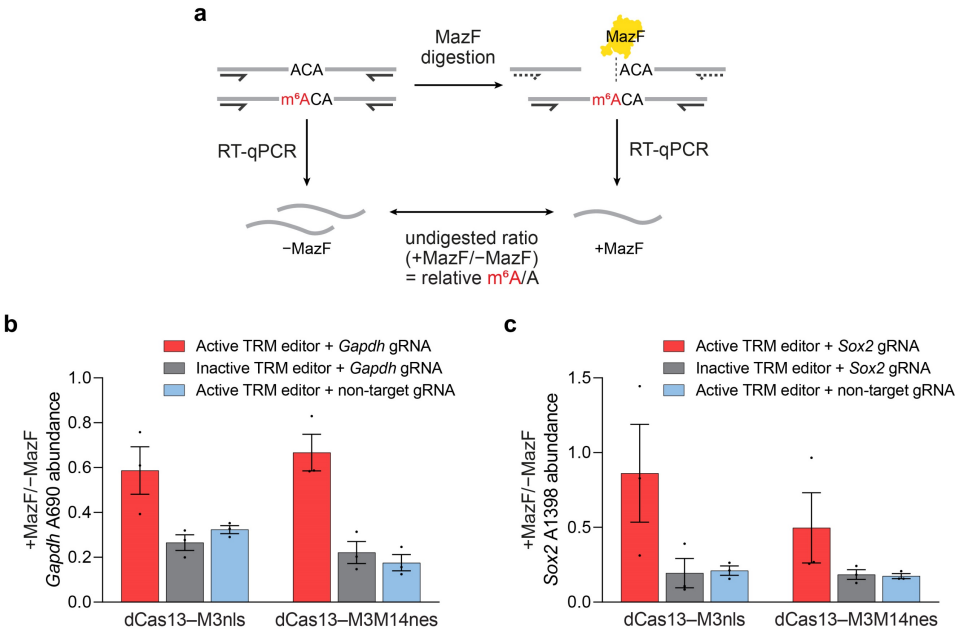
Supplementary Figure 8. Validation of targeted *Gapdh* methylation by TRM editors. (a) MeRIP-seq coverage of *Gapdh* mRNA from un-transfected HEK293T cells. **(b)** MeRIP-RT-qPCR of *Gapdh* A690 from HEK293T cells transfected with a control plasmid or full-length METTL3. **(c)** MeRIP-RT-qPCR of indicated TRM editors or TRM methyltransferase domains with *Gapdh*-targeting or non-targeting guide RNAs. M3 and M3M14 refer to methyltransferase components lacking dCas13. Values and error bars reflect the mean±s.e.m. of n=4 independent biological replicates.



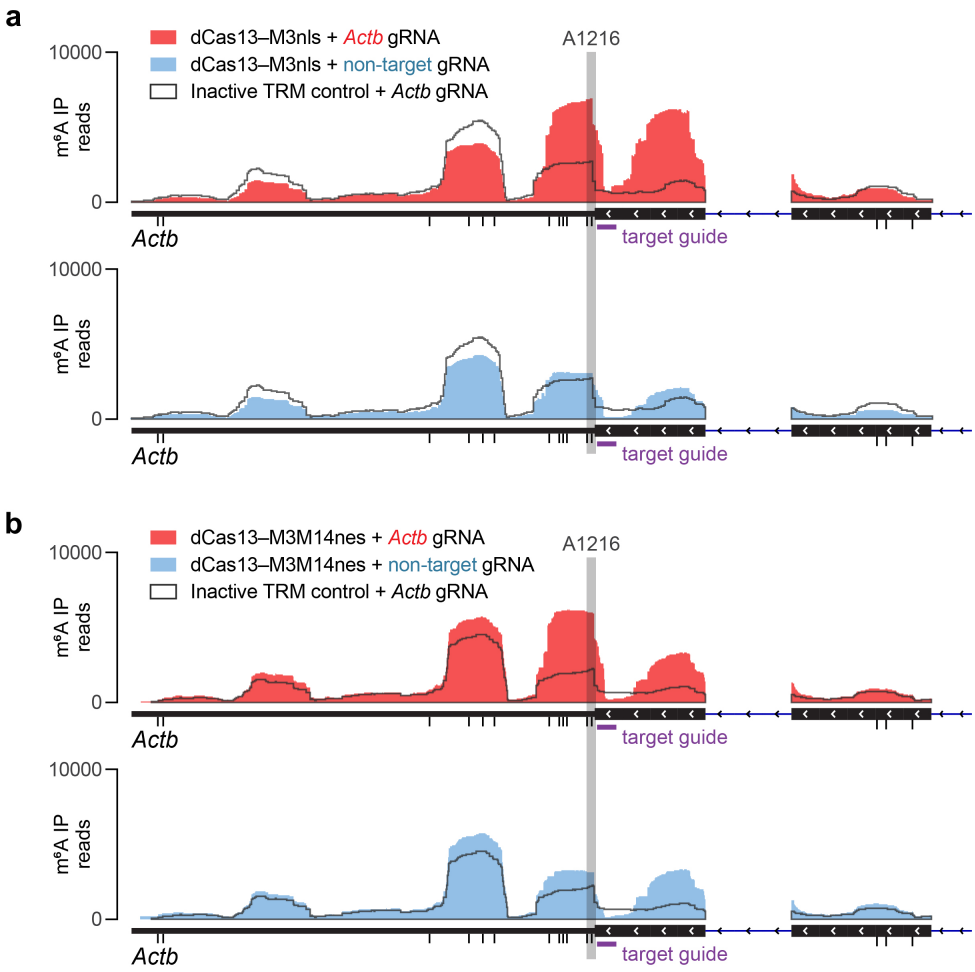
Supplementary Figure 9. Validation of targeted *Foxm1* methylation by TRM editors. (a) MeRIP-RT-qPCR of *Foxm1* 3'UTR m^{6}A sites (A3488 and A3504) from HEK293T cells transfected with a control plasmid or full-length METTL3. **(b)** MeRIP-RT-qPCR of indicated TRM editors or TRM methyltransferase domains with *Foxm1*-targeting or non-targeting guide RNAs. M3 and M3M14 refer to methyltransferase components lacking dCas13. Values and error bars reflect the mean±s.e.m. of n=3 independent biological replicates.



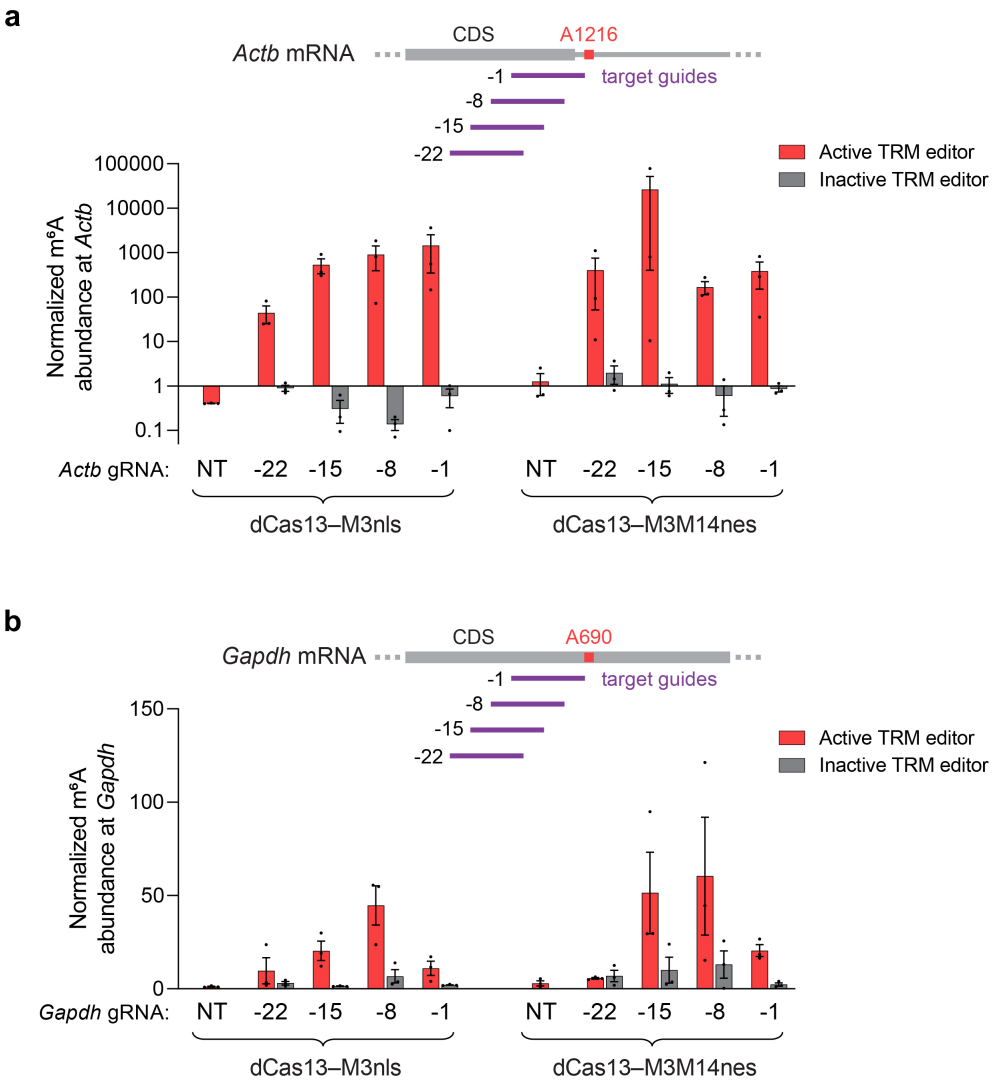
Supplementary Figure 10. Validation of targeted *Sox2* methylation by TRM editors. (a) MeRIP-RT-qPCR of *Sox2* 3'UTR m^{6}A sites (A1398 and A1405) from HEK293T cells transfected with a control plasmid or full-length METTL3. **(b)** MeRIP-RT-qPCR of indicated TRM editors or TRM methyltransferase domains with *Sox2*-targeting or non-targeting guide RNAs. M3 and M3M14 refer to methyltransferase components lacking dCas13. Values and error bars reflect the mean±s.e.m. of n=3 independent biological replicates.



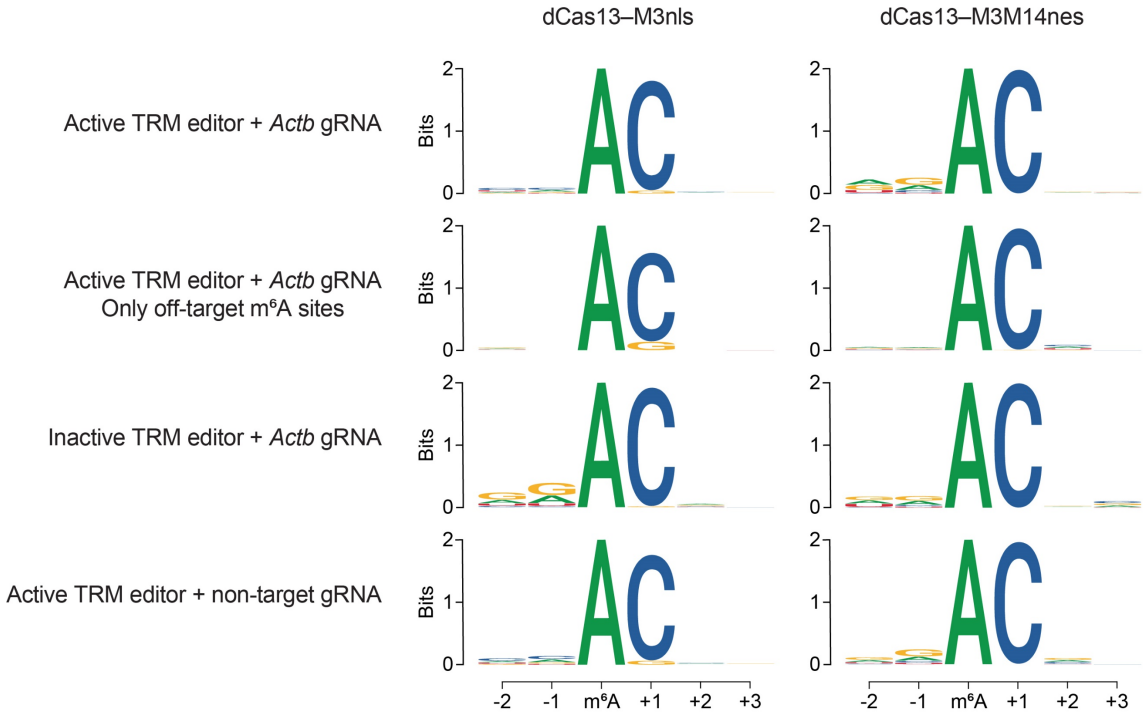
Supplementary Figure 11. Orthogonal validation of TRM with MazF-qPCR. (a) Schematic of MazF-qPCR method for base-resolution m^{6}A measurement. MazF ribonuclease specifically digests ACA but not m^{6}ACA, allowing m^{6}A detection within ACA motifs by measurement of MazF digestion efficiency. Cellular RNA samples are treated with (+MazF) or without MazF (−MazF), followed by RT-qPCR of a target amplicon spanning the interrogated ACA motif. +MazF/−MazF amplicon abundance provides single-base quantification of relative m^{6}A/A ratios at the targeted ACA motif. **(b)** MazF-qPCR of *Gapdh* A690. **(c)** MazF-qPCR of *Sox2* A1398. Inactive TRM editors contain a methyltransferase-inactivating D395A mutation within M3. Values and error bars reflect the mean±s.e.m. of n=3 independent biological replicates.



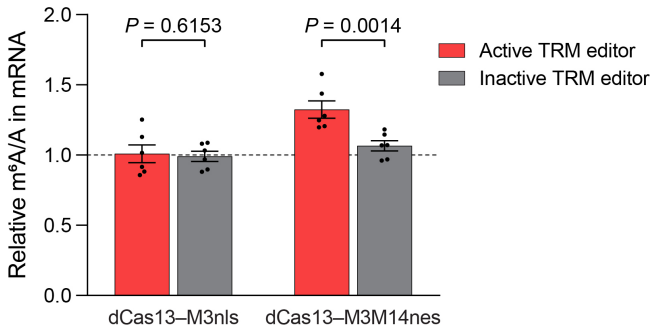
Supplementary Figure 12. MeRIP-seq of *Actb* mRNA targeted by TRM editors. (a,b) *Actb* read coverage of m^{6}A-immunoprecipitated (m^{6}A IP) RNA from MeRIP-seq of transfected HEK293T cells. *Actb* A1216 was targeted with (a) dCas13–M3nls or (b) dCas13–M3M14nes with the following conditions: active editor with a target guide RNA, inactive editor with a target guide RNA, and active editor with a non-target guide RNA. All DRACH motifs susceptible to TRM modification are annotated as ticks underneath the IGV track. The target guide RNA (purple) and targeted A1216 site (grey) are shown. Inactive TRM editors contain a methyltransferase-inactivating D395A mutation within M3. MeRIP-seq analysis was performed with n=5 independent biological replicates.



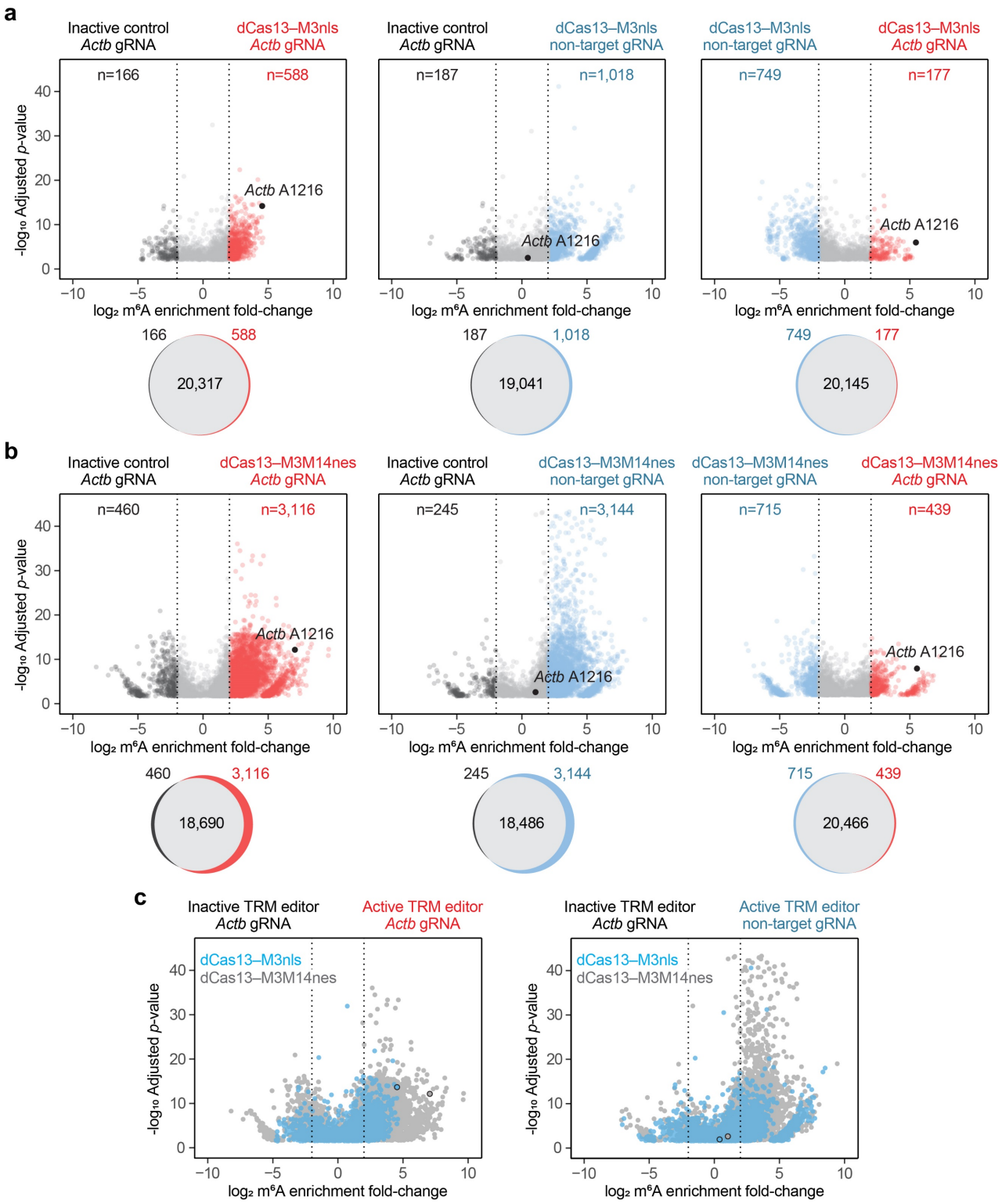
Supplementary Figure 13. TRM editing window on *Actb* and *Gapdh*. MeRIP-RT-qPCR of **(a)** *Actb* A1216 and **(b)** *Gapdh* A690 targeted by dCas13–M3nls and dCas13–M3M14nes with 30-nt guide RNAs (purple) ending at the indicated bp from the target adenine. NT = non-targeting. Inactive TRM editors contain a methyltransferase-inactivating D395A mutation within M3. Values and error bars reflect the mean±s.e.m. of n=3 independent biological replicates.



Supplementary Figure 14. Sequence preferences of TRM editors. Sequence logos of m^{6}A-enriched RNA from HEK293T cells expressing indicated constructs and guide RNAs. Logos for off-target sites from active dCas13–M3nls and dCas13–M3M14nes + *Actb* gRNA (2nd row) were created from 1,018 and 3,144 m^{6}A peaks, respectively, not found in the inactive TRM + *Actb* gRNA conditions. All other logos were generated from the 5,000 most-methylated m^{6}A peaks for each condition. The modified adenine is denoted as m^{6}A below the logos. Inactive TRM editors contain a methyltransferase-inactivating D395A mutation within M3. MeRIP-seq analysis for producing sequence logos was performed with n=5 independent biological replicates.

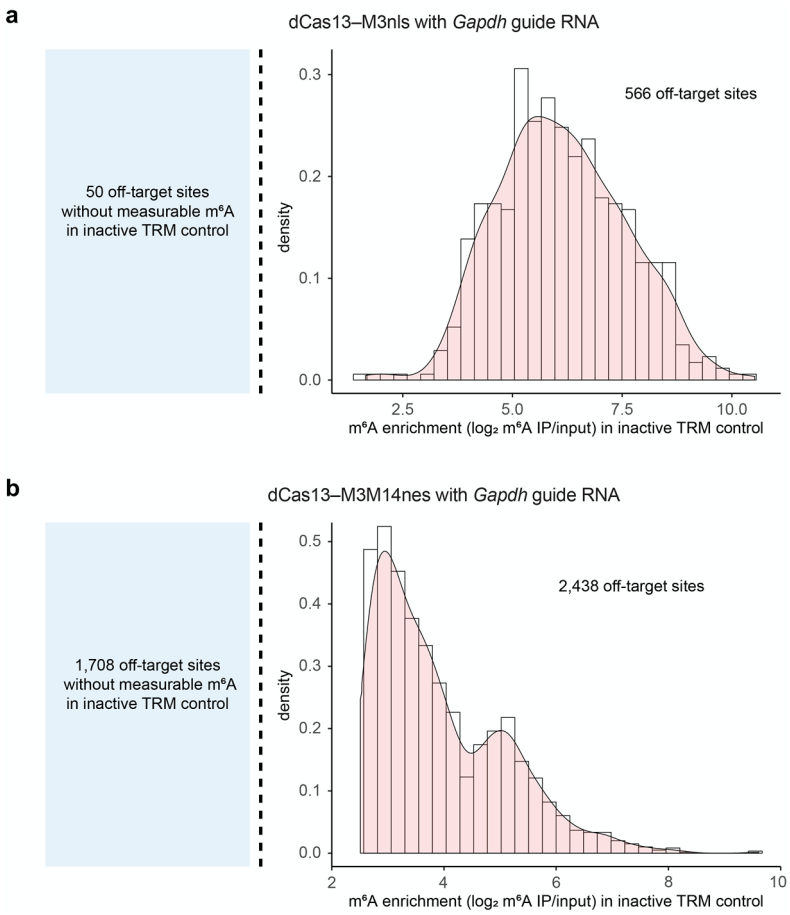


Supplementary Figure 15. Effect of TRM editors on cellular m^{6}A content. Relative m^{6}A/A in polyA+ RNA normalized to a transfected vector control. HEK293T cells were transfected with indicated TRM editors and non-targeting guide RNAs. Equal amounts of cellular mRNA were captured and detected with anti-m^{6}A antibodies to quantify m^{6}A/A ratios. Inactive TRM editors contain a methyltransferase-inactivating D395A mutation within M3. Values and error bars reflect the mean±s.e.m. of n=6 independent biological replicates. *P* values were calculated using a two-tailed Student's *t*-test.

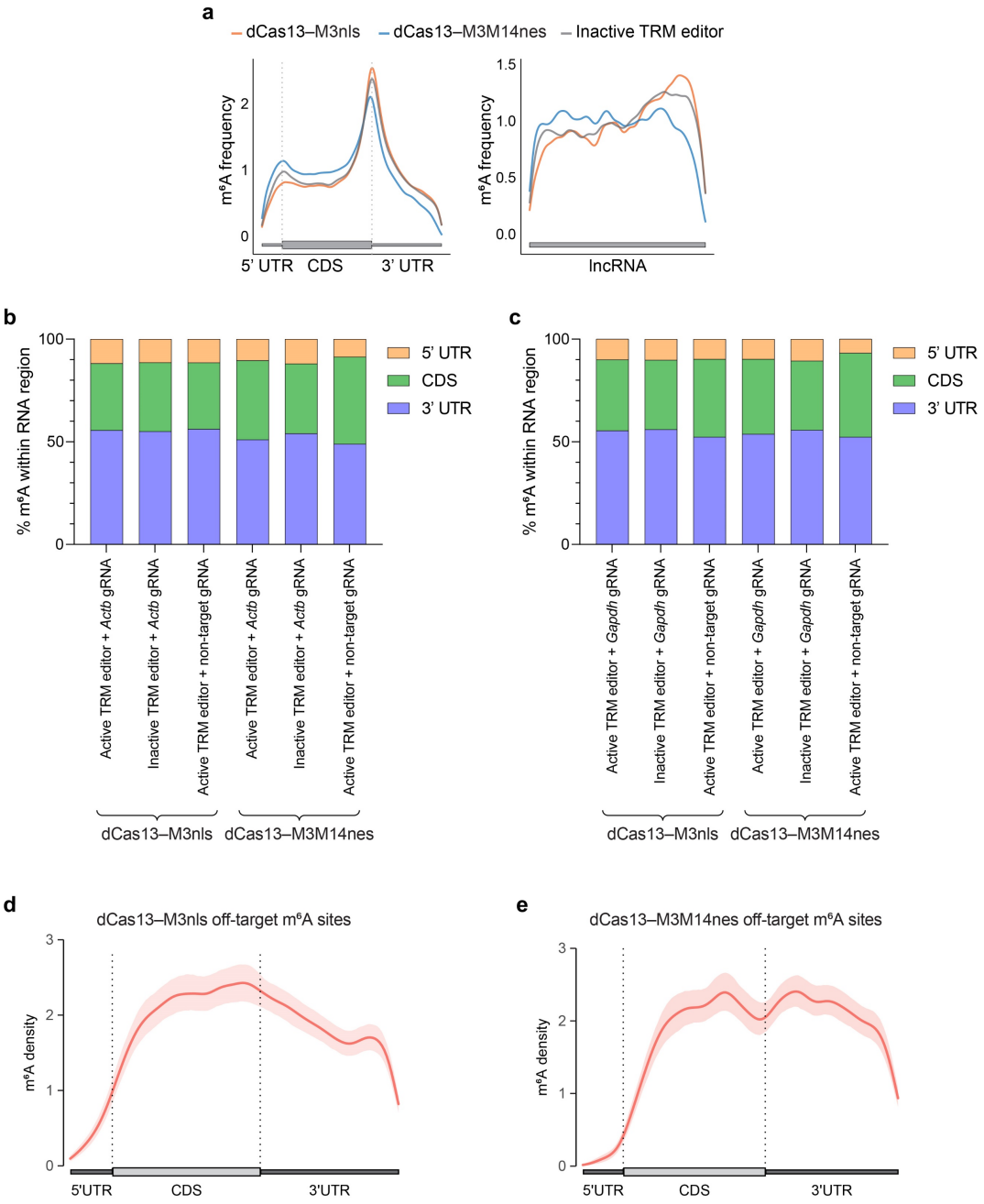


Supplementary Figure 16. Off-target methylation by TRM editors. (a,b) Differential m^{6}A enrichment of >21,000 methylated sites in HEK293T cells transfected with **(a)** dCas13–M3nls or **(b)** dCas13–M3M14nes and *Actb* A1216-targeting or non-target guide RNAs. Top: differential methylation of m^{6}A sites between conditions indicated above. Only differentially methylated sites with statistical significance (P<0.001) are shown. Bottom: Venn diagrams depicting overlap of all methylated m^{6}A sites for the above comparisons. **(c)** Overlay of dCas13–M3nls (blue) and dCas13–M3M14nes (grey) differential methylation with the following comparisons: left, active editors with *Actb* guide RNAs vs. inactive editors with *Actb* guide RNAs; right, active editors with non-target guide

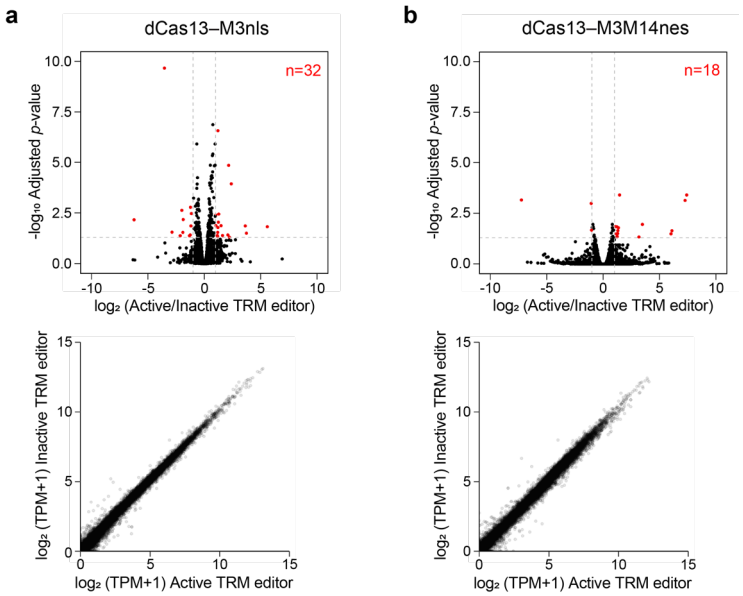
RNAs vs. inactive editors with *Actb* guide RNAs. The targeted *Actb* A1216 site is outlined in black. Inactive TRM editors contain a methyltransferase-inactivating D395A mutation within M3. MeRIP-seq analysis was performed with n=5 independent biological replicates. Statistical significance was calculated using a logs likelihood-ratio test with false discovery rate correction.



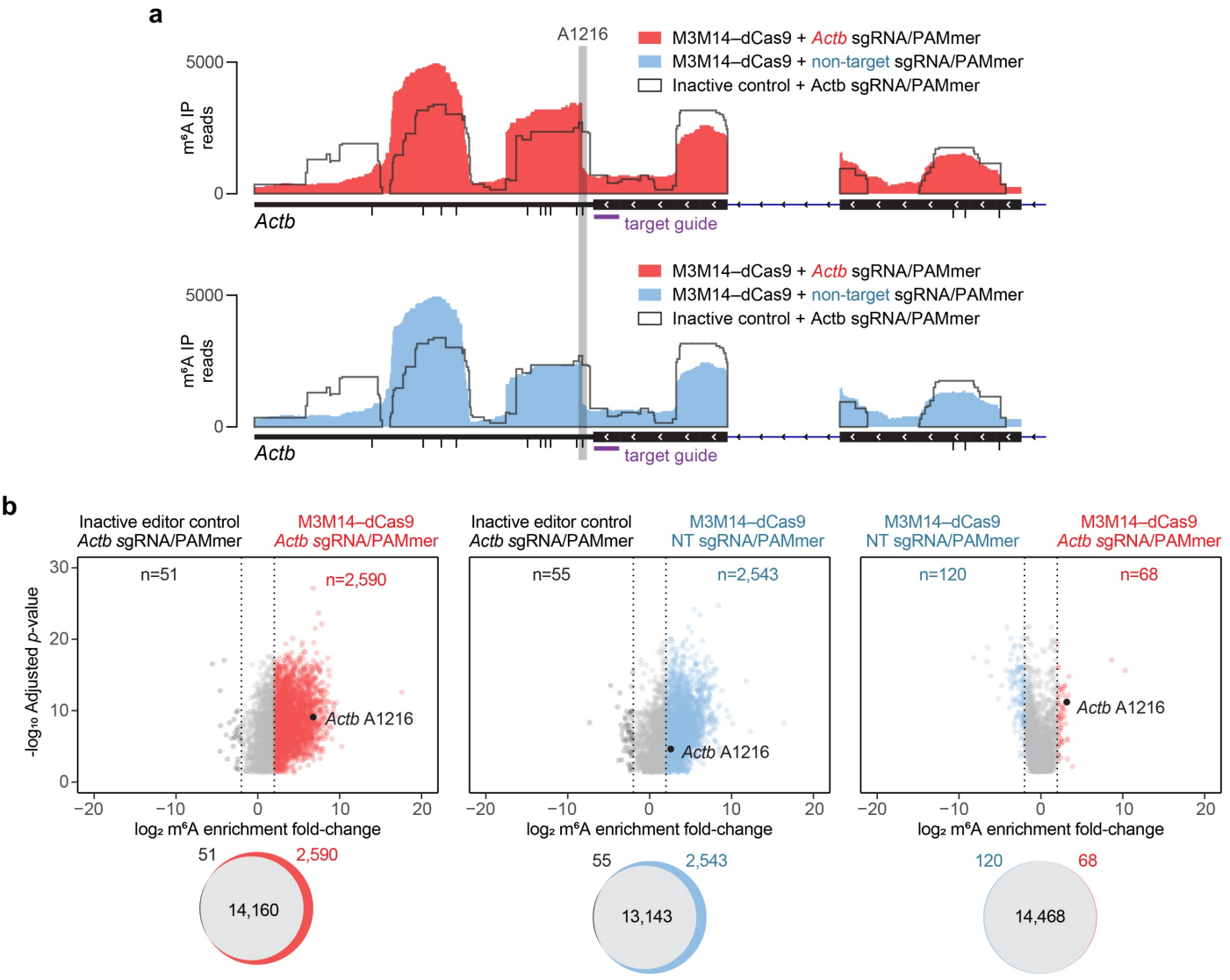
Supplementary Figure 17. Endogenous methylation levels of TRM editor off-targets. (a) Density plot depicting the m^6A enrichment (log_2 m^6A IP/input) of 566 dCas13–M3nls off-target sites in a methyltransferase-inactive TRM control. 50 off-targets did not have measurable methylation in the control. **(b)** Density plot depicting the m^6A enrichment (log_2 m^6A IP/input) of 2,438 dCas13–M3M14nes off-target sites in a methyltransferase-inactive TRM control. 1,708 off-targets did not have measurable methylation in the control. MeRIP-seq analysis was performed with n=5 independent biological replicates.



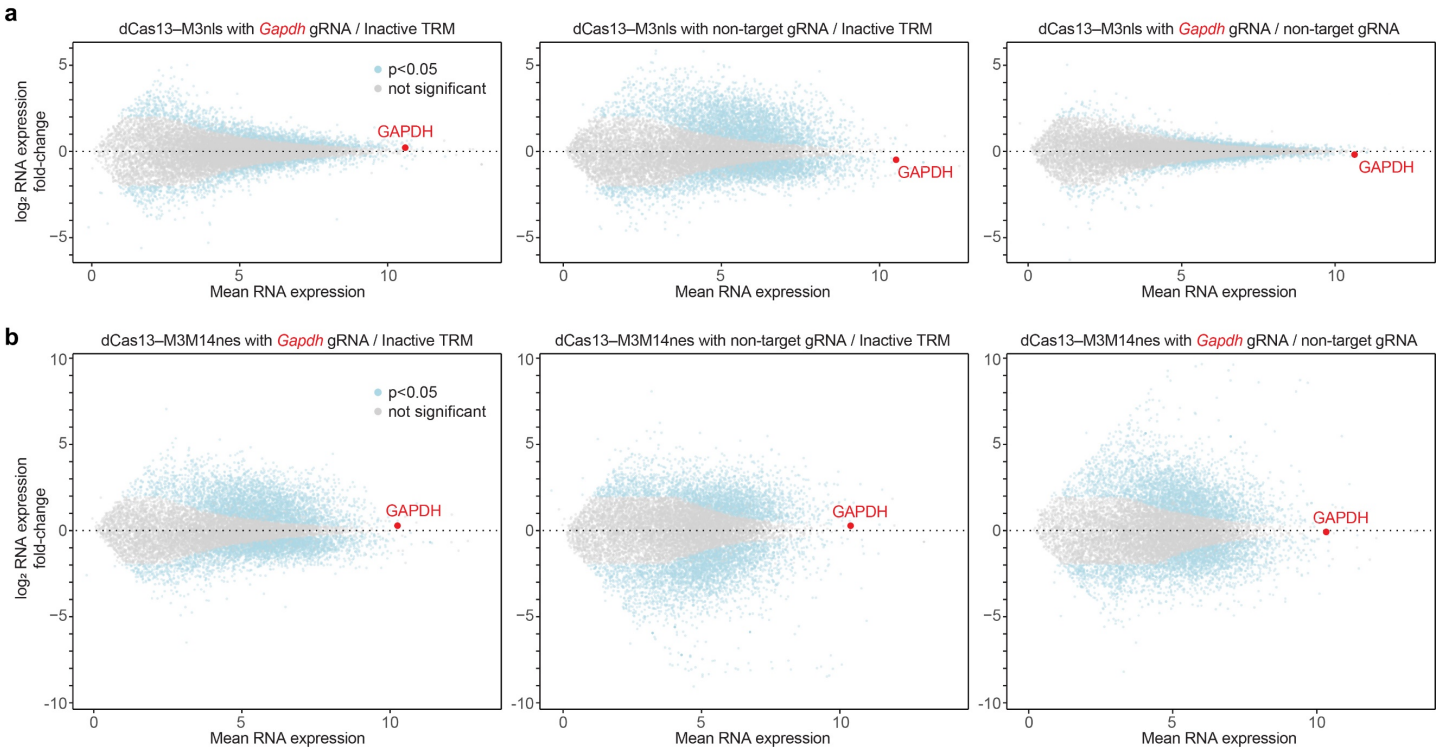
Supplementary Figure 18. Effects of TRM editors on transcriptome-wide distribution of m^{6}A. **(a)** Metagenome plots of m^{6}A distribution within mRNA (left) and lncRNA (right) in HEK293T cells transfected with indicated TRM editors and *Actb* guide RNAs. **(b,c)** Bar plots depicting the percentage of m^{6}A peaks within distinct mRNA regions, from HEK293T cells treated with the specified TRM editors and guide RNAs. *Actb* guide RNAs target the A1216 site, and *Gapdh* guide RNAs target the A690 site. 10,000 of the most-methylated m^{6}A peaks from each sample was used for each analysis. **(d,e)** Transcriptome-wide distribution of off-target m^{6}A sites from **(d)** dCas13–M3nls and **(e)** dCas13–M3M14nes, with 97.5% confidence intervals shaded in. Inactive TRM editors contain a methyltransferase-inactivating D395A mutation within M3. MeRIP-seq analysis for generating all plots was performed with n=5 independent biological replicates.



Supplementary Figure 19. Effects of TRM editors on cellular transcriptome abundances. Volcano and scatter plots depict differential gene transcript abundance in HEK293T cells transfected with **(a)** dCas13–M3nls and **(b)** dCas13–M3M14nes, each compared with its corresponding inactive TRM editor using methyltransferase-inactive M3^{D395} instead of M3. Cells were co-transfected with non-targeting guide RNAs for all conditions. Differentially expressed genes (P<0.05 and fold-change>2) are shown in red and counted in each volcano plot. TPM = transcripts per million. Differential RNA-seq analysis was performed with n=3 independent biological replicates, and more than 196,000 total transcripts from over 15,000 total genes were analyzed in each experiment. Statistical significance was calculated using a two-tailed Student's *t*-test with false discovery rate correction.



Supplementary Figure 20. Comparison of TRM editors with M3M14–dCas9 + PAMmer editors[3]. **(a)** *Actb* read coverage of m^{6}A-immunoprecipitated (m^{6}A IP) RNA fragments from MeRIP-seq of HEK293T cells transfected with indicated M3M14–dCas9 editors and single guide RNAs (sgRNAs) with corresponding PAMmers. The following conditions are shown: M3M14–dCas9 with sgRNA and PAMmers targeting *Actb* A1216 (red), M3M14–dCas9 with non-targeting sgRNA and PAMmers (blue), and inactive editor control with sgRNA and PAMmers targeting *Actb* A1216 (black outline). All DRACH motifs susceptible to TRM modification are shown as black tick marks underneath the IGV track. The target guide RNA (purple) and targeted A1216 site (grey) are shown. **(b)** Differential m^{6}A enrichment of >15,000 methylated sites in HEK293T cells transfected with M3M14–dCas9 and either *Actb* A1216-targeting sgRNA or non-targeting sgRNA with corresponding PAMmers. Top: differential methylation of m^{6}A sites between conditions indicated above. Only differentially methylated m^{6}A sites with statistical significance (p<0.01) are shown. Bottom: Venn diagrams depicting overlap of all methylated m^{6}A sites for the above comparisons. Inactive M3M14–dCas9 editor control bears a catalytically-inactivating D395 mutation within its METTL3 methyltransferase domain. MeRIP-seq analysis was performed on n=5 independent biological replicates. Statistical significance was calculated using a logs likelihood-ratio test with false discovery rate correction.



Supplementary Figure 21*. Gapdh* RNA expression changes from TRM editing. Differential RNA expression of HEK293T cells transfected with **(a)** dCas13–M3nls or **(b)** dCas13–M3M14nes and either the *Gapdh* A690-targeting guide RNA or a non-targeting guide RNA. The targeted GAPDH gene is marked in red. Inactive TRM control indicates editors containing a methyltransferase-inactivating M3 D395A mutation with a *Gapdh*-targeting guide RNA. Non-target gRNA control indicates methyltransferase-active TRM editors with a non-targeting guide RNA. Differential RNA-seq analysis was performed with n=5 independent biological replicates. Statistical significance was calculated using a two-tailed Student's *t*-test with false discovery rate correction.

Supplementary Table 1. RNA oligonucleotide sequences for *in vitro* kinetics assays. The adenine susceptible to methylation is bolded within the GGACU m^6A motif (red).

Name	RNA sequence
ssRNA	AGCUCGGUUCCGGUUGGGUUGG**A**CUGAAAUAUGGAUUACUUGGUAGAA

Supplementary Table 2. Cas13 guide RNA spacer sequences used in this study. A guanosine (lower-case g) was added to the 5' end of 30-nt guide RNA spacers (upper-case sequences) used in HEK293T cells, because the human U6 promoter has a strong preference for G at the transcription start site. TRM = targeted RNA methylation. IF = immunofluorescence.

Name	Guide RNA spacer sequence	Figures used in
Synthetic target guide	TGCAGAGTAGAGGCCGCAGGATAGTTTAGAAC	2c,d; SI 3
Non-targeting guide (*E. coli*)	GTAATTCCCTGGTCCGTCGACGCATAGTCTG	2c
Non-targeting guide (HEK293T)	gGTAATGCCTGGCTTGTCGACGCATAGTCTG	3b-c; 4a-e; 5a-e; 6a-d; SI 4a-b; SI 5b-d; SI 6b; SI 7; SI 8c; SI 9b; SI 10b; SI 11b-c; SI 12a-b; SI 13a-b; SI 14; SI 15; SI 16a-c; SI 18a-c; SI 19a-b; SI 21a-b
Cluc–syn guide 1	gTAGTCCGAGTCCAGTCCGGAGTCCGAGCAT	SI 5c
Cluc–syn guide 2	gTTCCAAACTATCCTGCGGCCTCTACTCTGC	3b; SI 5c
Cluc–Socs2 guide 1	gTACATAGCTGCATTCGGAGATACTCTATGT	3c; SI 4a-b; SI 5c
Cluc–Socs2 guide 2	gGATAAAACGCGACTTCAGCCTTTACATGCG	SI 5c
Actb guide IF	gGGGGGATGCTCGCTCCAACCGACTGCTGTC	4a
Actb A1216 guide -1	gCCGCCTAGAAGCATTTGCGGTGGACGATGG	SI 13a
Actb A1216 guide -8	gGAAGCATTTGCGGTGGACGATGGAGGGGCC	4b; 6a,b; SI 6b; SI 12a-b; SI 13a; SI 14; SI 16a-c; SI 18a-b
Actb A1216 guide -15	gTTGCGGTGGACGATGGAGGGGCCGGACTCG	SI 13a
Actb A1216 guide -22	gGGACGATGGAGGGGCCGGACTCGTCATACT	SI 13a
Gapdh A690 guide -1	gTCTGGAGAGCCCCGCGGCCATCACGCCACA	SI 13b
Gapdh A690 guide -8	gAGCCCCGCGGCCATCACGCCACAGTTTCCC	4c; 5a-e; SI 8c; SI 11b; SI 13b; SI 17a-b; SI 18c-e; SI 21a-b
Gapdh A690 guide -15	gCGGCCATCACGCCACAGTTTCCCGGAGGGG	SI 13b
Gapdh A690 guide -22	gCACGCCACAGTTTCCCGGAGGGGCCATCCA	SI 13b
Foxm1 A3488, A3504 guide	gGTATGATTGGGGACATTATCAGAGAAACAT	4d, SI 9b
Sox2 A1398, A1405 guide	gTCACATGTGTGAGAGGGGCAGTGTGCCGTT	4e, SI 10b; SI 11c
Cluc CDS guide 1	gAGGTTCTCCAGGGTGATGGACACCTTCCAG	SI 5b
Cluc CDS guide 2	gGTCTGTATCTTCCATCATTTTAAGATCTCC	SI 5b
Cluc CDS guide 3	gAATTGGTATCTTGCTTTGTCAAATGTATCG	SI 5b
Cluc CDS guide 4	gATGGATCTCTAATATGTACGACGAGCAGTT	SI 5b
Cluc CDS guide 5	gTACTTCTAGGGTGTCTCCATGCTTTATGTA	SI 5b
Cluc–*Nanog* guide 1	gGCCAGTGTCCAGACTGAAATTGAGTAATAT	SI 5c
Cluc–*Nanog* guide 2	gCACTCCAGCCTGGGCGACAGAGCAAGACTC	SI 5c
Hspa1a–Cluc guide 1	gGGGAAGCCTTGGGACAACGGGAGTCACTCT	SI 5d
Hspa1a–Cluc guide 2	gGGAACACTGGATCCGCGAGAAGAGCTCGGT	SI 5d
Hsph1–Cluc guide 1	gCCTCAGCCTTATGTATCGCACTGATCAGAA	SI 5d
Hsph1–Cluc guide 2	gCTCCGGCCCCCTGCCTGCTTCTCCTGCCGC	SI 5d

Brd8 guide 1	gTGAAGCATCCTGTTTGCGGGTAGAATCTCT	6c
Znf638 guide 1	gAAATGTGTAAGCCTGACATCTTGGTTCCAC	6d

Supplementary Table 3. Cas9 guide RNA spacer and PAMmer sequences used for experiments with M3M14-dCas9[3]. A guanosine (lower-case g) was added to the 5' end of 20-nt single guide RNA (sgRNA) spacers (upper-case sequences) used in HEK293T cells, because the human U6 promoter has a strong preference for G at the transcription start site.

Name	Sequence
M3M14-dCas9 *Actb* sgRNA	gCACCGCAAATGCTTCTAGGC
M3M14-dCas9 non-targeting sgRNA	gGTATTACTGATATTGGTGGG
M3M14-dCas9 *Actb* PAMmer	mUTmGCmGGmUGmUGGmGAmUGmGAmGGmGGmCCmGGmAC
M3M14-dCas9 non-targeting PAMmer	mGCmGGmAGmGAmGCTmUGmCGmAGmCGmGTmGGmUTmGG

Supplementary Table 4. Sequences of guide-quenching DNA oligonucleotides. Because guide RNAs interfere with RT-qPCR of targeted transcripts, DNA oligos were used to quench guides within total RNA purified from HEK293T cells. DNA oligos were designed to hybridize with guide RNA spacer sequences and the PspCas13b crRNA hairpin. The constant crRNA-binding region is shown in lower-case. A 2',3'-dideoxycytidine modification (/3ddc/) hybridizes with the guide RNA's 5' G and prevents polymerase extension of the guide-quenching oligo in subsequent reverse-transcription and PCR reactions.

Guide RNA name	**Guide-quenching DNA oligo sequence**
Non-targeting guide (HEK293T)	ggaccttccacaacCAGACTATGCGTCGACAAGCCAGGCATTAC/3ddc/
Cluc–syn guide 1	ggaccttccacaacGCAGAGTAGAGGCCGCAGGATAGTTTGGAA/3ddc/
Cluc–Socs2 guide 1	ggaccttccacaacACATAGAGTATCTCCGAATGCAGCTATGTA/3ddc/
Actb A1216 guide -1	ggaccttccacaacCCATCGTCCACCGCAAATGCTTCTAGGCGG/3ddc/
Actb A1216 guide -8	ggaccttccacaacGGCCCCTCCATCGTCCACCGCAAATGCTTC/3ddc/
Actb A1216 guide -15	ggaccttccacaacCGAGTCCGGCCCCTCCATCGTCCACCGCAA/3ddc/
Actb A1216 guide -22	ggaccttccacaacAGTATGACGAGTCCGGCCCCTCCATCGTCC/3ddc/
Gapdh A690 guide -1	ggaccttccacaacTGTGGCGTGATGGCCGCGGGGCTCTCCAGA/3ddc/
Gapdh A690 guide -8	ggaccttccacaacGGGAAACTGTGGCGTGATGGCCGCGGGGCT/3ddc/
Gapdh A690 guide -15	ggaccttccacaacCCCCTCCGGGAAACTGTGGCGTGATGGCCG/3ddc/
Gapdh A690 guide -22	ggaccttccacaacTGGATGGCCCCTCCGGGAAACTGTGGCGTG/3ddc/
Foxm1 A3488, A3504 guide	ggaccttccacaacATGTTTCTCTGATAATGTCCCCAATCATAC/3ddc/
Sox2 A1398, A1405 guide	ggaccttccacaacAACGGCACACTGCCCCTCTCACACATGTGA/3ddc/

Supplementary Table 5. Primers used for RT-qPCR in TRM methylation assays, mazF, or dCas13 targeting experiments. All probes were synthesized from Bio-Rad and used for TaqMan qPCR.

Name	Sequence
Synthetic target forward	CCGCAGGATAGTTTGGAACATGGACT
Synthetic target reverse	GGCCGCGGGGATCCAGACAT
Synthetic target probe	/56-FAM/CTTTGGACTTGATGAGCGGCCGCGGG/3IABkFQ/
Cluc–syn forward	GAAGAAACCCGTGGCCAAGATGCTC
Cluc–syn reverse	GAACATGGACTCTAGGACTGGACTTTG
Cluc-syn probe	/56-FAM/CCGGACTGGACTCGGACTAATGCAGAGTAGAGG/3IABkFQ/
Cluc-Socs2 forward	GGGGACTGCCTTTACCAACAAGACTAAAA
Cluc–Socs2 reverse	CTCAAGTTTTGGTTCTCTTTTACATAGCTGCATTC
Cluc-Socs2 probe	/56-FAM/CCAGGTATAAATGTTTCTCTTTTTTTAAACATGTCTCACATAGAGTATCTCC/3IABkFQ/
Actb A1216 target 1 forward	ATCGTCCACCGCAAATGCTT
Actb A1216 target 1 reverse	TCATCTTGTTTTCTGCGCAAGT
Actb A1216 target 1 probe	/56-FAM/AGTCCGCCT/ZEN/AGAAGCATTTGCGGT/3IABkFQ/
Actb A1216 target 2 forward	TCCATCGTCCACCGCAAAT
Actb A1216 target 2 reverse	TTGTTTTCTGCGCAAGTTAGGT
Actb A1216 target 2 probe	/5HEX/CCGCCTAGA/ZEN/AGCATTTGCGGTGG/3IABkFQ/
Actb reference control forward	Bio-Rad qHsaCEP0036280
Actb reference control reverse	Bio-Rad qHsaCEP0036280
Actb reference control probe	Bio-Rad qHsaCEP0036280 (Cy5-labeled)
Gapdh A690 target forward	CATCACTGCCACCCAGAAGA
Gapdh A690 target reverse	CAGTAGAGGCAGGGATGATGTT
Gapdh A690 target probe	/56-FAM/CCCTCCGGG/ZEN/AAACTGTGGCGT/3IABkFQ/
Gapdh reference control forward	TCAAGGCTGAGAACGGGAAG
Gapdh reference control reverse	GGACTCCACGACGTACTCAG
Gapdh reference control probe	/5Cy55/TCCAGGAGCGAGATCCCTCC/3IAbRQSp/
Foxm1 A3488, A3504 target 1 forward	TGCCCAGATGTGCGCTATTA
Foxm1 A3488, A3504 target 1 reverse	CTTCTCAAGCCTCCACCTGA
Foxm1 A3488, A3504 target 1 probe	/56-FAM/TCGTCAATG/ZEN/CCAGTCTCCCTGGT/3IABkFQ/
Foxm1 A3488, A3504 target 2 forward	GTGCCCAGATGTGCGCTAT
Foxm1 A3488, A3504 target 2 reverse	GTCAATGCCAGTCTCCCTGG
Foxm1 A3488, A3504 target 2 probe	/5HEX/CCCAATCAT/ZEN/ACCAGGGAGACTGGCA/3IABkFQ/

Foxm1 reference control forward	Bio-Rad qHsaCEP0050615
Foxm1 reference control reverse	Bio-Rad qHsaCEP0050615
Foxm1 reference control probe	Bio-Rad qHsaCEP0050615 (Cy5-labeled)
Sox2 A1398, A1405 target 1 forward	GGCCATTAACGGCACACTG
Sox2 A1398, A1405 target 1 reverse	TCTTTTGCACCCCTCCCATT
Sox2 A1398, A1405 target 1 probe	/56-FAM/TCACACATG/ZEN/TGAGGGCCGGACAG/3IABkFQ/
Sox2 A1398, A1405 target 2 forward	CGGCCATTAACGGCACACT
Sox2 A1398, A1405 target 2 reverse	CCCTCCCATTTCCCTCGTTTT
Sox2 A1398, A1405 target 2 probe	/5HEX/CATGTGAGG/ZEN/GCCGGACAGCGAAC/3IABkFQ/
Sox2 reference control forward	Bio-Rad qHsaCEP0039595
Sox2 reference control reverse	Bio-Rad qHsaCEP0039595
Sox2 reference control probe	Bio-Rad qHsaCEP0039595 (Cy5-labeled)
Sox2 A1398, A1405 MazF target forward	CATGTGAGGGCCGGACAG
Sox2 A1398, A1405 MazF target reverse	TCTTTTGCACCCCTCCCATT
Sox2 A1398, A1405 MazF target probe	/56-FAM/CGAGGGAAA/ZEN/TGGGAGGGGTGCAAA/3IABkFQ/
Sox2 MazF reference control forward	CATGGGTTCGGTGGTCAAG
Sox2 MazF reference control reverse	TGGAGTGGGAGGAAGAGGTA
Sox2 MazF reference control probe	/5Cy5/TCCGAGGCCAGCTCCAGCCC/3IAbRQSp/
EEF1A1 m^6A+ control forward	CGGTCTCAGAACTGTTTGTTTC
EEF1A1 m^6A+ control reverse	AAACCAAAGTGGTCCACAAA
EEF1A1 m^6A+ control probe	/5TEX615/CGTAAAACCTTCAGAAGGAAAGGAG/3IAbRQSp/
EEF1A1 m^6A- control forward	GGATGGAAAGTCACCCGTAAG
EEF1A1 m^6A- control reverse	TTGTCAGTTGGACGAGTTGG
EEF1A1 m^6A- control probe	/5Cy55/AACCACGCTGCTTGAGGCTC/3IAbRQSp/
Cypridina luciferase forward	ACTGTAAACGGTGGAGCTGA
Cypridina luciferase reverse	AAGCCTGGCATCTCAACAAC
Cypridina luciferase probe	/5Cy5/TCGGCGAGGTCACCATCGCT/3IAbRQSp/
Gaussia luciferase forward	ATCTGCCTGTCCCACATCAA
Gaussia luciferase reverse	CGGACTCTTTGTCGCCTTC

Gaussia luciferase probe	/56-FAM/CCCAGGACG/ZEN/CTGCCACACCT/3IABkFQ/

Supplementary Table 6. Primers used for semi-quantitative PCR in alternative splicing assays.

Name	Sequence
Znf638 exon 1 forward	CGTCGAGACTGGAGGCTGAG
Znf638 exon 3 reverse	TGAGTAGATGTATTTTGATGCTGAATCC
Brd8 exon 20 forward	GAGAGATTCTACCCGCAAACAGG
Brd8 exon 22 reverse	TATCTGCTTCAATGGCACAGCGG

Supplementary Sequences 1. Amino acid sequences of nucleus- and cytoplasm-localized TRM editors. Color coding is as follows:
green = HIV nuclear export signal
red = bipartite SV40 nuclear localization signal
blue = PspCas13b Δ984-1090 H133A (dCas13)
black = linkers
orange = METTL3$^{273-580}$ (for dCas13–M3nes and dCas13–M3nls) or METTL3$^{359-580}$ (for dCas13–M3M14nes and dCas13–M3M14nls)
brown = METTL14$^{111-456}$

dCas13–M3nes:
MNIPALVENQKKYFGTYSVMAMLNAQTVLDHIQKVADIEGEQNENNENLWFHPVMSHLYNAKNGYD
KQPEKTMFIIERLQSYFPFLKIMAENQREYSNGKYKQNRVEVNSNDIFEVLKRAFGVLKMYRDLTNAY
KTYEEKLNDGCEFLTSTEQPLSGMINNYYTVALRNMNERYGYKTEDLAFIQDKRFKFVKDAYGKKKS
QVNTGFFLSLQDYNGDTQKKLHLSGVGIALLICLFLDKQYINIFLSRLPIFSSYNAQSEERRIIIRSFGINS
IKLPKDRIHSEKSNKSVAMDMLNEVKRCPDELFTTLSAEKQSRFRIISDDHNEVLMKRSSDRFVPLLL
QYIDYGKLFDHIRFHVNMGKLRYLLKADKTCIDGQTRVRVIEQPLNGFGRLEEAETMRKQENGTFGN
SGIRIRDFENMKRDDANPANYPYIVDTYTHYILENNKVEMFINDKEDSAPLLPVIEDDRYVVKTIPSCR
MSTLEIPAMAFHMFLFGSKKTEKLIVDVHNRYKRLFQAMQKEEVTAENIASFGIAESDLPQKILDLISG
NAHGKDVDAFIRLTVDDMLTDTERRIKRFKDDRKSIRSADNKMGKRGFKQISTGKLADFLAKDIVLFQ
PSVNDGENKITGLNYRIMQSAIAVYDSGDDYEAKQQFKLMFEKARLIGKGTTEPHPFLYKVFARSIPA
NAVEFYERYLIERKFYLTGLSNEIKKGNRVDVPFIRRDQNKWKTPAMKTLGRIYSEDLPVELPRQMFD
NEIKSHLKSLPQMEGIDFNNANVTYLIAEYMKRVLDDDFQTFYQWNRNYRYMDMLKGEYDRKGSLQ
HCFTSVEEREGLWKERASRTERYRKQASNKIRSNRQMRNASSEEIETILDKRLSNSRNEYQKSEKVI
RRYRVQDALLFLLAKKTLTELADFDGERFKLKEIMPDAEKGILSEIMPMSFTFEKGGKKYTITSEGMKL
KNYGDFFVLASDKRIGNLLELVGSDIVSKEDGSLQLPPLERLTLSGSETPGTSESATPESQEFCDYGT
KEECMKASDADRPCRKLHFRRIINKHTDESLGDCSFLNTCFHMDTCKYVHYEIDACMDSEAPGSKDH
TPSQELALTQSVGGDSSADRLFPPQWICCDIRYLDVSILGKFAVVMADPPWDIHMELPYGTLTDDEM
RRLNIPVLQDDGFLFLWVTGRAMELGRECLNLWGYERVDEIIWVKTNQLQRIIRTGRTGHWLNHGKE
HCLVGVKGNPQGFNQGLDCDVIVAEVRSTSHKPDEIYGMIERLSPGTRKIELFGRPHNVQPNWITLG
NQLDGIHLLDPDVVARFKQRYPDGIISKPKNL

dCas13–M3nls:
MKRTADGSEFESPKKKRKVNIPALVENQKKYFGTYSVMAMLNAQTVLDHIQKVADIEGEQNENNENL
WFHPVMSHLYNAKNGYDKQPEKTMFIIERLQSYFPFLKIMAENQREYSNGKYKQNRVEVNSNDIFEV
LKRAFGVLKMYRDLTNAYKTYEEKLNDGCEFLTSTEQPLSGMINNYYTVALRNMNERYGYKTEDLAF
IQDKRFKFVKDAYGKKKSQVNTGFFLSLQDYNGDTQKKLHLSGVGIALLICLFLDKQYINIFLSRLPIFS
SYNAQSEERRIIIRSFGINSIKLPKDRIHSEKSNKSVAMDMLNEVKRCPDELFTTLSAEKQSRFRIISDD
HNEVLMKRSSDRFVPLLLQYIDYGKLFDHIRFHVNMGKLRYLLKADKTCIDGQTRVRVIEQPLNGFGR
LEEAETMRKQENGTFGNSGIRIRDFENMKRDDANPANYPYIVDTYTHYILENNKVEMFINDKEDSAPL
LPVIEDDRYVVKTIPSCRMSTLEIPAMAFHMFLFGSKKTEKLIVDVHNRYKRLFQAMQKEEVTAENIAS
FGIAESDLPQKILDLISGNAHGKDVDAFIRLTVDDMLTDTERRIKRFKDDRKSIRSADNKMGKRGFKQI
STGKLADFLAKDIVLFQPSVNDGENKITGLNYRIMQSAIAVYDSGDDYEAKQQFKLMFEKARLIGKGT
TEPHPFLYKVFARSIPANAVEFYERYLIERKFYLTGLSNEIKKGNRVDVPFIRRDQNKWKTPAMKTLG
RIYSEDLPVELPRQMFDNEIKSHLKSLPQMEGIDFNNANVTYLIAEYMKRVLDDDFQTFYQWNRNYR

YMDMLKGEYDRKGSLQHCFTSVEEREGLWKERASRTERYRKQASNKIRSNRQMRNASSEEIETILD
KRLSNSRNEYQKSEKVIRRYRVQDALLFLLAKKTLTELADFDGERFKLKEIMPDAEKGILSEIMPMSFT
FEKGGKKYTITSEGMKLKNYGDFFVLASDKRIGNLLELVGSDIVSKEDGSKRTADGSEFEPKKKRKVS
GSETPGTSESATPESQEFCDYGTKEECMKASDADRPCRKLHFRRIINKHTDESLGDCSFLNTCFHM
DTCKYVHYEIDACMDSEAPGSKDHTPSQELALTQSVGGDSSADRLFPPQWICCDIRYLDVSILGKFA
VVMADPPWDIHMELPYGTLTDDEMRRLNIPVLQDDGFLFLWVTGRAMELGRECLNLWGYERVDEII
WVKTNQLQRIIRTGRTGHWLNHGKEHCLVGVKGNPQGFNQGLDCDVIVAEVRSTSHKPDEIYGMIE
RLSPGTRKIELFGRPHNVQPNWITLGNQLDGIHLLDPDVVARFKQRYPDGIISKPKNL

dCas13–M3M14nes:
MNIPALVENQKKYFGTYSVMAMLNAQTVLDHIQKVADIEGEQNENNENLWFHPVMSHLYNAKNGYD
KQPEKTMFIIERLQSYFPFLKIMAENQREYSNGKYKQNRVEVNSNDIFEVLKRAFGVLKMYRDLTNAY
KTYEEKLNDGCEFLTSTEQPLSGMINNYYTVALRNMNERYGYKTEDLAFIQDKRFKFVKDAYGKKKS
QVNTGFFLSLQDYNGDTQKKLHLSGVGIALLICLFLDKQYINIFLSRLPIFSSYNAQSEERRIIIRSFGINS
IKLPKDRIHSEKSNKSVAMDMLNEVKRCPDELFTTLSAEKQSRFRIISDDHNEVLMKRSSDRFVPLLL
QYIDYGKLFDHIRFHVNMGKLRYLLKADKTCIDGQTRVRVIEQPLNGFGRLEEAETMRKQENGTFGN
SGIRIRDFENMKRDDANPANYPYIVDTYTHYILENNKVEMFINDKEDSAPLLPVIEDDRYVVKTIPSCR
MSTLEIPAMAFHMFLFGSKKTEKLIVDVHNRYKRLFQAMQKEEVTAENIASFGIAESDLPQKILDLISG
NAHGKDVDAFIRLTVDDMLTDTERRIKRFKDDRKSIRSADNKMGKRGFKQISTGKLADFLAKDIVLFQ
PSVNDGENKITGLNYRIMQSAIAVYDSGDDYEAKQQFKLMFEKARLIGKGTTEPHPFLYKVFARSIPA
NAVEFYERYLIERKFYLTGLSNEIKKGNRVDVPFIRRDQNKWKTPAMKTLGRIYSEDLPVELPRQMFD
NEIKSHLKSLPQMEGIDFNNANVTYLIAEYMKRVLDDDFQTFYQWNRNYRYMDMLKGEYDRKGSLQ
HCFTSVEEREGLWKERASRTERYRKQASNKIRSNRQMRNASSEEIETILDKRLSNSRNEYQKSEKVI
RRYRVQDALLFLLAKKTLTELADFDGERFKLKEIMPDAEKGILSEIMPMSFTFEKGGKKYTITSEGMKL
KNYGDFFVLASDKRIGNLLELVGSDIVSKEDGSLQLPPLERLTLSGGSSGGSSGSETPGTSESATPES
SGGSSGGSVGGDSSADRLFPPQWICCDIRYLDVSILGKFAVVMADPPWDIHMELPYGTLTDDEMRR
LNIPVLQDDGFLFLWVTGRAMELGRECLNLWGYERVDEIIWVKTNQLQRIIRTGRTGHWLNHGKEHC
LVGVKGNPQGFNQGLDCDVIVAEVRSTSHKPDEIYGMIERLSPGTRKIELFGRPHNVQPNWITLGNQ
LDGIHLLDPDVVARFKQRYPDGIISKPKNLGGSGGSGGSGGSGGSGGSGGSGGSGGSGSGQSLNP
HNDYCQHFVDTGHRPQNFIRDVGLADRFEEYPKLRELIRLKDELIAKSNTPPMYLQADIEAFDIRELTP
KFDVILLEPPLEEYYRETGITANEKCWTWDDIMKLEIDEIAAPRSFIFLWCGSGEGLDLGRVCLRKWG
YRRCEDICWIKTNKNNPGKTKTLDPKAVFQRTKEHCLMGIKGTVKRSTDGDFIHANVDIDLIITEEPEIG
NIEKPVEIFHIIEHFCLGRRRLHLFGRDSTIRPGWLTVGPTLTNSNYNAETYASYFSAPNSYLTGCTEEI
ERLRPKSPPPKSKSDRGGGAPRGGGRGGTSAGRGRERNRSNFRGERGGFRGGRGGAHRGGFPP
R

dCas13–M3M14nls:
MKRTADGSEFESPKKKRKVNIPALVENQKKYFGTYSVMAMLNAQTVLDHIQKVADIEGEQNENNENL
WFHPVMSHLYNAKNGYDKQPEKTMFIIERLQSYFPFLKIMAENQREYSNGKYKQNRVEVNSNDIFEV
LKRAFGVLKMYRDLTNAYKTYEEKLNDGCEFLTSTEQPLSGMINNYYTVALRNMNERYGYKTEDLAF
IQDKRFKFVKDAYGKKKSQVNTGFFLSLQDYNGDTQKKLHLSGVGIALLICLFLDKQYINIFLSRLPIFS
SYNAQSEERRIIIRSFGINSIKLPKDRIHSEKSNKSVAMDMLNEVKRCPDELFTTLSAEKQSRFRIISDD
HNEVLMKRSSDRFVPLLLQYIDYGKLFDHIRFHVNMGKLRYLLKADKTCIDGQTRVRVIEQPLNGFGR
LEEAETMRKQENGTFGNSGIRIRDFENMKRDDANPANYPYIVDTYTHYILENNKVEMFINDKEDSAPL
LPVIEDDRYVVKTIPSCRMSTLEIPAMAFHMFLFGSKKTEKLIVDVHNRYKRLFQAMQKEEVTAENIAS

FGIAESDLPQKILDLISGNAHGKDVDAFIRLTVDDMLTDTERRIKRFKDDRKSIRSADNKMGKRGFKQI
STGKLADFLAKDIVLFQPSVNDGENKITGLNYRIMQSAIAVYDSGDDYEAKQQFKLMFEKARLIGKGT
TEPHPFLYKVFARSIPANAVEFYERYLIERKFYLTGLSNEIKKGNRVDVPFIRRDQNKWKTPAMKTLG
RIYSEDLPVELPRQMFDNEIKSHLKSLPQMEGIDFNNANVTYLIAEYMKRVLDDDFQTFYQWNRNYR
YMDMLKGEYDRKGSLQHCFTSVEEREGLWKERASRTERYRKQASNKIRSNRQMRNASSEEIETILD
KRLSNSRNEYQKSEKVIRRYRVQDALLFLLAKKTLTELADFDGERFKLKEIMPDAEKGILSEIMPMSFT
FEKGGKKYTITSEGMKLKNYGDFFVLASDKRIGNLLELVGSDIVSKEDGSKRTADGSEFEPKKKRKVS
GGSSGGSSGSETPGTSESATPESSGGSSGGSVGGDSSADRLFPPQWICCDIRYLDVSILGKFAVVM
ADPPWDIHMELPYGTLTDDEMRRLNIPVLQDDGFLFLWVTGRAMELGRECLNLWGYERVDEIIWVKT
NQLQRIIRTGRTGHWLNHGKEHCLVGVKGNPQGFNQGLDCDVIVAEVRSTSHKPDEIYGMIERLSPG
TRKIELFGRPHNVQPNWITLGNQLDGIHLLDPDVVARFKQRYPDGIISKPKNLGGSGGSGGSGGSGG
SGGSGGSGGSGGSGGSGSGQSLNPHNDYCQHFVDTGHRPQNFIRDVGLADRFEEYPKLRELIRLKDELI
AKSNTPPMYLQADIEAFDIRELTPKFDVILLEPPLEEYYRETGITANEKCWTWDDIMKLEIDEIAAPRSFI
FLWCGSGEGLDLGRVCLRKWGYRRCEDICWIKTNKNNPGKTKTLDPKAVFQRTKEHCLMGIKGTVK
RSTDGDFIHANVDIDLIITEEPEIGNIEKPVEIFHIIEHFCLGRRRLHLFGRDSTIRPGWLTVGPTLTNSNY
NAETYASYFSAPNSYLTGCTEEIERLRPKSPPPKSKSDRGGGAPRGGGRGGTSAGRGRERNRSNF
RGERGGFRGGRGGAHRGGFPPR

Supplementary Sequences 2. Sequences of exogenous reporter transcripts targeted by TRM editor constructs. Color coding is as follows:
black = reporter sequence, with the TRM guide RNA-targeted protospacer underlined
green = *Cypridina* luciferase CDS
red = targeted m^{6}A motifs (DRACH), with target adenines marked in **bold**

Synthetic target (*E. coli* Tuner DE3):
ACCGACCAGAAUCAUGCAAGUGCGUAAGAUAGUCGCGGGCCGGGGAGTTTAATACGACTCACT
ATAGGGGAATTGTGAGCGGATAACAATTCCCCCTCTAGAAATAATTTTGTTTAACATTTAAATCGG
ACUCCGG**A**CUGG**A**CUCGG**A**CUAAUGCAGAGUAGAGGCCGCAGGAUAGUUUGGAACAUGG**A**C
UCUAGGACUGG**A**CUUUGG**A**CUUGUGUUUAUCCGCUCACAAUUCCACACAACAUACGAGCCGG
AAGCAUAAAGTGACTTCCCATCTCCGGTTTAAACAGGCCTGCATGCAGCGTAATCATGGTCATTA
ATTAACCTCCTGTGTGAAATTGTTATCCGCTCTGCGTTAGCAATTTAACTGTGAACCGACCAGAA
UCAUGCAAGUGCGUAAGAUAGUCGCGGGCCGGGGAGTT

Cluc–syn (HEK293T):
AUGAAGACCUUAAUUCUUGCCGUUGCAUUAGUCUACUGCGCCACUGUUCAUUGCCAGGACUG
UCCUUACGAACCUGAUCCACCAAACACAGUUCCAACUUCCUGUGAAGCUAAAGAAGGAGAAUG
UAUUGAUAGCAGCUGUGGCACCUGCACGAGAGACAUACUAUCAGAUGGACUGUGUGAAAAUA
AACCAGGAAAAACAUGUUGUCGAAUGUGUCAGUAUGUAAUUGAAUGCAGAGUAGAGGCCGCA
GGAUGGUUUAGAACAUUCUAUGGAAAGAGAUUCCAGUUCCAGGAACCUGGUACAUACGUGUU
GGGUCAAGGAACCAAGGGCGGCGACUGGAAGGUGUCCAUCACCCUGGAGAACCUGGAUGGA
ACCAAGGGGGCUGUGCUGACCAAGACAAGACUGGAAGUGGCUGGAGACAUCAUUGACAUCGC
UCAAGCUACUGAGAAUCCCAUCACUGUAAACGGUGGAGCUGACCCUAUCAUCGCCAACCCGU
ACACCAUCGGCGAGGUCACCAUCGCUGUUGUUGAGAUGCCAGGCUUCAACAUCACCGUCAUU
GAGUUCUUCAAACUGAUCGUGAUCGACAUCCUCGGAGGAAGAUCUGUAAGAAUCGCCCCAGA
CACAGCAAACAAAGGAAUGAUCUCUGGCCUCUGUGGAGAUCUUAAAAUGAUGGAAGAUACAG
ACUUCACUUCAGAUCCAGAACAACUCGCUAUUCAGCCUAAGAUCAACCAGGAGUUUGACGGU
UGUCCACUCUAUGGAAAUCCUGAUGACGUUGCAUACUGCAAAGGUCUUCUGGAGCCGUACAA
GGACAGCUGCCGCAACCCCAUCAACUUCUACUACUACACCAUCUCCUGCGCCUUCGCCCGCU
GUAUGGGUGGAGACGAGCGAGCCUCACACGUGCUGCUUGACUACAGGGAGACGUGCGCUGC
UCCCGAAACUAGAGGAACCUGCGUUUUGUCUGGACAUACUUUCUACGAUACAUUUGACAAAG
CAAGAUACCAAUUCCAGGGUCCCUGCAAGGAGAUUCUUAUGGCCGCCGACUGUUUCUGGAAC
ACUUGGGAUGUGAAGGUUUCACACAGGAAUGUUGACUCUUACACUGAAGUAGAGAAAGUACG
AAUCAGGAAACAAUCGACUGUAGUAGAACUCAUUGUUGAUGGAAAACAGAUUCUGGUUGGAG
GAGAAGCCGUGUCCGUCCCGUACAGCUCUCAGAACACUUCCAUCUACUGGCAAGAUGGUGAC
AUACUGACUACAGCCAUCCUACCUGAAGCUCUGGUGGUCAAGUUCAACUUCAAGCAACUGCU
CGUCGUACAUAUUAGAGAUCCAUUCGAUGGUAAGACUUGCGGUAUUUGCGGUAACUACAACC
AGGAUUUCAGUGAUGAUUCUUUUGAUGCUGAAGGAGCCUGUGAUCUGACCCCCAACCCACCG
GGAUGCACCGAAGAACAGAAACCUGAAGCUGAACGACUCUGCAAUAGUCUCUUCGCCGGUCA
AAGUGAUCUUGAUCAGAAAUGUAACGUGUGCCACAAGCCUGACCGUGUCGAACGAUGCAUGU
ACGAGUAUUGCCUGAGGGGACAACAGGGUUUCUGUGACCACGCAUGGGAGUUCAAGAAAGAA
UGCUACAUAAAGCAUGGAGACACCCUAGAAGUACCAGAUGAAUGCAAAUAAAUGCUCGG**A**CUC
CGG**A**CUGG**A**CUCGG**A**CUAAUGCAGAGUAGAGGCCGCAGGAUAGUUUGGAACAUGG**A**CUCUA
GGACUGG**A**CUUUGG**A**CUUGAUGAGCGGCCGCGGGGAUCCAGACAUGA

Cluc–*Socs2* (HEK293T):

AUGAAGACCUUAAUUCUUGCCGUUGCAUUAGUCUACUGCGCCACUGUUCAUUGCCAGGACUG
UCCUUACGAACCUGAUCCACCAAACACAGUUCCAACUUCCUGUGAAGCUAAAGAAGGAGAAUG
UAUUGAUAGCAGCUGUGGCACCUGCACGAGAGACAUACUAUCAGAUGGACUGUGUGAAAAUA
AACCAGGAAAAACAUGUUGUCGAAUGUGUCAGUAUGUAAUUGAAUGCAGAGUAGAGGCCGCA
GGAUGGUUUAGAACAUUCUAUGGAAAGAGAUUCCAGUUCCAGGAACCUGGUACAUACGUGUU
GGGUCAAGGAACCAAGGGCGGCGACUGGAAGGUGUCCAUCACCCUGGAGAACCUGGAUGGA
ACCAAGGGGGCUGUGCUGACCAAGACAAGACUGGAAGUGGCUGGAGACAUCAUUGACAUCGC
UCAAGCUACUGAGAAUCCCAUCACUGUAAACGGUGGAGCUGACCCUAUCAUCGCCAACCCGU
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