Supporting Information

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A

TGTACAAAAAAGCAGGCTTTAAAGGAACCAATTCAGTCGACTGGATCCGGTACCAAGGTCGGGCAGGAAGAGGGCCTATTTCCCATGATT
CCTTCATATTTGCATATACGATACAAGGCTGTTAGAGAGATAATTAGAATTAATTTGACTGTAAACACAAAGATATTAGTACAAAATACGTGA
CGTAGAAAGTAATAATTTCTTGGGTAGTTTGCAGTTTTAAAATTATGTTTTAAAATGGACTATCATATGCTTACCGTAACTTGAAAGTATTTC
GATTTCTTGGCTTTATATATCTTGTGGAAAGGACGAAACACCG**GTCACTGTGAAGCTGCAAT**GTTTTAGAGCTAGAAATAGCAAGTTAAAA
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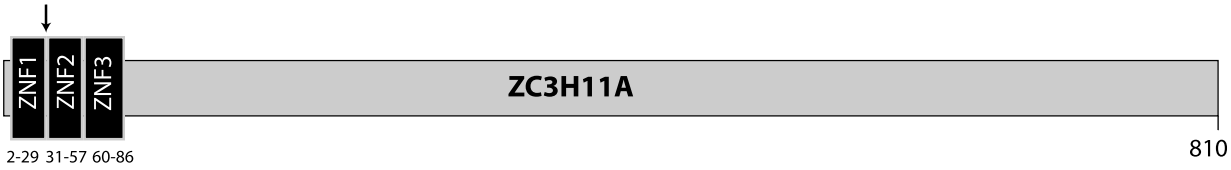
B



C

WT human ZC3H11A

MPNQGEDCYFFFYSTCTKGDSCPFRHCEAAIGNETVCTLWQEGRCFRQVCRFRHMEIDKKRSEIPCYWENQPTGCQKLNCAFHHNRGRYVDG
LFLPPSKTVLPTVPESPEEEVKASQLSVQQNKLSVQSNPSPQLRSVMKVESSENVPSPTHPPVVINAADDDEDDDDQFSEEGDETKTPTLQPTP
EVHNGLRVTSVRKPAVNIKQGECLNFGIKTLEEIKSKKMKEKSKKQGEGSSGVSSLLLHPEPVPGPEKENVRTVVRTVTLSTKQGEEPLVRLSLTE
RLGKRKFSAGGDSDPPLKRSLAQRLGKKVEAPETNIDKTPKKAQVSKSLKERLGMSADPDNEDATDKVNKVGEIHVKTLEEILLERASQKRGELQ
TKLKTEGPSKTDDSTSGARSSSTIRIKTFSEVLAEKKHRQQEAERQKSKKDTTCIKLKIDSEIKKTVVLPPIVASRGQSEEPAGKTKSMQEVHIKTLE
EIKLEKALRVQQSSESSTSSPSQHEATPGARRLLRITKRTGMKEEKNLQEGNEVDSQSSIRTEAKEASGETTGVDITKIQVKRCETMREKHMQKQ
QEREKSVLTPLRGDVASCNTQVAEKPVLTAVPGITRHLTKRLPTKSSQKVEVETSGIGDSLLNVKCAAQTLEKRGKAKPKVNVKPSVVKVVSSPK
LAPKRKAVEMHAAVIAAVKPLSSSSVLQEPPAKKAAVAVVPLVSEDKSVTVPEAENPRDSLVLPPTQSSSDSSPPEVSGPSSSQMSMKTRRLSSA
STGKPPLSVEDDFEKLIWEISGGKLEAEIDLDPGKDEDDLLLELSEMIDS**Stop**

ZC3H11A-KO clone #1, 13bp deletion of nucleotides ATAGGAAATGAAA

MPNQGEDCYFFFYSTCTKGDSCPFRHCEAALFAHYGKKGAVFDRCAGFGTWRLIKNAVKFLVIGKISQQDVKN**Stop**TALSITIEDDMLMAFSYLR
AKLCCPLCLSHQKRK**Stop**RLANFQFSRTNCLSSPILPLSCGAL**Stop**K**Stop**KVPKMFLAPRIHQL**Stop**LMLQMMMKMMMISFLRKVMKPKHLPC
NQLLKFTMDYE**Stop**LLSGNLQSI**Stop**SKVNV**Stop**ILE**Stop**KLLRKLSQRK**Stop**RKNLRSKVRVLQEFPVFYSTLSPFQVLKKKMSGLW**Stop**GQ
StopLSPPNKEKNPWLD**Stop**VLLRDWGNENFQQAVTVILH**Stop**SVAWHRG**Stop**GRKLKLQKLTLTKHQRKLKFPSLLRSD**Stop**ACQLIQIMRMQ
QIKLIKLVRSM**Stop**RH**Stop**KKFFLKEPVRNVENCKLNSRQKDLQKLMILLQEQEAPPLSVSKPSLRSWLKKNIGSRKQRDKKAKRIQLASS**Stop**R
LIVKLKKQ**Stop**FCHPLLPAEDNQRSLQVKQSLCRRCTSRRWKKLNWRRH**Stop**GCSRALRAAPAPRLNTRPLQGQGGCCESPKEQG**Stop**KKR
RTFRKEMKLILRAVLEQKLKRLQVRPQELTSLKFKSRDVRP**Stop**ERSTCRNSRRGKNQS**Stop**HLFGEM**Stop**PLAIPKWQRNQCSLLCQESHGT
StopPSGFPQSHPRRWR**Stop**KPQGLETHY**Stop**M**Stop**NVQHRPWKKGVKLNPK**Stop**T**Stop**SHLWLKLCHPPNWPQNVRQWRCTLLSLPL**Sto**
pSHSAPAVSYRNPQPKRQLWLLSRLSLRTNQSLCLKQKILETVLCCLQPSPLQIPHPRRCLALPHPK**Stop**A**Stop**KLADSALPQQESPHSLWRMIL
RN**Stop**YGRFQEANWKLRLTWILGKMKMTFCLSYQK**Stop**LIA

ZC3H11A-KO clones #2 and #3, 323bp insertion of random nucleotides

MPNQGEDCYFFFYSTCTKGDSCPFRHCEAAKAGFKGTNSVDWIRYQGRAGRGPISHDSFIFAYTIQGC**Stop**RDN**Stop**N**Stop**FDCKHKDISTKY
VT**Stop**KVIISWVVCSFKIMF**Stop**NGLSYAYRNLKVFRFLGFIYLVERTKHRSL**Stop**Q**Stop**EMKLFAHYGKKGAVFDRCAGFGTWRLIKNAVKFLVI
GKISQQDVKNS**top**TALSITIEDDMLMAFSYLRAKLCCPLCLSHQKRK**Stop**RLANFQFSRTNCLSSPILPLSCGAL**Stop**K**Stop**KVPKMFLAPRIHQL
StopLMLQMMMKMMMISFLRKVMKPKHLPCNQLLKFTMDYE**Stop**LLSGNLQSI**Stop**SKVNV**Stop**ILE**Stop**KLLRKLSQRK**Stop**RKNLRSKVRVL
QEFPVFYSTLSPFQVLKKKMSGLW**Stop**GQ**Stop**LSPPNKEKNPWLD**Stop**VLLRDWGNENFQQAVTVILH**Stop**SVAWHRG**Stop**GRKLKLQKLTL
TKHQRKLKFPSLLRSD**Stop**ACQLIQIMRMQQIKLIKLVRSM**Stop**RH**Stop**KKFFLKEPVRNVENCKLNSRQKDLQKLMILLQEQEAPPLSVSKPSL
RSWLKKNIGSRKQRDKKAKRIQLASS**Stop**RLIVKLKKQ**Stop**FCHPLLPAEDNQRSLQVKQSLCRRCTSRRWKKLNWRRH**Stop**GCSRALRAAPA
PRLNTRPLQGQGGCCESPKEQG**Stop**KKRRTFRKEMKLILRAVLEQKLKRLQVRPQELTSLKFKSRDVRP**Stop**ERSTCRNSRRGKNQS**Stop**HL
FGEMS**top**PLAIPKWQRNQCSLLCQESHGT**Stop**PSGFPQSHPRRWR**Stop**KPQGLETHY**Stop**M**Stop**NVQHRPWKKGVKLNPK**Stop**T**Stop**SHL
WLKLCHPPNWPQNVRQWRCTLLSLPL**Stop**SHSAPAVSYRNPQPKRQLWLLSRLSLRTNQSLCLKQKILETVLCCLQPSPLQIPHPRRCLALPHP
K**Stop**A**Stop**KLADSALPQQESPHSLWRMILRN**Stop**YGRFQEANWKLRLTWILGKMKMTFCLSYQK**Stop**LIA

Fig. S1. (*A*) Sequence of the gRNA expression fragment. The specific *ZC3H11A* gRNA sequence is shown in bold. (*B*) Schematic description of the ZC3H11A protein showing the location of the three zinc fingers. The arrow indicates the corresponding location of the gRNA in the ZC3H11A protein. (*C*, *Top*) Amino acid sequence of the WT human ZC3H11A protein. (*C*, *Middle* and *Bottom*) Amino acid sequences of the ZC3H11A proteins expressed in KO clones 1–3. The red-highlighted sequences represent the WT ZC3H11A sequence, whereas the yellow-highlighted sequences represent the short out-of-frame added peptide sequences caused by the deletion of 13 bp or the insertion of 323 bp. "Stop" indicates a translational stop codon in the ORF.

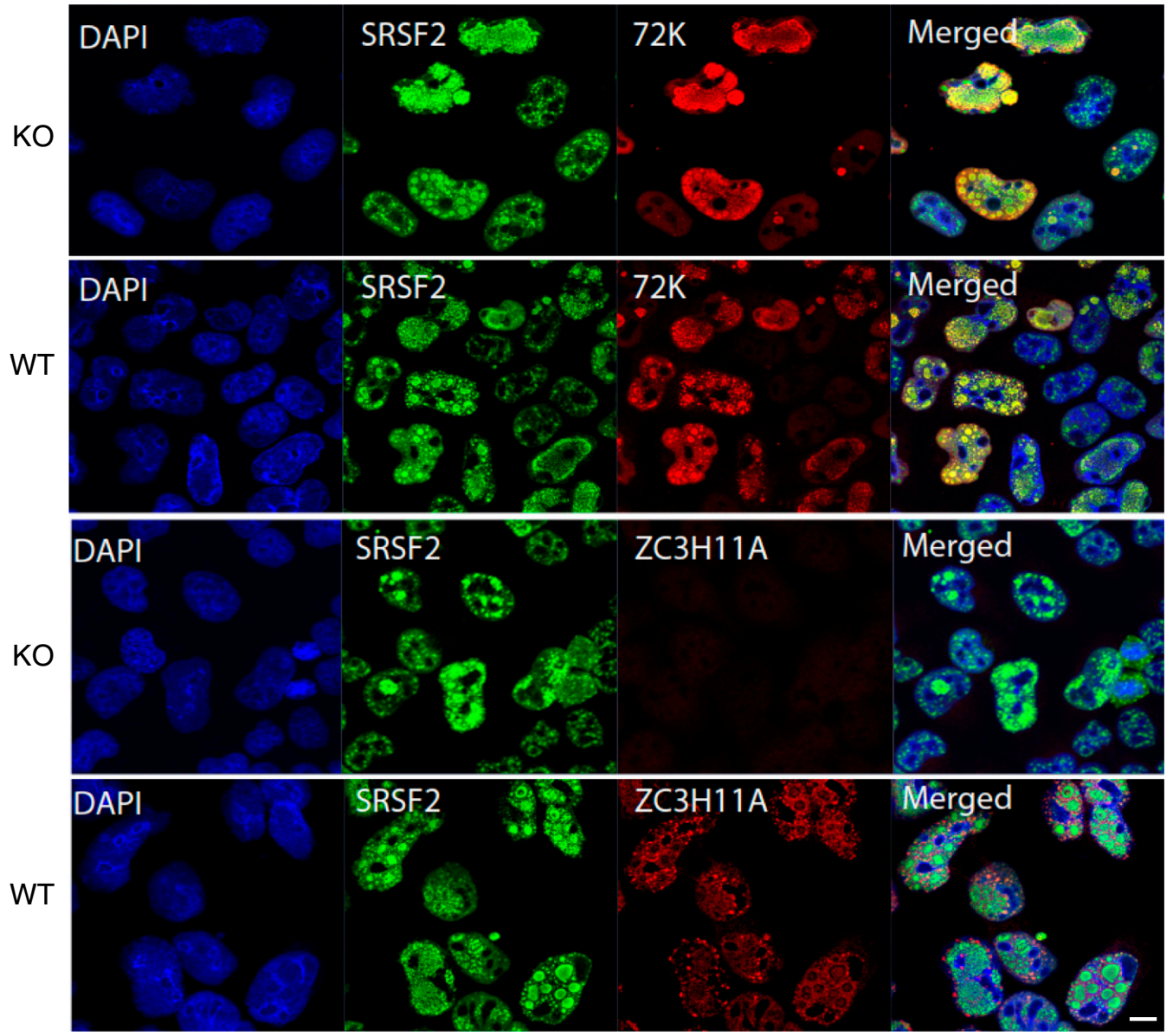


Fig. S2. Immunofluorescence staining using anti-SRSF2 and anti-72K antibodies in HeLa WT and ZC3H11A-KO cells infected with HAdV-5 virus showing that the formation of viral replication centers is not affected by *ZC3H11A* knockdown. (Scale bar: 5 μm.)

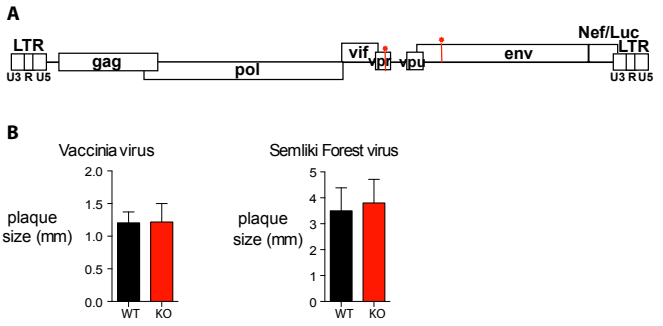


Fig. S3. (*A*) Schematic diagram of the HIV-1 pNL4-Luc. The firefly luciferase gene was inserted into the pNL4-3 Nef gene. Two frameshift mutations in the *Env* and *Vpr* coding sequences (indicated with a red line) render this clone *Env*$^-$ and *Vpr*$^-$ (13). (*B*) Plaque sizes (mean ± SEM) formed by vaccinia virus Western Reserve strain and Semliki Forest virus infections in HeLa WT and ZC3H11A-KO cells.

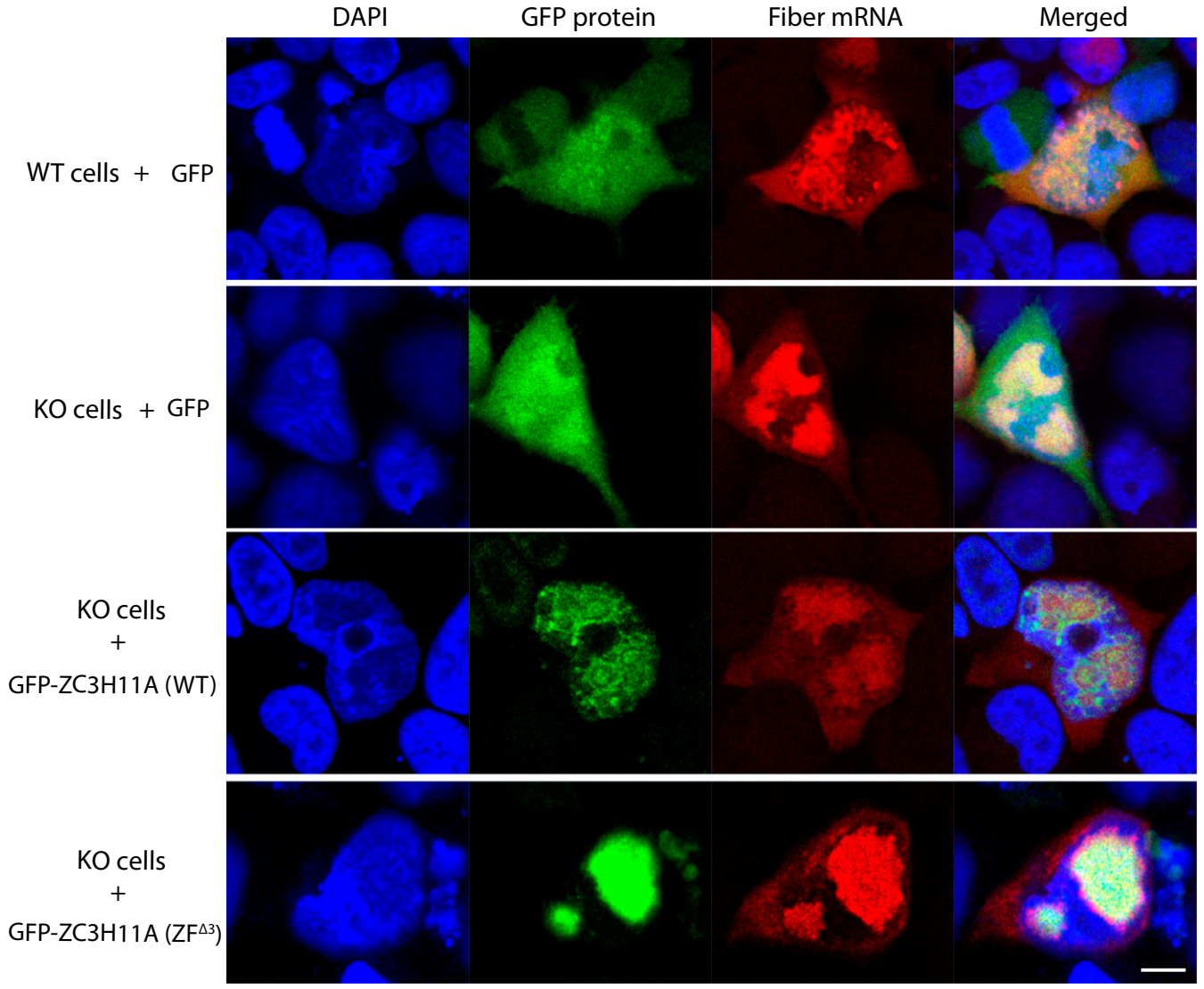


Fig. S4. RNA-FISH of fiber mRNA in WT and ZC3H11A-KO cells transfected with plasmids expressing full-length GFP-ZC3H11A or the mutant GFP-ZC3H11A protein lacking zinc fingers (ZF$^{\Delta 3}$). The GFP-expressing plasmid construct was used as a control. GFP expression was measured by fluorescence microscopy. (Scale bar: 6 μm.)

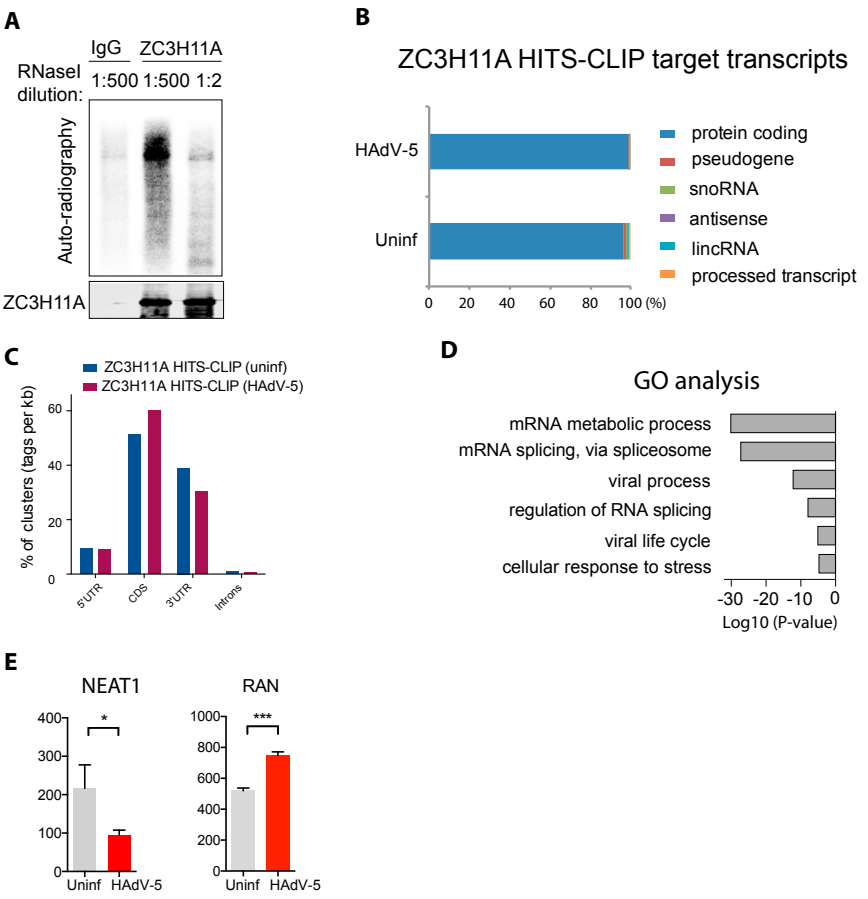


Fig. S5. (*A*) SDS/PAGE of cross-linked ^{32}P-labeled RNA/protein complexes obtained after IgG or ZC3H11A immunoprecipitation and treatment with RNaseI (dilutions: 1:500 and 1:2). (*B*) Distribution of ZC3H11A HITS-CLIP targets in uninfected (Uninf) and HAdV-5–infected cells. snoRNA, small nucleolar RNA. (*C*) Distribution of the proportion of ZC3H11A HITS-CLIP mapped reads (tags per kilobase) over the various elements of a gene in Uninf and HAdV-5–infected cells. (*D*) GO analysis of the endogenous gene targets of ZC3H11A shared before and after HAdV-5 infection. Gray bars, multiple testing corrected *P* values for enriched GO categories. (*E*) Expression of the *NEAT1* and *RAN* mRNAs in Uninf and HAdV-5–infected HeLa cells (***$P < 0.001$, *$P < 0.05$).

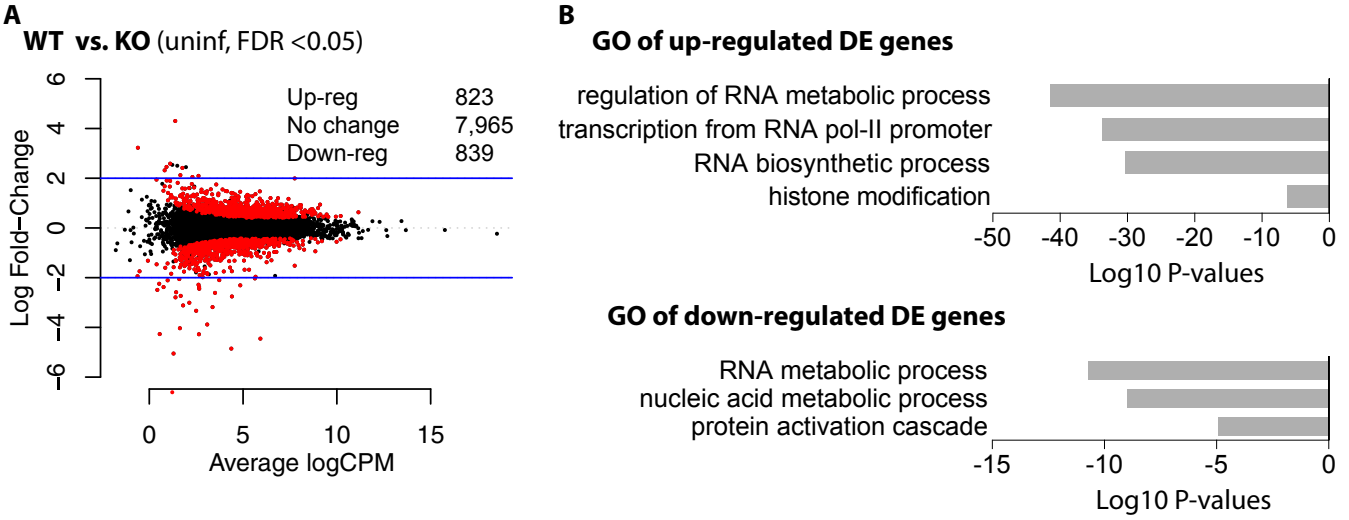
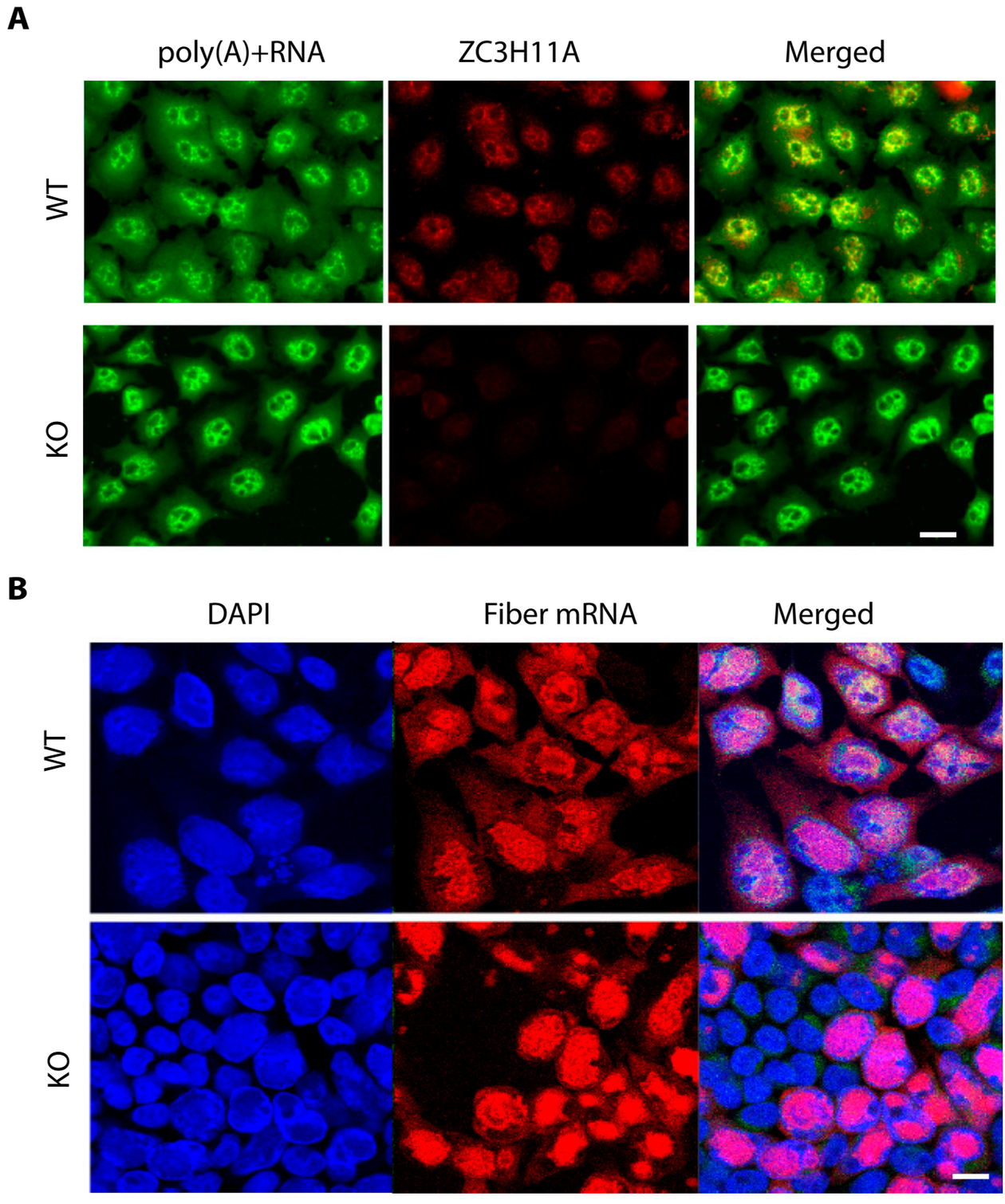


Fig. S6. Results of RNA-seq analysis of uninfected (uninf) WT and ZC3H11A-KO cells. (*A*) Smear plot of log-fold changes against expression levels measured as the log count per million (logCPM) of cytoplasmic mRNA in WT vs. ZC3H11A-KO cells. The lines indicate twofold changes, and red dots indicate significant fold change [false discovery rate (FDR) < 0.05]. reg, regulated. (*B*) GO analysis of significantly up- and down-regulated transcripts in uninf WT and ZC3H11A-KO cells. Gray bars, multiple testing corrected *P* values for enriched GO categories.



C Fiber probe sequence

TTGAAGGTATCTTCAGACGGTCTTGCGCGCTTCATTGCGACTG
TGACTGGTTAGACGCCTTTCTCGAGAG

Fig. S7. (*A*) RNA-FISH detecting poly(A)$^+$ RNA accumulation in WT and ZC3H11A-KO cells. (Scale bar: 15 μm.) (*B*) RNA-FISH of fiber mRNA accumulation in WT and ZC3H11A-KO cells. (Scale bar: 7 μm.) (*C*) Sequence of the FISH probe used to detect the fiber mRNA.

Table S1. List of antibodies

Antibody	Type	Host	Dilution	Application	Source
ZC3H11A	Primary	Rabbit	1:3,000	WB	Abcam
ZC3H11A	Primary	Rabbit	1:500	IF	Human Protein Atlas
ZC3H11A	Primary	Rabbit	4–5 μg	IP	Human Protein Atlas
GFP	Primary	Rabbit	1:1,000	WB	Novus Biologicals
HAdV-5 capsid proteins	Primary	Rabbit	1:5,000	WB	Abcam
VP16	Primary	Rabbit	1:5,000	WB	Abcam
VP5	Primary	Rabbit	1:5,000	WB	Abcam
β-Actin	Primary	Goat	1:10,000	WB	Abcam
Influenza A M2 (14C2)	Primary	Mouse	1:200	WB	Abcam
Influenza A NP (5D8)	Primary	Mouse	1:200	WB	Santa Cruz Biotechnology
ZBED6	Primary	Rabbit	1:2,000	WB	In-house
HSP-70	Primary	Mouse	1:1,000	WB	Invitrogen
SUMO 2+3	Primary	Mouse	2–5 μg	IP	Abcam
HIV1 (p55 + p24 + p17)	Primary	Rabbit	1:1,000	WB	Abcam
Anti-mouse IgG Alexa Fluor-488	Secondary	Goat	1:500	IF	Invitrogen
Anti-rabbit IgG Alexa Fluor-594	Secondary	Goat	1:500	IF	Invitrogen
Fluorescent anti-rabbit IgG (green)	Secondary	Goat	1:10,000	WB	LI-COR
Fluorescent anti-mouse IgG (red)	Secondary	Goat	1:10,000	WB	LI-COR

IF, immunofluorescence; IP, immunoprecipitation; WB, Western blot.

Table S2. Primer sequences

ID	Primer	Sequences
1	ZBED6 (forward)	TCTTTCATGTTGACCCCCAGTA
2	ZBED6 (reverse)	CTGACGCTTTTCTCACAGAGGTT
3	ZC3H11A (forward)	AAGGAAGGACTTACCCATTTTGATATT
4	ZC3H11A (reverse)	TGGGTCAGATTTCCCTATGAGAA
5	β-Actin (forward)	GCAAAGACCTGACGCCAAC
6	β-Actin (reverse)	ACATCTGCTGGAAGGTGGAC
7	Gag/Pol (forward)	TTCTTCAGAGCAGACCAGAGC
8	Gag/Pol (reverse)	GCTGCCAAAGAGTGATCTGA
9	Tat1 (forward)	AGGGGCGGCGACTGAATTGGGT
10	Tat1 (reverse)	CTCGGGATTGGGAGGTGGGT
11	Renilla (forward)	GATGGATTGCACGCAGGTTC
12	Renilla (reverse)	CAGCCGATTGTCTGTTGTGC

Dataset S1. Results of HITS-CLIP analysis

Dataset S1