

Plant mitochondrial RNA editing factors can perform targeted C-to-U editing of nuclear transcripts in human cells

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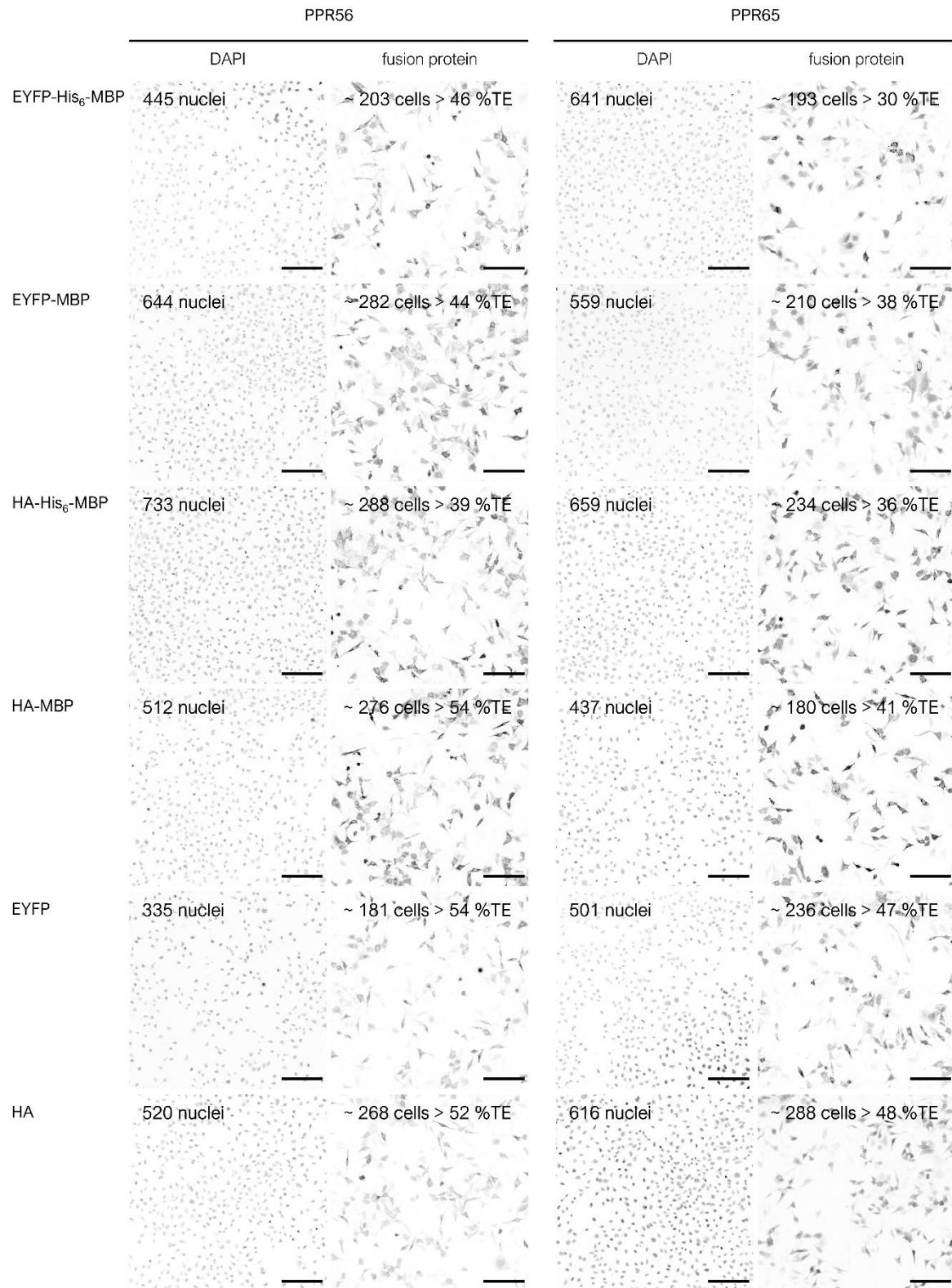
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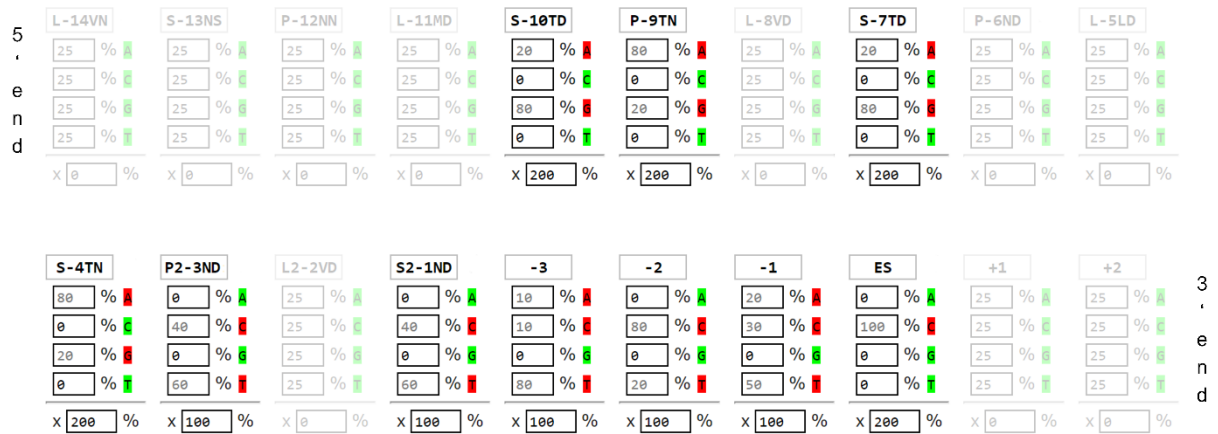
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Supplementary Material



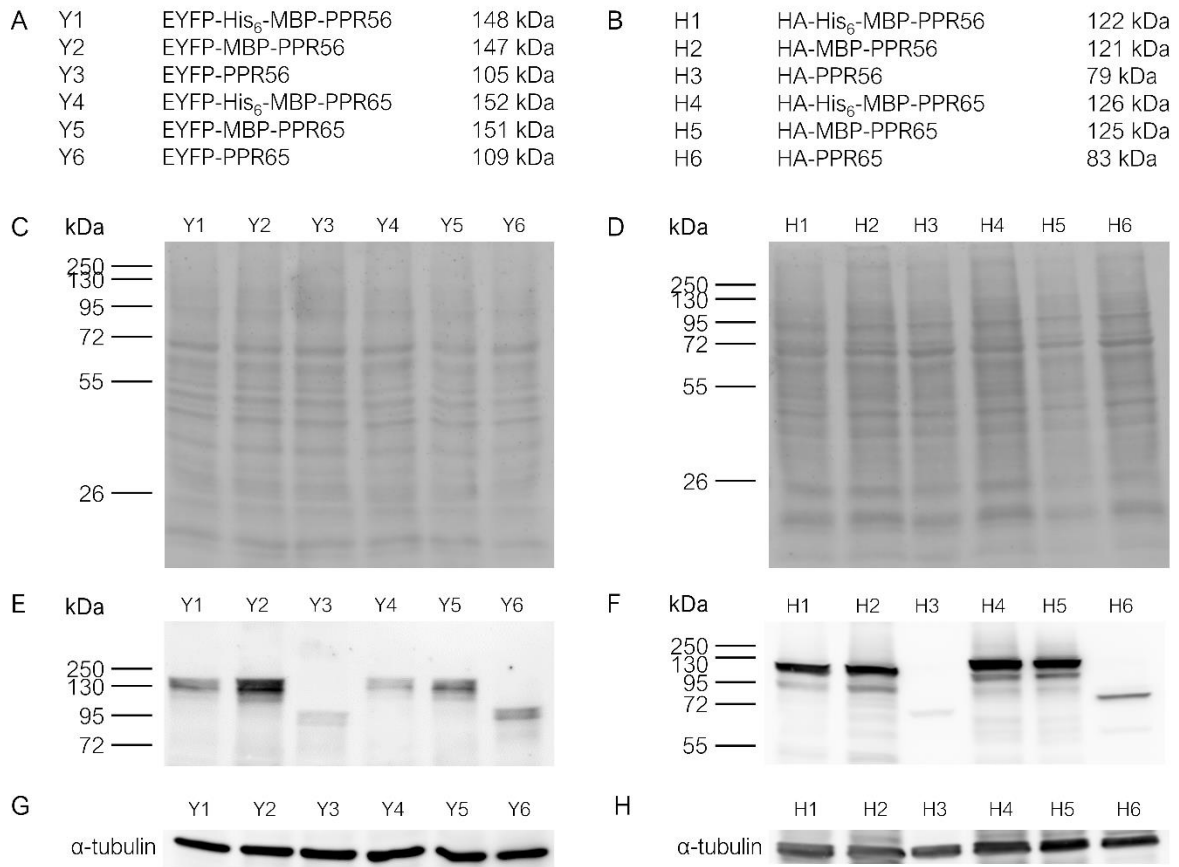
Supplementary Figure S1. Transfection rate calculation via HA and EYFP immunostained fusion proteins per DAPI stained nuclei.

After fixation and immunostaining of HeLa cells, expressing N-terminally tagged PPR56 or PPR65, 960.93x960.93 μm overview images were taken with a Nikon Eclipse Ti2 system equipped with a PlanFluor 40x Oil objective (NA 1.3) and 1.5x Zoom using the NIS-Elements AR software. In ImageJ/Fiji version 1.53c for Windows, the color channels were split. The DAPI image was transformed into a B&W binary and close nuclei were separated using the 'Watershed' and counted using the 'Analyze Particles' function with a size threshold of 1000 μm^2 . Alexa594 fluorescence was displayed as inverted grayscale for counting fusion protein expressing cell. Transfection efficiencies (%TE) were defined as the number of fusion proteins expressing cells per total nuclei. Scale bars = 200 μm .



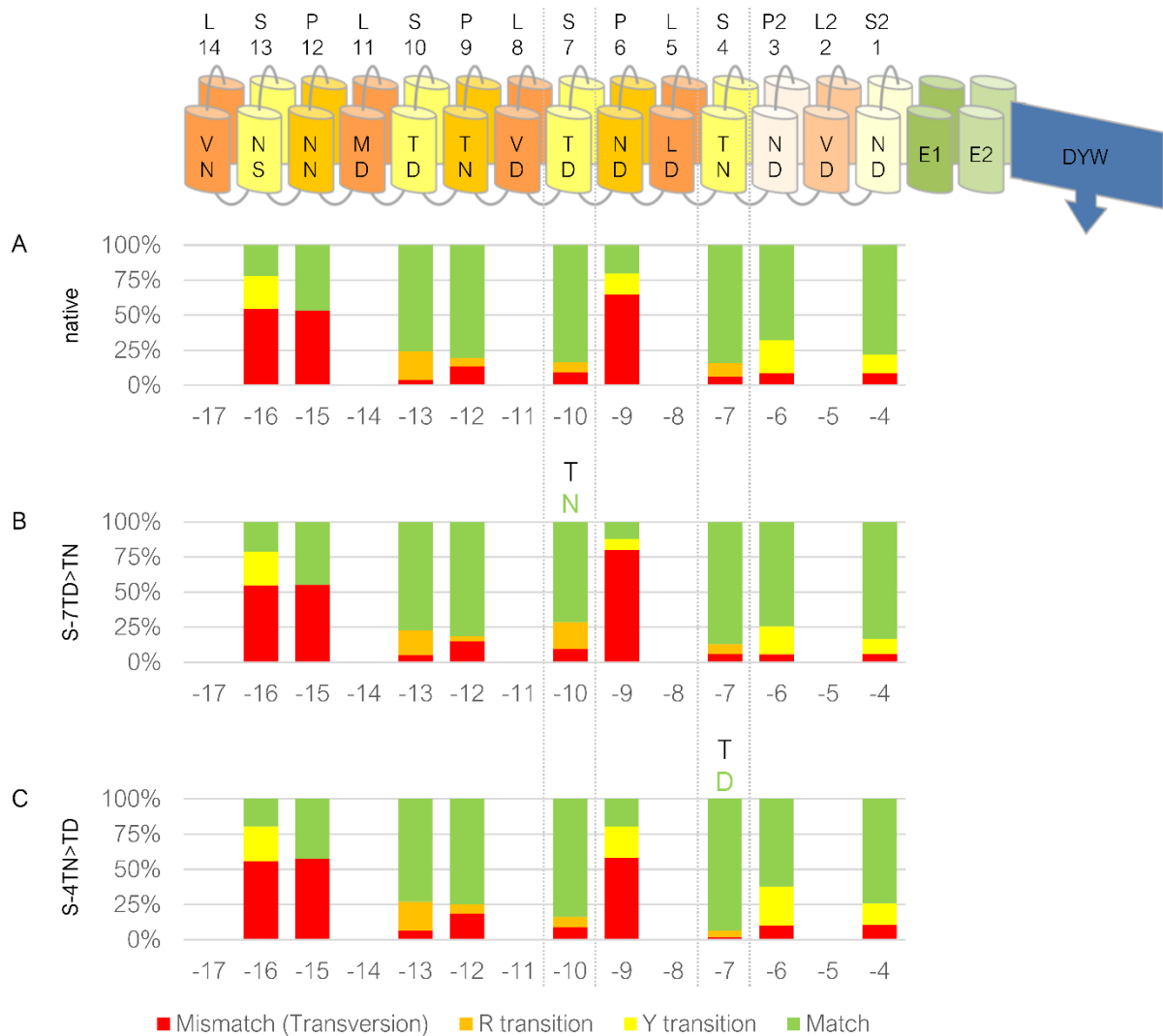
Supplementary Figure S2. Weight matrix used for scanning the custom IMR-90 transcriptome for putative PPR56 editing sites by the TargetScan tool of PREPACT (1).

Each column corresponds to one nucleotide position and is labelled either (i) with type, position counting from S2, and fifth and last amino acid of the corresponding PPR or (ii) with the position relative to the editing site (ES). Nucleotide distributions were set according to the published PPR-RNA binding code (2) and specific knowledge about PPR56 off-target sequence conservation (3). Positional weights are shown below. Positions with no given weight (0%) and no influence on target selection are shaded in light grey.



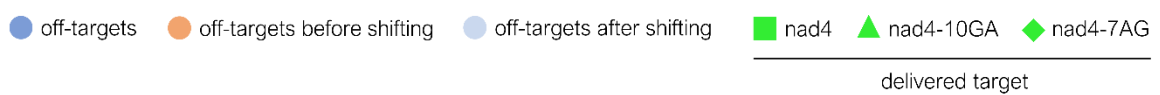
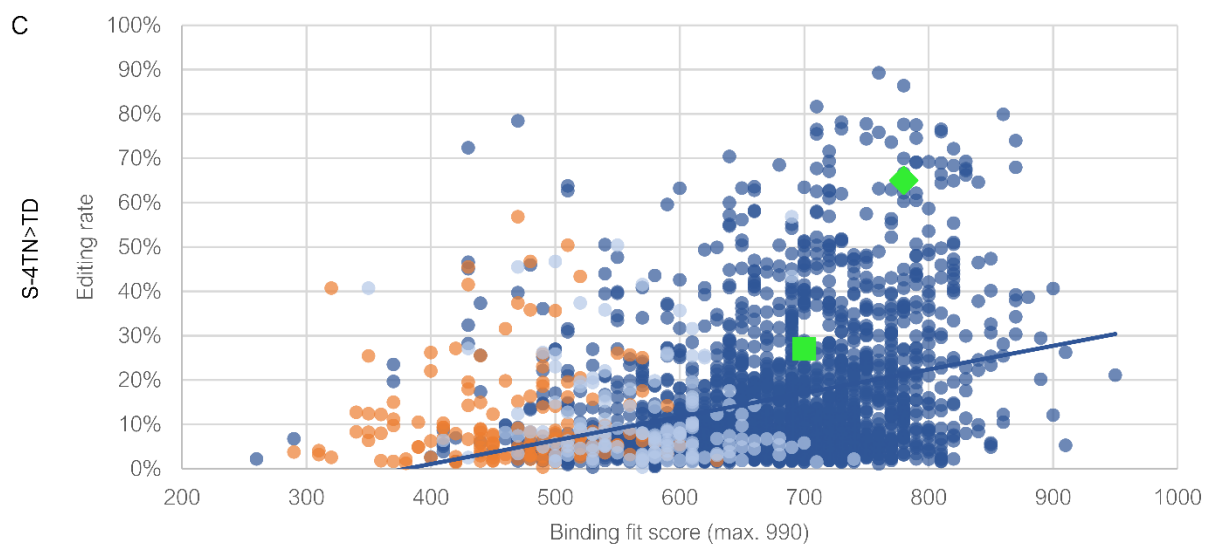
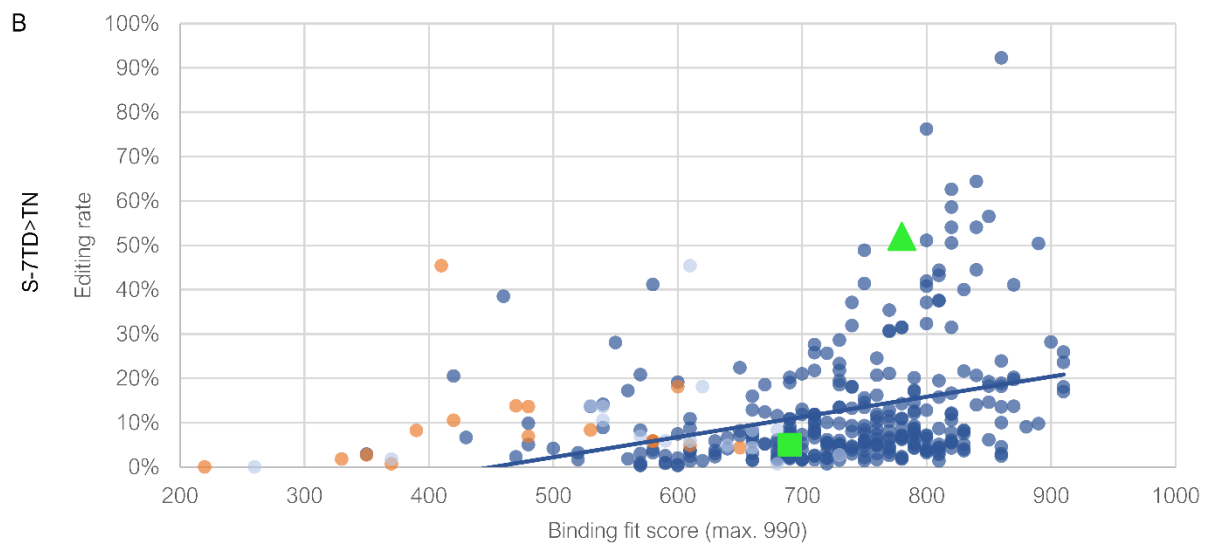
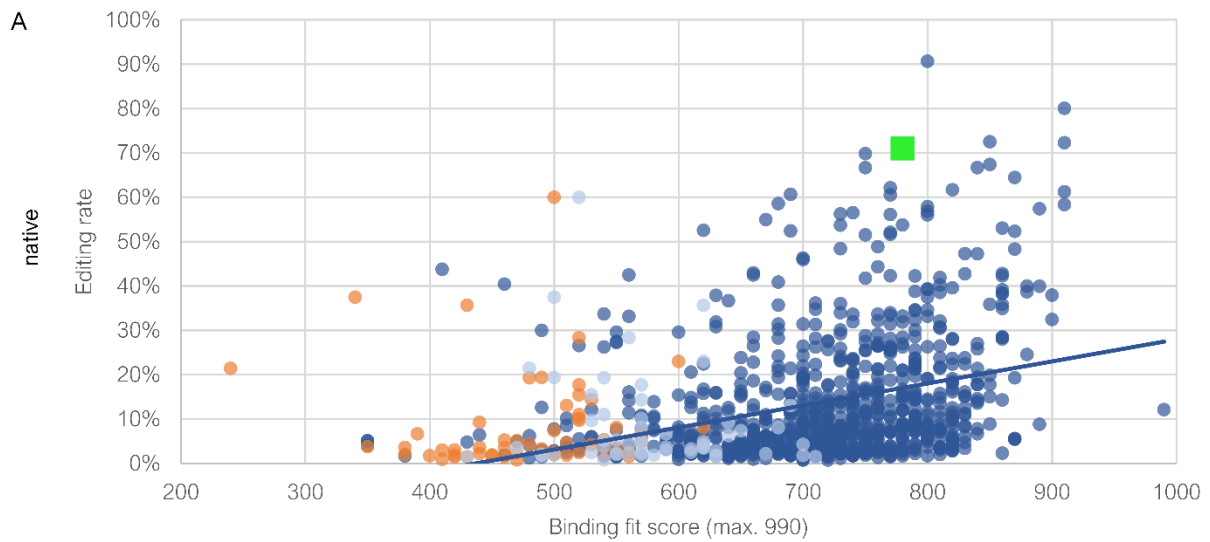
Supplementary Figure S3. Western Blot analyses of fusion protein expression in HeLa cells.

A, B: Investigated fusion proteins and corresponding molecular weights in kDa (calculated with BCCM Genecorner, https://www.genecorner.ugent.be/protein_mw.html). C, D: Total protein stained with Ponceau. E: Immunodetection of EYFP-tagged PPR proteins with α -GFP (mouse, Roth) and peroxidase labeled secondary antibody (Jackson ImmunoResearch). F: Immunodetection of HA-tagged PPR proteins with α -HA (rabbit, abcam ab91110) and peroxidase labeled secondary antibody (Jackson ImmunoResearch). G, H: Immunodetection of α -tubulin with α - α -tubulin (rabbit, Cell Signaling cat. #2125) and peroxidase labeled secondary antibody (Jackson ImmunoResearch) as loading control.



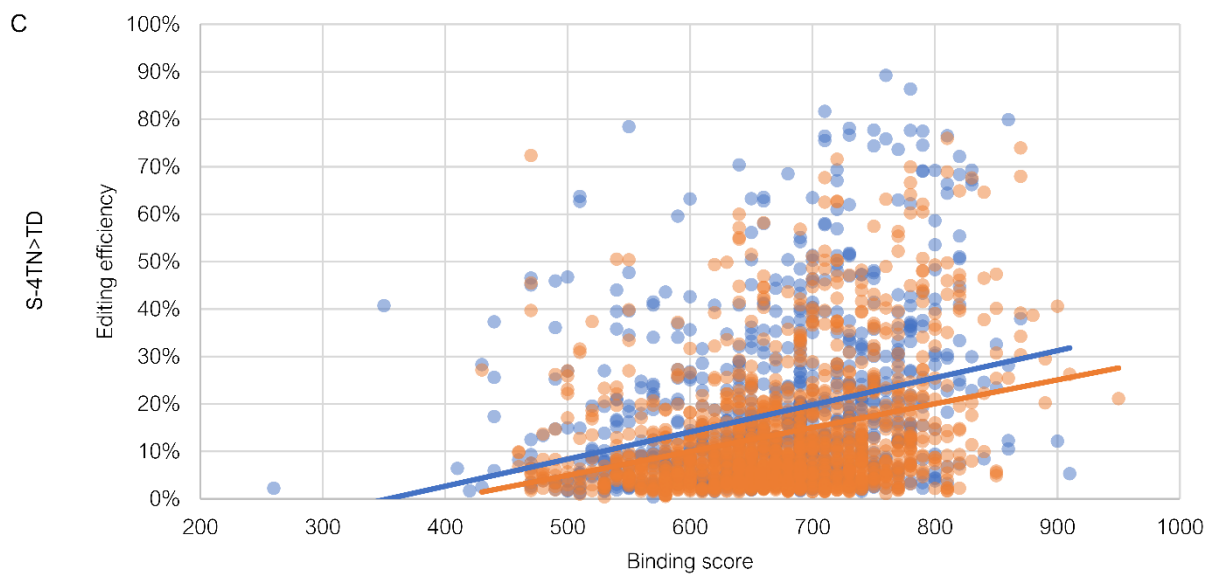
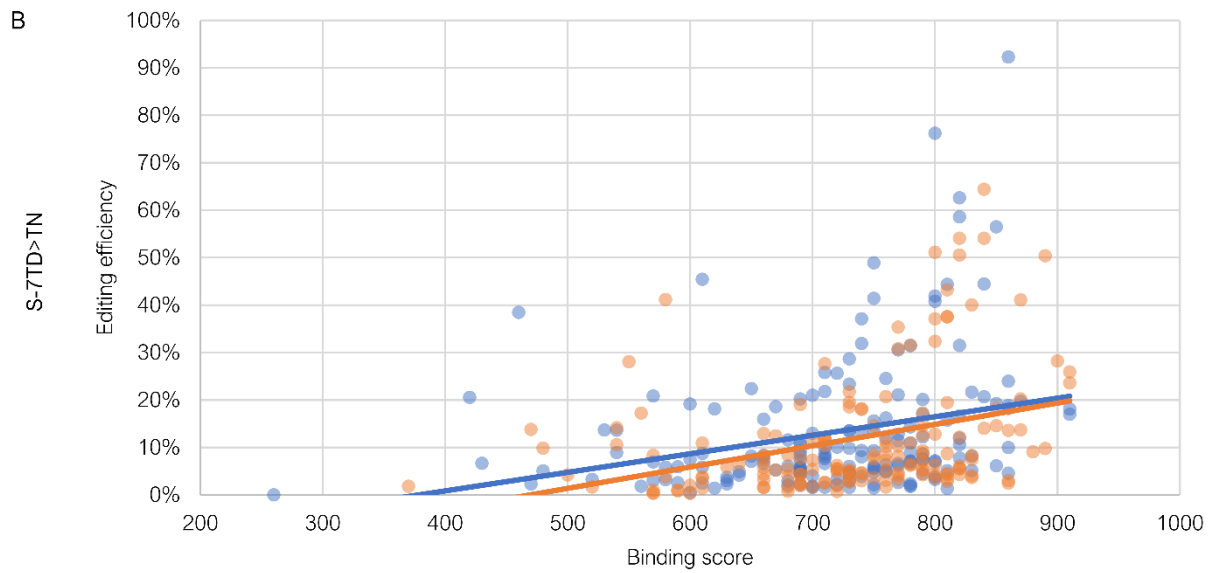
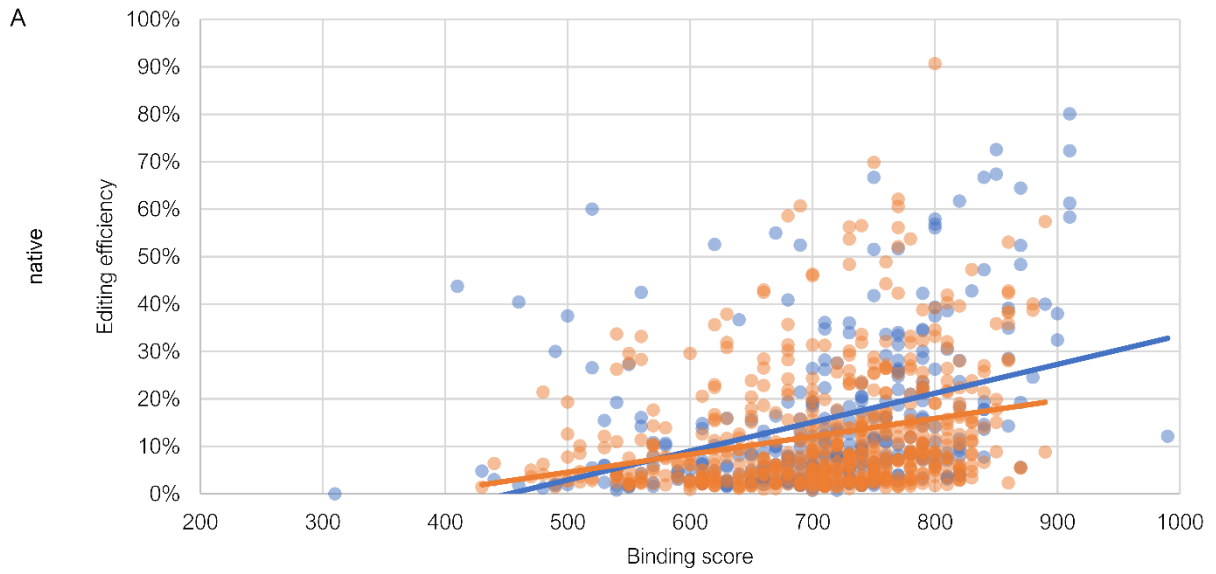
Supplementary Figure S4. Matches of individual PPRs and their corresponding nucleotides for the off-target data sets of PPR56 and the two investigated mutants S-4TN-TD and S-7TD-TN

The bar charts show the relative amounts of conceptual matches between PPRs and their target nucleotides (green), pyrimidine and purine transitions (yellow and orange, respectively) and transversion mismatches (red) at the respective positions observed for the off-targets determined for the native PPR56 protein (A) and the two investigated mutants (B and C).



Supplementary Figure S5. Scatter Plots correlating RNA editing efficiencies with the conceptual overall binding score at the off-targets for wild-type and the two mutant variants of PPR56.

Scatter plot of RNA editing efficiencies on the y-axis displayed in relation to conceptual matching scores (as described in figure 9) on the x-axis for the off-targets (dark blue dots) determined for the native PPR56 protein (A) and the two investigated mutants (B and C). Better matching scores are observed for ca. 5-6 % of off-targets (orange dots; 59 of 906 for PPR56, 16 of 323 for the S-7TN mutant and 147 of 2265 for the S-4TD mutant, respectively) after one- or two-nucleotide-shifts (light blue dots), away from the canonical juxtaposition of the terminal S2-type PPR with position -4 upstream of the editing sites. Editing at the co-delivered targets (green symbols) are added for reference.



● off-target in non-coding region ● off-targets in coding region

Supplementary Figure S6. Scatter Plots distinguishing off-targets occurring in coding vs. non-coding transcript regions.

Off-targets as analyzed in supplementary figure S5 (after nucleotide shifting for improved matching scores where applicable) classified by their occurrence in coding sequences (orange) or in non-coding regions (blue).

Supplementary Table S1

Oligonucleotides/ gBlocks used in this study.

#	Name	Purpose/ target	Sequence (5'-3')
P1	PPR56_P9_R	Cloning of PPR56 S motif mutant versions	GTTTGAATCCTTCCTGTTCC
P2	PPR56_P6_F	Cloning of PPR56 S motif mutant versions	GGCATGAAGCATATGATCTG
P3	PPR56_P6_R	Cloning of PPR56 S motif mutant versions	CAGATCATATGCTTCATGCC
P4	PPR56_P3_F	Cloning of PPR56 S motif mutant versions	GCTTGGCACAACATGGTC
P5	Gw_PPR56_-n_for	Cloning PPR into petG41K	GGGGACAAGTTTGTACAAAAAAGCAGGCTTAGAGAATCTTTATTTTCAGGGCGTGGTTCTGATGAACCGTC
P6	Gw_PPR56_-n_rev	Cloning PPR into petG41K	GGGGACCACCTTTGTACAAGAAAGCTGGGTCTCACCAGTAATCACCACATG
P7	175641Ppfor	Cloning PPR into petG41K	GGGGACAAGTTTGTACAAAAAAGCAGGCTTAGAGAATCTTTATTTTCAGGGCTCTCAGATCAGATGCCAGGG
P8	175641Ppcomprev	Cloning PPR into petG41K	GGGGACCACCTTTGTACAAGAAAGCTGGGTCTCACCACAAAAATCCCCACAAGAACATAC
P9	EGFP_MBP_for_Scal	Cloning PPR-target expression cassette	GCCAGTACTCCATGAAAAATCGAAGAAGG
P10	EGFP_TEV_for_Scal	Cloning PPR-target expression cassette	GCCAGTACTTAGAGAATCTTTATTTTCAGG
P11	EGFP_tT7_rev_Apal	Cloning PPR-target expression cassette	ATGGGCCCTAGTTATTGCTCAGCGGTGG
P12	CMV_His_for_Mscl	Cloning PPR-target expression cassette	ATTATATGGCCAAACATCACCATCACCATCACC
P13	CMV_MBP_for_Mscl	Cloning PPR-target expression cassette	ATTATATGGCCACCATGAAAAATCGAAGAAGG
P14	CMV_TEV_for_Mscl	Cloning PPR-target expression cassette	ACTATATGGCCATAGAGAATCTTTATTTTCAGG
P15	CMV_tT7_rev_NotI	Cloning PPR-target expression cassette	ATTATAGCGGCCGCTAGTTATTGCTCAGCGGTGG
P16	pCMV-HA_seq_for	Cloning PPR-target expression cassette	CGGAATTGTACCCGCGGGCC
P17	pCMV-HA_seq_rev	Cloning PPR-target expression cassette	ATTCTAGTTGTGGTTTGTCC
P18	EGFP_His_for_Scal_MOD	Cloning PPR-target expression cassette	GCCAGTACTTACATCACCATCACCATCACC
P19	PPR56_4-7ag_F	Cloning target	AAATATCAATTTTTATATAGgtatagacggtGtctcttcATTTTGG
P20	PPR56_4-7ag_R	Cloning target	CGCGCCAAAATGAAGAGACACCGTCTATACCTATATAAAAAATTGATATT
P21	PPR56_4-10ga_F	Cloning target	AAATATCAATTTTTATATAGgtatagacAgtatctcttcATTTTGG
P22	PPR56_4-10ga_R	Cloning target	CGCGCCAAAATGAAGAGATACTGTCTATACCTATATAAAAAATTGATATT
P23	PPR56_nad4_F	Cloning target	TCAAACATCAATTTTTATATAGgtatagacggtatctcttcATTTT
P24	PPR56_nad4_R	Cloning target	CGCGAAAATGAAGAGATACCGTCTATACCTATATAAAAAATTGATGTTTGA
P25	HindIIIccmFC_for	Cloning target	agcttATACTATTTCAATGGTTGGTAAGTAGAGATGTTCCACAtt
P26	Bsp119iccmFC_rev	Cloning target	CGAATGTGGGAACATCTCTACTTACCAACCATTGAAATAGTATA
P27	petG_30K40Kseqrev	Editing analysis/ Sequencing	GGTTATGCTAGTTATTGCTCAGC
P28	PPR56_CTDH_R	PPR mutagenesis DYW domain	CTTAGTTGCTCCATGAGCATCGGTGCACACCCG
P29	PPR56_CTDH_F	PPR mutagenesis DYW domain	CGGGTGTGCACCCGATGCTCATGGAGCAACTAAG
P30	PPR65_HSARLAfor	PPR mutagenesis DYW domain	CTTTTCTCTCACAGTGCGAGGCTGGCGATCAC
P31	PPR65_HSARLArev	PPR mutagenesis DYW domain	GTGATCGCCAGCCTCGCACTGTGAGAGAAAAG
P32	PPR65_CGDAH_for	PPR mutagenesis DYW domain	CGAGTGTGTGGGATGCTCATACGGCGACGAAG
P33	PPR65_CGDAH_rev	PPR mutagenesis DYW domain	CTTCGTGCGCGTATGAGCATCCCCACACACTCG
P34	PPR56_HSERLA_F	PPR mutagenesis DYW domain	CTCACAGTGAGaggTTGGCAATTGTG
P35	PPR56_HSERLA_R	PPR mutagenesis DYW domain	CACAATTGCCAACCTCTCACTGTGAG
P36	PPR56_S4TNTD_for	PPR mutagenesis S4 motif (gBlock IDT)	GTGGGCATGAAGCATATGATCTGTTTCTTCAGATGCAGACAGAGGGCTGTAAGCCAGATGCAATCATGTTTTGAGCATCTTGAATGCATGTGCAAGTGC AGGGGCATTGGAGTGGGTGAAGAAGATTACAGGCATGCCTTGATTGAGGGCTTGAGGTTGATGTGCGTGTGGGCACTGCACCTGTTACATGTA CAAAGAGTGGTAGCATTGATGATGCGCGAGTGGTGTGGACAGAATGAAGGTGCGCgATGTTGATCATGGAATGCGATGATTTCTGGCTTGGCACAAC ATGGTCTTGGGCAAGGACGCTTTG
P37	PPR56_S-7TDTN_for	PPR mutagenesis S7 motif (gBlock IDT)	TGGAACAGGAAGGATTCAAACCAATGCAATCACTTATGTGAGCATCTTGAACGCATGTGCAAGTGAAGGGGCACTTGAAGTGGGTGAAGAGGGTTCAC AGGCATGCCTTGGATGCAGGGCTTGGATTGGATGTGCGTGTGGGCACTGCACCTGTTCAAATGTACGCAAAGAGTGGTAGCATTGATGCACGAGT GGTGTGGACAGAATGAAGGTGCGTgATGTTGATCATGGAATGTGATGATAGGCGCATTGACAGAACATGGGCGTGGGCATGAAGCATATGATCTGTT CTTC
P38	PPR56_int_for	PPR56 internal/ Sequencing	TTGAATGCATGTGCAAGTGC
P39	PPR65_intseq_rev	PPR65 internal/ Sequencing	CCAGACTTGCATCCATGC
P40	175641KOfor	PPR65 internal/ Sequencing	ATGGAAATTGTTGAAGCTGGACTGG

P41 PPR65_PAbox_rev	PPR65 internal/ Sequencing	CTTCAATCCAGCTCTGCGCCGGCTCCTTGACAACATCC
P42 PPR65edit_f	PPR65 internal/ Sequencing	TATCTACGCTGCTGCTGGCC
P43 pEGFP-C3_seq_for	Sequencing	GATCACTCTCGGCATGGACG
P44 pEGFP-CR_seq_rev	Sequencing	CTCTACAAATGTGGTATGGC
P45 mbppetG40Kseqfor	Sequencing	GAT GTC CGC TTT CTG GTA TGC
P46 TEVrev	Sequencing	CCTGAAAATAAAGATTCTC
P47 EYFPfor	Sequencing	CACCTTGACCTACGGCGTGC

Supplementary Table S2

Results of all individual RNA editing experiments in the heterologous human cell and *E. coli* setups. The experimental conditions (type and number of cells seeded for transfection [human cells]/ of cells induced by addition of 0.4 mM isopropyl β -D-1-thiogalactopyranoside (IPTG) [*E. coli*], incubation time after transfection [human cells]/ after induction [*E. coli*], media and zinc supplementation) and C-U RNA editing rate of each individual replicate is listed.

#	Organism	Experiment	Cell line	Construct	Incubation	Cell number	Media	Zinc	Editing	Study
1	Human	Cell line	HEK-293	HA-His ₆ -MBP-PPR56+nad4_native	20h	2x10 ⁵	DMEM+FCS+Penstrep	25 μ M	46%	This study
2	Human	Cell line	HEK-293	HA-His ₆ -MBP-PPR56+nad4_native	20h	2x10 ⁵	DMEM+FCS+Penstrep	25 μ M	57%	This study
3	Human	Cell line	HEK-293	HA-His ₆ -MBP-PPR56+nad4_native	20h	2x10 ⁵	DMEM+FCS+Penstrep	25 μ M	50%	This study
4	Human	Cell line	HEK-293	HA-His ₆ -MBP-PPR65+ccmFC_native	20h	2x10 ⁵	DMEM+FCS+Penstrep	25 μ M	34%	This study
5	Human	Cell line	HEK-293	HA-His ₆ -MBP-PPR65+ccmFC_native	20h	2x10 ⁵	DMEM+FCS+Penstrep	25 μ M	39%	This study
6	Human	Cell line	HEK-293	HA-His ₆ -MBP-PPR65+ccmFC_native	20h	2x10 ^b	DMEM+FCS+Penstrep	25 μ M	36%	This study
7	Human	Cell line	HEK-293	EYFP-PPR56+nad4_native	20h	2x10 ⁵	DMEM+FCS+Penstrep	25 μ M	50%	This study
8	Human	Cell line	HEK-293	EYFP-PPR56+nad4_native	20h	2x10 ⁵	DMEM+FCS+Penstrep	25 μ M	52%	This study
9	Human	Cell line	HEK-293	EYFP-PPR56+nad4_native	20h	2x10 ⁵	DMEM+FCS+Penstrep	25 μ M	55%	This study
10	Human	Cell line	HEK-293	EYFP-PPR65+ccmFC_native	20h	2x10 ⁵	DMEM+FCS+Penstrep	25 μ M	17%	This study
11	Human	Cell line	HEK-293	EYFP-PPR65+ccmFC_native	20h	2x10 ⁵	DMEM+FCS+Penstrep	25 μ M	13%	This study
12	Human	Cell line	HEK-293	EYFP-PPR65+ccmFC_native	20h	2x10 ⁵	DMEM+FCS+Penstrep	25 μ M	13%	This study
13	Human	Cell line	IMR-90	HA-His ₆ -MBP-PPR56+nad4_native	20h	1.8x10 ⁵	MEM+FCS+Penstrep	25 μ M	59%	This study
14	Human	Cell line	IMR-90	HA-His ₆ -MBP-PPR56+nad4_native	20h	1.8x10 ⁵	MEM+FCS+Penstrep	25 μ M	60%	This study
15	Human	Cell line	IMR-90	HA-His ₆ -MBP-PPR56+nad4_native	20h	1.8x10 ⁵	MEM+FCS+Penstrep	25 μ M	61%	This study
16	Human	Cell line	IMR-90	HA-His ₆ -MBP-PPR65+ccmFC_native	20h	1.8x10 ⁵	MEM+FCS+Penstrep	25 μ M	33%	This study
17	Human	Cell line	IMR-90	HA-His ₆ -MBP-PPR65+ccmFC_native	20h	1.8x10 ^b	MEM+FCS+Penstrep	25 μ M	30%	This study
18	Human	Cell line	IMR-90	HA-His ₆ -MBP-PPR65+ccmFC_native	20h	1.8x10 ⁵	MEM+FCS+Penstrep	25 μ M	30%	This study
19	Human	Cell line	IMR-90	EYFP-PPR56+nad4_native	20h	1.8x10 ⁵	MEM+FCS+Penstrep	25 μ M	69%	This study
20	Human	Cell line	IMR-90	EYFP-PPR56+nad4_native	20h	1.8x10 ⁵	MEM+FCS+Penstrep	25 μ M	73%	This study
21	Human	Cell line	IMR-90	EYFP-PPR56+nad4_native	20h	1.8x10 ⁵	MEM+FCS+Penstrep	25 μ M	73%	This study
22	Human	Cell line	IMR-90	EYFP-PPR65+ccmFC_native	20h	1.8x10 ⁵	MEM+FCS+Penstrep	25 μ M	2%	This study
23	Human	Cell line	IMR-90	EYFP-PPR65+ccmFC_native	20h	1.8x10 ⁵	MEM+FCS+Penstrep	25 μ M	2%	This study
24	Human	Cell line	IMR-90	EYFP-PPR65+ccmFC_native	20h	1.8x10 ⁵	MEM+FCS+Penstrep	25 μ M	0%	This study
25	Human	Cell line	MCF-7	HA-His ₆ -MBP-PPR56+nad4_native	20h	2x10 ^b	DMEM+FCS+Penstrep	25 μ M	64%	This study
26	Human	Cell line	MCF-7	HA-His ₆ -MBP-PPR56+nad4_native	20h	2x10 ⁵	DMEM+FCS+Penstrep	25 μ M	71%	This study
27	Human	Cell line	MCF-7	HA-His ₆ -MBP-PPR56+nad4_native	20h	2x10 ⁵	DMEM+FCS+Penstrep	25 μ M	74%	This study
28	Human	Cell line	MCF-7	HA-His ₆ -MBP-PPR65+ccmFC_native	20h	2x10 ^b	DMEM+FCS+Penstrep	25 μ M	61%	This study
29	Human	Cell line	MCF-7	HA-His ₆ -MBP-PPR65+ccmFC_native	20h	2x10 ⁵	DMEM+FCS+Penstrep	25 μ M	63%	This study
30	Human	Cell line	MCF-7	HA-His ₆ -MBP-PPR65+ccmFC_native	20h	2x10 ⁵	DMEM+FCS+Penstrep	25 μ M	63%	This study
31	Human	Cell line	MCF-7	EYFP-PPR56+nad4_native	20h	2x10 ⁵	DMEM+FCS+Penstrep	25 μ M	37%	This study
32	Human	Cell line	MCF-7	EYFP-PPR56+nad4_native	20h	2x10 ⁵	DMEM+FCS+Penstrep	25 μ M	47%	This study
33	Human	Cell line	MCF-7	EYFP-PPR56+nad4_native	20h	2x10 ⁵	DMEM+FCS+Penstrep	25 μ M	44%	This study
34	Human	Cell line	MCF-7	EYFP-PPR65+ccmFC_native	20h	2x10 ⁵	DMEM+FCS+Penstrep	25 μ M	25%	This study

119	Human	S motif mutants	IMR-90	EYFP-PPR56_S7mut+nad4_native	20h	2x10 ⁵	MEM+FCS+Penstrep	25μM	0%	This study
120	Human	S motif mutants	IMR-90	EYFP-PPR56_S7mut+nad4_native	20h	2x10 ⁵	MEM+FCS+Penstrep	25μM	0%	This study
121	Human	S motif mutants	IMR-90	EYFP-PPR56_S4mut+nad4_-7ag	20h	2x10 ⁵	MEM+FCS+Penstrep	25μM	70%	This study
122	Human	S motif mutants	IMR-90	EYFP-PPR56_S4mut+nad4_-7ag	20h	2x10 ⁵	MEM+FCS+Penstrep	25μM	67%	This study
123	Human	S motif mutants	IMR-90	EYFP-PPR56_S4mut+nad4_-7ag	20h	2x10 ⁵	MEM+FCS+Penstrep	25μM	58%	This study
124	Human	S motif mutants	IMR-90	EYFP-PPR56_S7mut+nad4_-10ga	20h	2x10 ⁵	MEM+FCS+Penstrep	25μM	31%	This study
125	Human	S motif mutants	IMR-90	EYFP-PPR56_S7mut+nad4_-10ga	20h	2x10 ⁵	MEM+FCS+Penstrep	25μM	29%	This study
126	Human	S motif mutants	IMR-90	EYFP-PPR56_S7mut+nad4_-10ga	20h	2x10 ⁵	MEM+FCS+Penstrep	25μM	30%	This study
127	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	14h	1x10 ⁵	DMEM+FCS+Penstrep	100μM	39%	This study
128	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	14h	1x10 ⁵	DMEM+FCS+Penstrep	100μM	42%	This study
129	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	14h	1x10 ⁵	DMEM+FCS+Penstrep	100μM	27%	This study
130	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	20h	1x10 ⁵	DMEM+FCS+Penstrep	100μM	51%	This study
131	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	20h	1x10 ⁵	DMEM+FCS+Penstrep	100μM	44%	This study
132	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	20h	1x10 ⁵	DMEM+FCS+Penstrep	100μM	39%	This study
133	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	40h	1x10 ⁵	DMEM+FCS+Penstrep	100μM	45%	This study
134	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	40h	1x10 ⁵	DMEM+FCS+Penstrep	100μM	47%	This study
135	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	40h	1x10 ⁵	DMEM+FCS+Penstrep	100μM	42%	This study
136	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	14h	1x10 ⁵	DMEM+FCS+Penstrep	25μM	40%	This study
137	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	14h	1x10 ⁵	DMEM+FCS+Penstrep	25μM	36%	This study
138	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	14h	1x10 ⁵	DMEM+FCS+Penstrep	25μM	29%	This study
139	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	20h	1x10 ⁵	DMEM+FCS+Penstrep	25μM	45%	This study
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141	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	20h	1x10 ⁵	DMEM+FCS+Penstrep	25μM	32%	This study
142	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	40h	1x10 ⁵	DMEM+FCS+Penstrep	25μM	49%	This study
143	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	40h	1x10 ⁵	DMEM+FCS+Penstrep	25μM	46%	This study
144	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	40h	1x10 ⁵	DMEM+FCS+Penstrep	25μM	47%	This study
145	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	14h	1x10 ⁵	DMEM+FCS+Penstrep	no	42%	This study
146	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	14h	1x10 ⁵	DMEM+FCS+Penstrep	no	30%	This study
147	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	14h	1x10 ⁵	DMEM+FCS+Penstrep	no	35%	This study
148	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	20h	1x10 ⁵	DMEM+FCS+Penstrep	no	43%	This study
149	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	20h	1x10 ⁵	DMEM+FCS+Penstrep	no	54%	This study
150	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	20h	1x10 ⁵	DMEM+FCS+Penstrep	no	36%	This study
151	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	40h	1x10 ⁵	DMEM+FCS+Penstrep	no	48%	This study
152	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	40h	1x10 ⁵	DMEM+FCS+Penstrep	no	62%	This study
153	Human	Zinc/time	HeLa	EYFP-PPR56+nad4_native	40h	1x10 ⁵	DMEM+FCS+Penstrep	no	50%	This study
154	<i>Escherichia coli</i>	S motif mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR56_S4mut+nad4_native	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	98%	This study
155	<i>Escherichia coli</i>	S motif mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR56_S4mut+nad4_native	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	98%	This study
156	<i>Escherichia coli</i>	S motif mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR56_S4mut+nad4_native	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	81%	This study
157	<i>Escherichia coli</i>	S motif mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR56_S7mut+nad4_native	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	0%	This study
158	<i>Escherichia coli</i>	S motif mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR56_S7mut+nad4_native	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	0%	This study
159	<i>Escherichia coli</i>	S motif mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR56_S7mut+nad4_native	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	0%	This study
160	<i>Escherichia coli</i>	S motif mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR56_S7mut+nad4_-10ga	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	0%	This study

161	<i>Escherichia coli</i>	S motif mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR56_S7mut+nad4_-10ga	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	0%	This study
162	<i>Escherichia coli</i>	S motif mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR56_S7mut+nad4_-10ga	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	0%	This study
163	<i>Escherichia coli</i>	S motif mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR56_S4mut+nad4_-7ag	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	100%	This study
164	<i>Escherichia coli</i>	S motif mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR56_S4mut+nad4_-7ag	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	100%	This study
165	<i>Escherichia coli</i>	S motif mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR56_S4mut+nad4_-7ag	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	98%	This study
166	<i>Escherichia coli</i>	DYW domain mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR56_HSERLA+nad4_native	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	79%	This study
167	<i>Escherichia coli</i>	DYW domain mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR56_HSERLA+nad4_native	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	80%	This study
168	<i>Escherichia coli</i>	DYW domain mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR56_HSERLA+nad4_native	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	78%	This study
169	<i>Escherichia coli</i>	DYW domain mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR56_CxDAH+nad4_native	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	0%	This study
170	<i>Escherichia coli</i>	DYW domain mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR56_CxDAH+nad4_native	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	0%	This study
171	<i>Escherichia coli</i>	DYW domain mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR56_CxDAH+nad4_native	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	0%	This study
172	<i>Escherichia coli</i>	DYW domain mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR56+nad4_native	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	100±0%	(3)
173	<i>Escherichia coli</i>	DYW domain mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR65+ccmFC_native	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	96±4%	(3)
174	<i>Escherichia coli</i>	DYW domain mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR65_HSARLA+ccmFC_native	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	0±0%	(3)
175	<i>Escherichia coli</i>	DYW domain mutants	Rosetta 2 (DE3)	His ₆ -MBP-PPR65_CxDAH+ccmFC_native	20h	3.2x10 ⁸ to 4.6x10 ⁸	LB+KanCam	0.4mM	0±0%	(3)

Supplementary Table S3

Transfected human IMR-90 cell lines sent for RNA sequencing. The following information is included: cell line, RNASeq batch, expressed fusion protein, delivered target, replicate number of respective constructs, FACS sorting prior to RNA preparation, amounts of raw reads, reads left for mapping after quality trimming and numbers of removed plasmid matching reads (in brackets are percentages of mapped reads to EYFP-plasmid vs. HA-His₆-MBP-PPR65 plasmid for co-transfected cells), editing efficiencies of the respective delivered targets (%editing) and the fraction of off-targets, which are confirmed in the individual replicates. The corresponding SRA data is deposited within BioProject PRJNA832818.

Name	Cell line	RNASeq batch	Expressed protein	Delivered target	Replicate	FACS sorted	Raw reads	After quality trimming > % of raw reads	Matching plasmid > % of clean reads	Reads for mapping	%editing > delivered target	Off-targets confirmed
IMR1	IMR-90	2	EYFP-PPR56	nad4	1	Yes	99106154	87380226 (88.2%)	32.4%	59007884	71%	727/906 (80.2%)
IMR2	IMR-90	1	EYFP-PPR56	nad4	2	Yes	117428040	101532360 (86.5%)	27.2%	73903770	71%	839/906 (92.6%)
IMR3	IMR-90	1	EYFP-PPR56	nad4	3	Yes	110474636	95441720 (86.4%)	28.4%	68253350	70%	814/906 (89.8%)
ELIM1	IMR-90	2	EYFP-PPR56	nad4	4	No	115482810	103838880 (89.9%)	26.1%	76716486	72%	533/906 (58.8%)
IMR5	IMR-90	1	EYFP-PPR56-CxDAH	nad4	1	Yes	124316786	107041332 (86.1%)	27.0%	78065000	0%	/
I-1-2	IMR-90	3	EYFP-PPR56_S4TN>TD	nad4	1	Yes	128657316	115191558 (89.5%)	38.3%	70994072	27%	2063/2265 (91.1%)
I-3	IMR-90	3	EYFP-PPR56_S4TN>TD	nad4-7AG	2	Yes	112121016	100559526 (89.7%)	37.4%	62881814	65%	1987/2265 (87.7%)
I-4	IMR-90	3	EYFP-PPR56_S4TN>TD	nad4-7AG	3	Yes	122435238	108661546 (88.8%)	38.5%	66747124	64%	2028/2265 (89.5%)
I-6-2	IMR-90	3	EYFP-PPR56_S7TD>TN	nad4	1	Yes	126830966	113777666 (89.7%)	39.2%	69104234	5%	284/323 (87.9%)
I-7	IMR-90	3	EYFP-PPR56_S7TD>TN	nad4-10GA	2	Yes	104658340	93086300 (88.9%)	44.6%	51480558	53%	261/323 (80.8%)
I-8	IMR-90	3	EYFP-PPR56_S7TD>TN	nad4-10GA	3	Yes	133100572	118817108 (89.3%)	44.4%	65931130	50%	290/323 (89.8%)
I-9	IMR-90	3	EYFP + HA-His ₆ -MBP-PPR65 (co-transfection)	ccmFC	1	Yes	249859798	221774196 (88.8%)	30.1% (5.6%/ 24.5%)	154861296	19%	4/7 (57.1%)
I-10	IMR-90	3	EYFP + HA-His ₆ -MBP-PPR65 (co-transfection)	ccmFC	2	Yes	274709688	242595378 (88.3%)	30.5% (4.9%/ 25.6%)	168492352	20%	7/7 (100%)
I-12	IMR-90	3	EYFP + HA-His ₆ -MBP-PPR65 (co-transfection)	ccmFC	3	Yes	255509484	225227710 (88.2%)	30.6% (6.5%/ 24.1%)	156244988	20%	5/7 (71.4%)
IMR8	IMR-90	1	EYFP	none	Control PPR56	Yes	110753644	98081586 (88.6%)	19.4%	78948974	no PPR/ target	/
I-13	IMR-90	3	EYFP + HA-His ₆ -MBP (co-transfection)	none	Control PPR65	Yes	139760184	124347896 (89.0%)	25.0% (5.5%/ 19.5%)	93136262	no PPR/ target	/

ARNT-NM_001197325.2eU1108-NP_001184254.1aa3597T	3	9%	4%	9%	6%	0	0	290	160	780	230	400	T	C	T	C	C	G	A	C	A	C	A	C	A	C	A	T	T	G	A	G	G	T	A	T	C	T	T	C	A	G	T	T	T	T	G	13%	5%	10%	
ASAH2B-NM_001079516.4eU550-NP_001072984.1aa158PL	4	49%	28%	61%	17%	0	1	7%	260	300	690	430	160	C	T	G	T	C	A	T	A	C	T	T	T	C	A	T	T	T	G	A	A	G	G	C	A	G	T	C	C	C	G	G	C	T	72%	70%	41%	13%	7%
ASB16-NM_0008663.5eU1914-3UTR+466	2	22%	11%	30%	0	0	3	25%	220	110	490	200	200	G	G	C	T	C	T	T	G	A	G	A	C	C	T	G	C	T	G	A	C	T	T	C	C	C	G	G	A	C	T	23%	29%	23%					
ASCC3-NM_0006828.4eU6781-NP_006619.2aa2157YY	4	5%	1%	4%	1%	0	0	520	230	680	330	180	A	C	C	T	T	C	T	T	G	A	A	T	A	C	C	T	G	G	A	A	G	G	T	A	T	T	C	T	C	A	G	A	C	5%	3%	5%	6%		
ASH1L-NM_001396177.2eU4968-NP_001353106.1aa1406PP	3	16%	4%	19%	4%	0	0	210	360	800	260	380	A	G	A	T	T	A	G	G	T	T	A	C	T	A	T	G	A	A	G	A	G	A	G	A	G	A	G	A	G	A	G	A	G	22%	16%	14%			
ASXL2-NM_001369346.1eU3253-NP_001356275.1aa1044TI	2	8%	4%	8%	4%	0	0	260	130	610	290	300	A	C	T	C	A	G	T	T	T	C	C	A	G	A	G	C	T	G	C	C	A	A	A	A	A	A	A	A	A	A	A	A	5%	10%					
ATG101-NM_001098673.2eU437-NP_001092143.1aa146SF	2	2%	1%	2%	1%	0	0	350	240	740	400	180	A	G	T	C	C	A	C	T	T	C	A	A	G	A	G	A	G	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	1%	2%					
ATC14-NM_014924.5eU096-NP_055793.2aa326PP	4	2%	14%	34%	11%	0	0	300	230	540	490	370	T	G	T	T	C	T	C	A	T	A	T	A	C	T	T	G	A	T	G	A	A	A	A	A	A	A	A	A	A	A	A	45%	24%	32%	12%	7%	6%		
ATC-NM_004044.7eU625-NP_004035.2aa181HY	3	2%	1%	2%	1%	0	0	220	280	770	150	300	A	G	C	G	C	C	A	T	T	A	G	C	T	T	G	A	G	A	G	A	G	A	G	A	G	A	G	A	G	A	G	A	2%	2%	3%				
ATP13A3-NM_001367549.1eU1382-NP_001354478.1aa286SF	3	6%	4%	10%	3%	0	0	380	260	730	260	310	G	A	G	T	A	A	T	G	A	A	G	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	2%	2%					
ATP5F1A-NM_001001935.3eU1572-NP_001001935.1aa428PP	4	2%	1%	2%	1%	3%	28%	3%	190	380	620	330	440	T	G	A	T	T	G	C	T	G	A	G	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	2%	2%						
ATP5P0-NM_001697.3eU505-NP_001688.1aa155SF	4	8%	4%	10%	1%	0	0	380	200	720	210	290	C	T	G	C	T	C	T	T	T	T	G	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	11%	10%	2%	24%	30%	29%		
ATP6V1D-NM_015994.4eU309-NP_057078.1aa68SL	4	8%	3%	9%	1%	0	3	70%	480	210	700	270	300	T	G	C	G	C	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	10%	9%	8%	3%	70%	71%	69%
ATPSCKMT-NM_199133.4eU470-NP_954584.2aa152SL	3	10%	0%	10%	0%	0	0	570	230	820	550	320	A	T	T	T	C	A	G	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	10%	10%	10%					
ATRX-NM_000489.6eU10170-3UTR+2476	3	10%	4%	10%	4%	0	0	390	220	820	410	180	T	A	A	G	T	A	C	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	8%	15%	10%					
ATXN3-NM_001127696.2eU1694-3UTR+623	3	27%	11%	27%	11%	0	1	9%	430	170	550	510	230	C	A	C	A	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	22%	40%	20%	9%				
AVL9-NM_015060.3eU686-NP_055875.1aa149Q*	4	23%	0%	27%	3%	0	0	220	420	780	450	470	C	A	T	C	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	3%	19%	2%	20%	16%			
B3GALT9-NM_001386823.1eU841-NP_001373752.1aa68SS	3	28%	0%	32%	6%	0	0	340	260	740	470	150	G	A	C	T	C	G	A	C	C	C	C	T	A	A	G	A	G	A	G	A	G	A	G	A	G	A	G	A	G	A	G	A	26%	38%	19%				
BAD-NM_004322.3eU727-NP_004313.1aa131RC	4	5%	2%	6%	1%	0	0	510	270	730	370	240	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	7%	7%	5%	2%				
BANF1-NM_001143985.1eU464-NP_001137457.1aa5Q*	4	12%	6%	14%	2%	0	1	2%	200	320	740	220	370	A	A	G	A	T	T	A	A	G	C	T	G	A	T	C	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	15%	16%	12%	4%	2%	
BANF1-NM_003860.4eU97-NP_003851.1aa5Q*	4	12%	6%	14%	1%	0	1	1%	200	320	740	220	370	G	A	T	T	A	A	G	C	T	G	A	T	C	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	16%	13%	14%	5%				
BANF1-XM_017018514.1eU514-XP_016874003.1aa5Q*	3	15%	5%	15%	5%	0	0	200	320	740	220	370	A	A	T	T	A	A	G	C	T	G	A	T	C	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	19%	16%	10%	1%				
BAX-NM_001291429.2eU1107-3UTR+448	2	2%	0%	2%	0%	0	0	200	350	710	200	330	A	G	T	C	C	T	G	T	C	T	C	A	G	A	G	A	G	A	G	A	G	A	G	A	G	A	G	A	G	A	G	A	2%	2%					
BBX-NM_001142568.3eU2664-NP_001136040.1aa788SL	4	15%	8%	16%	9%	0	0	430	210	830	220	230	A	T	G	C	A	T	T	C	A	C	C	T	A	C	A	G	A	G	A	G	A	G	A	G	A	G	A	G	A	G	A	G	27%	12%	9%	11%			
BBX-NM_0020235.7eU2574-NP_064620.2aa758SL	4	14%	7%	15%	8%	0	0	430	210	830	220	230	A	T	G	C	A	T	T	C	A	C	C	T	A	C	A	G	A	G	A	G	A	G	A	G	A	G	A	G	A	G	A	G	24%	9%	13%	11%			
BCAR1-NM_001170714.3eU868-NP_001164185.1aa195SL	3	3%	1%	3%	1%	3%	28%	8%	280	240	550	350	180	C	T	C	C	C	C	A	G	C	C	A	A	G	A	G	A	C	A	C	C	C	T	T	T	T	T	T	T	T	T	2%	4%	3%	25%	37%	22%		
BCAS2-NM_005872.3eU792-3UTR+90	4	5%	2%	6%	1%	3%	17%	4%	300	360	700	320	250	A	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	5%	7%	5%	2%					
BCL2L12-NM_001040668.1eU1096-NP_001035758.1aa138PP	4	4%	1%	4%	2%	0	3	60%	290	220	700	240	350	T	T	C	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	6%	3%	4%	3%					
BCL2L12-NM_001282515.1eU1099-NP_001269445.1aa139PP	3	5%	1%	5%	1%	0	3	64%	290	220	700	240	350	T	T	C	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	6%	3%	4%	3%					
BCOR-NM_001123385.2eU792-NP_00116857.1aa33SL	3	26%	15%	26%	15%	0	0	210	120	790	120	440	A	G	T	C	C	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	43%	15%	19%						
BIRC2-NM_001166.5eU1932-NP_001157.1aa171SL	4	5%	1%	6%	1%	0	0	390	140	780	250	150	T	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	5%	6%	5%	4%					
BLMH-NM_000386.4eU1294-NP_000377.1aa390TI	4	10%	4%	12%	2%	0	3	63%	300	270	640	380	310	C	A	G	A	A	G	A	G	A	T	G	A	T	C	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	14%	9%	12%	5%				
BMP1-NM_006129.5eU3253-3UTR+258	4	3%	1%	3%	1%	0	0	320	280	680	220	110	C	T	C	C	A	A	G	A	G	A	C	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	4%	5%	2%	3%					
BNIP3-NM_004052.4eU1380-3UTR+678	4	38%	10%	43%	5%	0	3	7%	270	170	830	190	100	A	T	G	A	T	C	A	G	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	48%	38%	42%	26%	4%					

EGFR-NM_001346897.2eU408-NP_00133326.1aa49LL	4	20%	11%	24%	9%	0	0	390	210	650	420	350	G	C	A	G	T	T	G	G	G	C	A	C	T	T	T	G	A	A	G	A	T	T	C	T	C	T	A	G	C	C	T	33%	24%	15%	9%
EID2-NM_153232.4eU747-NP_694964.3aa222PL	2	4%	2%	4%	2%	0	0	250	150	650	180	230	T	A	G	C	T	C	T	G	A	C	T	G	C	T	C	T	G	A	A	G	T	T	A	C	A	A	C	C	T	2%	5%				
EIF1AD-NM_001242461.2eU571-NP_001229410.1aa69LL	2	5%	1%	6%	0%	0	0	440	260	800	350	440	G	A	A	C	A	C	T	C	T	G	G	A	T	C	A	A	G	A	G	G	G	G	G	G	G	G	G	G	G	6%	6%	4%			
EIF2AK2-NM_001135651.3eU1767-NP_00129123.1aa504SL	4	21%	9%	26%	1%	0	3	220	160	840	160	290	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	26%	26%	25%	6%				
EIF2AK2-NM_001135651.3eU892-NP_001129123.1aa209SL	4	6%	3%	7%	1%	0	3	270	320	770	140	200	C	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	7%	6%	6%	2%				
EIF2A-NM_001319043.2eU1206-NP_001305972.1aa397PL	4	4%	0%	4%	1%	0	0	320	290	710	440	110	C	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	5%	5%	3%	2%				
EIF2S3-NM_001415.4eU110-NP_001406.1aa339HY	4	10%	4%	12%	1%	0	2	310	130	720	300	250	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	12%	11%	12%	4%				
EIF3J-NM_001284335.2eU1268-3UTR+605	3	2%	0%	2%	0%	0	0	360	320	450	500	490	A	A	A	A	C	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	2%	2%	2%	1%				
EIF4G1-NM_001194946.2eU3101-NP_001181875.2aa981RC	3	2%	0%	2%	0%	0	3	340	220	710	220	410	A	A	A	T	C	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	3%	48%	2%					
EIF5A-NM_001970.5eU367-NP_001961.1aa74PL	3	2%	0%	2%	0%	0	0	210	150	720	280	310	T	A	C	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	2%	1%	2%					
ELLAVL1-NM_001419.3eU543-NP_0014140.2aa127SF	4	17%	6%	20%	2%	0	2	410	150	850	140	370	G	A	C	C	A	G	A	G	A	G	A	C	T	A	G	A	G	A	G	A	G	A	G	A	G	A	G	A	G	2%	2%				
ELL2-NM_012081.6eU1079-NP_036213.2aa317FS	4	2%	1%	2%	1%	0	0	190	340	610	60	560	G	T	A	G	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	22%	16%	19%	8%			
EMC1-NM_001271427.2eU3137-3UTR+143	2	2%	0%	2%	0%	0	0	570	230	470	200	520	A	A	A	A	G	A	G	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	2%	2%						
EMP1-NM_001423.3eU1297-3UTR+634	4	4%	2%	4%	2%	0	1	700	210	620	520	240	G	G	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	1	3%						
ENGASE-NM_001042573.3eU196-3UTR+1843	2	32%	8%	38%	0%	0	0	330	270	340	500	230	A	C	T	G	A	C	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	4%	3%	6%	2%				
EPB41L3-NM_001281533.2eU361-NP_001268462.1aa89Q*	4	7%	2%	8%	2%	0	0	320	290	650	260	570	T	A	T	C	G	C	A	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	3%						
EPS8-NM_004447.6eU348-3UTR+730	4	4%	1%	4%	1%	0	0	620	60	680	380	230	A	A	G	C	A	A	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	8%	10%	6%	5%				
EPS8-NM_004447.6eU3450-3UTR+732	4	7%	3%	8%	3%	0	3	320	200	620	60	680	A	G	C	A	A	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	39%	3%						
ERBIN-NM_001006600.3eU695-NP_001006600.1aa136Q*	2	3%	1%	3%	1%	0	0	280	420	610	130	360	C	T	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	2%							
ERCC3-NM_000122.2eU335-NP_0001113.1aa91PL	2	2%	0%	2%	0%	0	0	290	210	660	530	70	A	T	G	C	C	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	2%							
ERMAP-NM_001017922.2eU2020-3UTR+339	2	1%	0%	1%	0%	0	0	350	210	790	260	360	A	C	T	G	A	G	G	G	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	2%							
ESF1-NM_001276380.2eU2167-NP_001263309.1aa695PL	4	16%	3%	17%	2%	0	1	460	220	670	440	120	C	G	T	A	A	A	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	1	2%						
EXO5-NM_001346946.2eU1568-3UTR+17	4	30%	9%	34%	4%	0	1	240	400	760	120	360	A	A	A	A	G	C	C	A	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	8%							
EZH2-NM_001203247.2eU821-NP_001190176.1aa229SL	4	30%	10%	35%	3%	0	3	280	190	800	320	230	C	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	3%	3%	1%					
EZH2-NM_004456.5eU821-NP_004447.2aa229SL	4	88%	9%	91%	9%	0	1	280	190	800	320	230	C	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	1	50%						
EZR-NM_00111077.2eU327-NP_001104547.1aa68Q*	4	1%	1%	2%	0%	0	3	210	240	770	280	310	T	G	G	T	G	A	G	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	3	31%	3%					
FAF2-NM_014613.3eU1247-NP_055428.1aa408PP	4	5%	3%	6%	2%	0	0	250	240	570	640	390	A	A	A	A	G	A	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	2%						
FAF2-NM_014613.3eU2540-3UTR+1179	4	22%	7%	26%	3%	0	0	420	300	770	310	40	G	C	T	G	T	G	T	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	2%						
FAM111B-NM_001142703.2eU1577-NP_001136175.1aa488TI	3	2%	0%	2%	0%	0	0	380	110	600	520	270	T	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	2%							
FAM118B-NM_001330446.1eU255-NP_001313715.1aa24PP	3	8%	3%	8%	3%	0	0	340	350	500	430	330	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	11%							
FAM217B-NM_001190827.2eU203-5UTR-163	4	31%	9%	35%	9%	1	19%	260	150	790	400	220	A	T	A	A	G	A	G	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	4%							
FAM27B-NM_001100910.2eU1037-NP_001094380.1aa62TT	2	5%	2%	5%	2%	0	0	270	180	650	460	270	A	G	T	G	A	C	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	6%							
FAM91A1-NM_001317917.2eU2730-NP_001304846.2aa578SL	3	6%	1%	6%	1%	0	3	320	160	760	180	290	A	A	A	A	C	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	5%							
FAM98B-NM_173611.4eU583-NP_075882.2aa183SL	2	2%	0%	2%	0%	0	0	260	350	670	310	520	A	G	T	G	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	2%							
FANCA-NM_000135.4eU474-NP_0001026.2aa148LL	3	15%	1%	15%	1%	0	3	250	220	740	320	390	A	C	T	G	G	A	G	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	39%							
FASTKD2-NM_001136193.2eU1118-NP_001129665.1aa309TI	4	8%	5%	9%	4%	0	3	250	150	790	140	520	A	G	C	A	G	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	74%							
FASTK-NM_006712.5eU626-NP_006703.1aa195PS	2	2%	0%	2%	0%	0	0	260	250	700	400	100	A	G	C	T	C	C	G	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	2%							
FAT1-NM_005245.4eU9222-NP_005236.2aa3004SL	4	13%	4%</																																												

NPPB-NM_002521.3eU137-NP_002512.1aa9RW	4	4%	1%	5%	0%	3	37%	1%	0	290	180	800	250	310	A	G	A	G	A	C	A	T	G	G	A	T	C	C	C	C	A	G	A	C	A	G	C	A	C	T	T	C	C	G	G	G	C	5%	5%	5%	2%	36%	37%	38%
NRC2-NM_001921694.2eU2093-NP_001278623.1aa577PL	0					3	4%	1%	0	320	280	740	140	420	T	G	G	C	A	A	T	G	T	T	T	G	A	T	A	G	A	C	A	G	C	A	T	A	T	T	C	C	A	C	A	C	7%	10%			3%	5%	5%	
NRC1-NM_001159895.3eU1145-NP_001153467.1aa332SF	0					2	9%	2%	0	370	400	760	170	310	G	A	G	A	C	A	C	G	G	A	C	A	C	A	G	T	G	A	A	G	C	A	T	A	T	T	T	T	T	T	T	T	T	2%	2%					
NJAK2-NM_030952.3eU2390-3UTR+386	0					3	6%	1%	0	330	230	680	430	270	A	C	T	G	T	G	T	C	T	T	A	G	A	G	A	C	A	G	C	T	C	T	T	T	T	T	T	T	T	T	T	T	7%	5%	6%					
NUP43-NM_198887.3eU2393-3UTR+1216	0					3	10%	4%	0	370	290	720	320	300	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	12%	6%	13%						
OB1-NM_024546.4eU1989-NP_078822.3aa652Q*	0					3	18%	3%	3	380	240	740	350	440	C	C	A	G	C	T	G	T	G	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	14%	20%	20%	15%	23%	20%			
OSBPL2-NM_001278649.3eU1600-3UTR+61	0					3	12%	4%	0	380	280	780	140	460	A	C	A	G	A	C	T	G	A	G	T	C	G	C	A	G	A	T	A	G	C	A	T	C	A	T	C	T	C	A	T	11%	17%	9%						
OSBPL9-NM_001350208.2eU225-5UTR-538	3	4%	2%	5%	1%	3	44%	4%	0	290	160	810	190	280	G	T	A	T	A	G	C	G	A	T	A	G	A	G	A	C	A	G	C	T	C	A	A	T	A	A	A	A	A	A	A	5%	4%		2%	40%	46%	47%		
OTUD7B-NM_020205.4eU2588-NP_064590.2aa748SF	0					2	6%	1%	0	510	150	680	270	230	A	C	C	C	T	A	G	C	C	C	C	A	G	A	G	A	C	A	G	C	A	T	C	T	T	T	T	T	T	T	T	5%	7%							
PABPC4-NM_001324436.2eU1785-NP_001129125.1aa316PS	0					3	4%	1%	0	400	350	810	460	400	G	A	T	G	A	G	A	T	A	A	T	T	A	A	G	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	4%	6%	3%					
PAPOLG-NM_022894.4eU3708-3UTR+1286	0					2	44%	8%	0	430	220	840	220	280	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	50%	39%							
PARL-NM_001324436.2eU596-NP_001311365.1aa188LL	0					3	7%	2%	0	470	110	760	230	370	C	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	9%	5%	8%							
PCLAF-NM_014736.6eU415-3UTR+9	0					2	2%	1%	0	440	210	680	500	370	C	A	C	A	A	T	G	A	T	A	A	A	A	G	A	T	A	G	A	C	T	T	T	T	T	T	T	T	T	T	T	2%	2%							
PCSK9-NM_174936.4eU279-5UTR-13	0					3	13%	8%	0	350	340	730	160	300	C	T	A	G	T	C	T	C	T	C	G	C	A	G	A	C	A	G	C	A	C	C	C	C	C	C	C	C	C	C	C	9%	22%	9%						
PCYOX1-NM_016297.4eU5208-3UTR+3662	0					3	16%	7%	0	230	210	750	120	420	T	A	G	T	A	A	T	A	C	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	8%	22%	17%							
PEG10-NM_001040152.2eU3267-3UTR+1855	0					2	2%	0%	3	380	300	690	290	540	C	A	T	C	T	C	A	G	G	C	T	G	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	2%	2%								
PHACTR2-NM_001100164.2eU6956-3UTR+4819	0					3	41%	6%	0	240	230	600	180	440	A	C	C	A	A	T	G	C	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	39%	36%	47%	8%	10%	9%				
PHF6-NM_001015877.2eU2407-3UTR+1107	0					3	5%	1%	0	350	290	860	260	490	A	A	G	A	A	C	C	A	G	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	4%	5%	5%							
PHF6-NM_001015877.2eU4050-3UTR+2750	0					2	4%	2%	0	400	100	740	220	390	G	A	G	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	4%	5%	2%							
PHYKPL-XM_024446242.1eU1589-5UTR-2097	0					2	21%	6%	0	340	250	570	420	410	A	G	T	G	G	T	T	A	A	A	C	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	17%	25%								
PK3CB-NM_001256045.2eU2677-3UTR+582	0					2	13%	5%	0	260	240	730	220	200	T	A	A	T	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	17%	10%									
PLSCR3-NM_001369421.1eU185-5UTR-73	0					2	7%	2%	0	300	160	790	80	380	C	A	G	A	A	C	T	A	A	G	C	A	C	A	G	A	C	A	G	C	A	T	C	T	C	A	T	C	T	C	A	6%	6%	9%						
PLXNA1-NM_032242.4eU6746-3UTR+809	0					2	6%	0%	0	370	180	750	250	350	A	G	A	G	G	C	C	C	G	G	T	C	C	A	G	A	G	A	G	A	G	A	G	A	G	A	G	A	G	A	6%	6%	6%							
PMS2-NM_000535.7eU2687-3UTR+68	0					3	32%	7%	3	470	160	740	220	470	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	28%	31%	40%	36%	29%	38%					
POU2F2-NM_001207025.4eU5695-3UTR+4220	4	37%	19%	40%	22%	3	38%	8%	1	390	80	460	280	180	A	C	C	T	G	A	C	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	40%	19%	63%	27%								
PPP2R3B-NM_0013239.5eU1286-NP_037371.2aa357SL	0					3	2%	1%	0	310	150	910	100	420	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	25%	22%	24%								
PPP2R5C-NM_001161725.2eU1123-NP_001155197.1aa313TM	0					2	2%	0%	0	280	160	690	210	390	T	G	C	A	G	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	3%	2%									
PPP6R3-NM_001352354.2eU135-5UTR-227	0					2	7%	1%	0	490	100	650	370	370	C	A	T	T	C	T	C	A	A	T	A	T	A	G	T	A	G	A	C	T	T	T	T	T	T	T	T	T	T	8%	7%									
PRPL-NM_001042385.2eU829-NP_001035844.1aa265LL	0					2	14%	4%	3	380	180	470	530	310	C	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	11%	17%										
PRKCA-NM_002737.3eU6655-3UTR+4397	0					3	9%	2%	0	290	250	610	420	310	A	A	G	G	T	G	A	T	A	C	C	T	C	T	G	A	C	A	G	T	C	A	T	T	T	T	T	T	T	10%	6%	10%								
PRKCI-NM_002740.6eU493-NP_002731.4aa83Q*	0					2	5%	2%	0	250	110	720	120	360	G	A	G	A	A	C	G	A	A	C	C	C	G	T	T	A	C	A	G	T	A	C	A	T	C	A	T	C	A	4%	6%									
PRMT6-NM_018137.3eU2095-3UTR+925	0					2	9%	1%	0	290	410	780	480	540	A	A	C	T	C	A	C	T	A	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	10%	8%									
PRR11-NM_018304.4eU376-NP_060774.2aa104PL	0					2	4%	2%	0	320	240	800	320	320	G	T	T	A	G	A	A	G	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	9%	10%	11%								

Table with columns: Accession ID, Count, Percent, and Sequence alignment. Contains genomic coordinates and nucleotide alignments (A, C, G, T, N) for various sequences. The table ends with a summary of overall counts for each nucleotide across the entire dataset.

ZNF451-NM_001031623.3eU2201-NP_001026794.1aa712PL	0	0	3	9%	3%	320 340	650	350 200	A A A C T T C A A T T T A A A A C	G A A G A T G A T T T T C	A G T A A	11%	8%	6%
ZNF451-NM_001257273.2eU1721-NP_001244202.1aa552PL	0	0	2	14%	7%	460 260	590	370 200	C C T A T A G A T A T A G A A G	A A G G A G A C T C T C	A G A A C	10%	18%	
ZNF461-NM_001297623.3eU134-5UTR-96	0	0	2	9%	1%	380 310	470	370 340	T C C G A A T C A G A G C T T G G	A C C A G A G C T T C C	T G A G A	10%	8%	
ZNF461-NM_001297623.3eU1756-NP_001284552.1aa510HY	0	0	3	31%	2%	320 230	770	290 290	A A A C C T T A T C A A T G C G G	G A G G C G T T T A A T	C A T A G A	33%	29%	30%
ZNF462-NM_001347997.2eU4898-NP_001334926.1aa1560Q*	0	0	2	8%	4%	250 260	680	420 560	C C A T G T G A A T T T T G T G G	A C G G C G T T T T C A	C A G G G C	10%	5%	
ZNF473-NM_001006656.4eU2238-NP_001006657.1aa684TI	0	3	2	6%	3%	440 300	690	100 420	T T A A A T G T A G T A A G T G T	G A C A G A G T C T T C A	C C C A G A	6%	5%	5%
ZNF518A-NM_001278524.2eU822-5UTR-83	0	0	2	41%	20%	520 140	700	250 220	G A A A A T C C T G T A T A T T G A	A G A T G T C T C T A	C A C A G T	4%	27%	8%
ZNF518B-NM_001375816.1eU4914-3UTR-973	0	0	2	14%	0%	410 70	780	260 270	T C C A A A G A C C A T G G G T G	G A T G C T T T C T	T C T T C	8%	10%	14%
ZNF566-NM_001145343.1eU1270-NP_001138815.1aa379Q*	3	11%	1%	11%	1%	280 240	820	280 370	G A A T G T A A G A T A T G T G G	G A A G G C T T A T T C T	C A G A G T	12%	11%	10%
ZNF592-NM_014630.3eU240-5UTR-93	0	0	3	12%	9%	330 260	580	490 270	A C G G G A G A A G A C A A A G	G A A G A C G T C C C C	G T A C G	5%	10%	22%
ZNF598-NM_178167.4eU1411-NP_835461.2aa446PL	0	0	2	5%	2%	250 290	680	360 180	C A G C C C G A A G C T C A A G	G A C G A A G A C T T C C	C A G C C	3%	6%	6%
ZNF623-NM_001082480.2eU881-NP_001075949.1aa178HY	0	0	3	18%	2%	420 300	830	290 330	A A A T G T G C G C A G T G T G G	G A A G G C G T T T T G T	C A C A G T	15%	19%	19%
ZNF638-NM_001014972.3eU3685-NP_001014972.1aa1133PP	0	0	3	4%	2%	350 270	500	430 400	A C C T A A T G A G C T T G A A G	A A G A A A G T A C T C C	C A G C A T	3%	4%	6%
ZNF639-NM_001375800.1eU440-5UTR-186	0	0	3	27%	6%	350 260	760	460 470	T T T G T A T T T T C A G T C G G	A C G G G G T T T C C C	C A T G T T	20%	3%	29%
ZNF664-NM_001204298.2eU1521-NP_001191227.1aa182Q*	0	0	3	6%	2%	260 280	700	230 360	A A A T G T T A T G A G T G T G G	G A A G G C G T T C A G T	C A G A G T	4%	7%	8%
ZNF670-NM_001204220.2eU1677-3UTR+295	0	0	3	19%	10%	320 230	730	220 380	A G T G G T T C A T T G T T A A A	G A A G G T G T C A T A	C T T T A G	12%	16%	30%
ZNF689-NM_138447.3eU710-NP_612456.1aa121PL	0	0	3	14%	2%	430 180	610	530 290	G A A G A A G A A T G G G G A G	A A G A A G T A T T C C	C G C C T A	12%	14%	15%
ZNF697-NM_001080470.2eU4946-3UTR+2695	0	0	3	12%	3%	470 170	350	410 350	T A A A T G C A T T C A G G A G G	T T G T T G T T T C T T	C T A T C T	11%	10%	16%
ZNF71-NM_001370214.1eU2657-3UTR+1057	0	0	2	27%	5%	250 200	530	310 480	G T A G T G C T T G T G T C T T G	A T G T G T G T C T T C A	G T G G T	3%	7%	7%
ZNF736-NM_001170905.3eU5870-3UTR+4396	0	0	2	35%	14%	470 60	650	290 130	A A C T C T T A C A C T T T G A T	G C T G T G T C T T	C A T T T C	25%		44%
ZNF740-NM_001004304.4eU1000-NP_001004304.1aa193LL	0	0	2	15%	11%	350 260	610	300 560	C A G T C C A A G A C T T T C G G	A C G G C A G T T T T C T	T A T A G	7%	22%	
ZNF765-NM_001040185.3eU4054-3UTR+2364	0	0	2	11%	5%	420 320	690	500 410	T T T G T A T T T T T A G T A G A	G A T G G G G T T T C T C	C A T G T T	8%		15%
ZNF780B-NM_001005851.3eU5120-3UTR+2530	0	0	2	30%	16%	300 70	750	80 470	T A A G T A A T C T A T G G A G A	G A C G T T G T C A T C T	T G A A A	19%		41%
ZNF865-NM_001195605.2eU540-NP_001182534.1aa143PL	0	0	3	20%	6%	250 350	630	400 260	C C C C T C C T C C C C T C T T T	G A C G C T G C T T T C C	C A C T C	26%	18%	15%
ZNRF1-NM_032268.5eU1685-3UTR+353	0	0	2	19%	4%	310 330	570	430 450	A A A A A G T C A G T G T C G	A C T G T G T T T T C C	C T C G T G	16%		21%
ZRANB2-NM_005455.5eU582-NP_005446.2aa181SL	0	0	3	6%	2%	260 230	650	230 330	A G G A T G A G G A T G A A G A T	G A C G C T G A T C T C T	C A A A A T	3%	7%	7%
ZSCAN12-NM_001163391.2eU1681-NP_001156863.1aa508Q*	0	0	3	22%	3%	320 280	840	290 390	A A A T G T G A T A A G T G T G G	G A A G G C G T T T A C T	C A A A G A	22%	20%	25%
ZSCAN12-NR_028077.2eU3726-ncr	0	0	3	20%	10%	350 260	690	460 410	T T T G T A T T T T T A G T A G A	G A C G G G T T T C A C	C A T G T T	31%	13%	16%
ZSCAN23-XM_011514406.2eU1848-3UTR+499	0	0	2	18%	5%	260 130	810	290 400	C A T G A G G T T T A A G G A A A	G A A G C T G T C C A	C A A C A T	16%		21%
ZSWIM7-NM_001042697.2eU308-NP_001036162.1aa66YY	0	0	3	39%	12%	360 450	740	260 240	A A T C T C A T C A C C C A G G	G A A G G C G T G T T A	C A G G T	26%	48%	44%
ZSWIM8-NM_001242487.2eU4898-NP_001229416.1aa1542PL	0	0	2	8%	2%	210 140	620	320 290	T G C C T C A C A T G C C C C G G	C C T G C C G T C T T C C	C T G T G C	10%		7%
ZYX-NM_001010972.2eU358-NP_001010972.1aa94PL	0	0	3	2%	1%	310 390	680	330 370	A T G C A G A G G G T G C T G G	A G T G C T T C C	G C C G C	2%	2%	3%

Supplementary Table S5

PPR65 off-targets detected in the IMR-90 transcriptome. Off-targets appearing in at least two replicates of HA-His₆-MBP-PPR65 in the transcriptome of IMR-90. The off-target label reveals the gene ID, the transcript accession with the position of the edited C, the corresponding protein accession with the position of the affected amino acid and the induced amino acid change. Off-targets in non-coding regions are labeled with 'npc'. Shown are the number of replicates, in which the site was called (Rep.) and the mean C-U conversion rates (%CU) with standard deviations (STABW). Nucleotides from position -30 up- to +5 downstream of the editing sites are listed and nucleotide shading is based on the PPR-RNA binding code with perfect matches in green and moderate matches (transition) in yellow (see Figure 1). Conversion rates of individual replicates (labeled as in Supplementary table S3) are depicted.

>total: 7

Offtarget	Rep.	%CU	STABW	Binding region	% editing in individual replicates		
					I-9	I-10	I-12
TCP11L1-NM_001145541.1eU481-NP_001139013.1aa89-Q*	2	8%	1%	G C C C A T G A A A T T G T A G T A A A T G G A G A C T T T C A G A T T	7%	9%	
NDUFAF3-NM_199069.2eU523-npc	3	6%	2%	T G T C A T G A A G G C C G A G T A A C T G G A G C T G C T C T C A T C	8%	6%	4%
EML4-NM_001145076.3eU1496-NP_001138548.2aa413-LF	2	3%	1%	T G T T T A G C A T T C T T G G G A A T G G A G A T G T T C T T A C T		3%	2%
MRPL14-NM_001318767.2eU572-npc	2	2%	0%	C T G G T T G C A G G A C T C G T G A A T G G A G C A G T T C T G A G A		2%	2%
SPDYE16-XM_017012887.2eU206-npc	3	13%	5%	G G A G A A A A T C A G A A A G A T G G C T T A G A G A A G C C A G C A	9%	18%	12%
HMG1-NM_011529543.2eU1210-npc	2	7%	1%	C A G C C T C T G A T G A A G C A G G A G A G A A A A G A G A A G C C A A G T	6%	7%	
EPB41L1-XM_024451852.1eU174-npc	2	6%	3%	C C C G C G C C G C C C G C C C G C C C G G C C T G C C T G C C A G A G		8%	4%

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