# **Supplementary Data**

# A cytosine base editor toolkit with varying activity windows and target scopes for versatile gene manipulation in plants

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hAID	1	MDSLLMNRRKFLYQFKNVRWAKGRRETYLCYVVKRRDSATSFSLDFGYLR	50
hAID*∆	1	MDSLLMNRR <mark>E</mark> FLYQFKNVRWAKGRRETYLCYVVKRRDSATSFSLDFGYLR	50
AID10	1	MDSLLMNRR <mark>E</mark> FLYQFKNVRWAKGRRETYLCYVV <mark>E</mark> RRD <mark>C</mark> ATSFSLDFGYLR	50
hAID	51	NKNGCHVELLFLRYISDWDLDPGRCYRVTWFTSWSPCYDCARHVADFLRG	100
hAID* $\Delta$	51	NKNGCHVELLFLRYISDWDLDPGRCYRVTWF <mark>I</mark> SWSPCYDCARHVADFLRG	100
AID10	51	NKNGCHVELLFLRYISDWDLDPGRCYRVTWF <mark>I</mark> SWSPCYDCARHVADFLRG	100
hAID	101	NPNLSLRIFTARLYFCEDRKAEPEGLRRLHRAGVQIAIMTFKDYFYCWNT	150
hAID* $\Delta$	101	NPNLSLRIFTARLYFCEDRKAEPEGLRRLHRAGVQIAIMTFKDYFYCWNT	150
AID10	101	NPNLSLRIF <mark>A</mark> ARLYFCEDRKAEPEGLRRL <mark>R</mark> RAGVQIAIMTFKDYFYCWNT	150
hAID	151	FVENHERTFKAWEGLHENSVRLSRQLRRILLPLYEVDDLRDAFRTLGL	198
hAID* $\Delta$	151	FVENH <mark>G</mark> RTFKAWEGLHENSVRLSRQLRRILLPLYEVDDLRDAFRT	195
AID10	151	F <mark>A</mark> ENH <mark>G</mark> RTFKAWEGLHENSVRLS <mark>G</mark> QLRRIL	180

# Supplementary Figure S1. Protein sequence alignment of wild-type hAID, hAID\* $\Delta$ , and AID10.

Amino acid mutations are shadowed in yellow and truncations are indicated by dash lines.

	OsER1-T1	
WT	5' <u>AGAACTCGAGTCGCAAGTCG<b>AGG</b></u> 3'	Clone number
		6/0
APOBEC1-nCas9-0GI	5AGAATIIGAGICGCAAGICGAGG5.	6/9
	5'AGAACTTGAGTCGCAAGTCG <b>AGG</b> 3'	2/9
	5'AGAACT <mark>T</mark> GAGTTGCAAGTCG <b>AGG</b> 3'	1/9
AID10-nCas9-UGI	5'AGAATTCGAGTCGCAAGTCG <b>AGG</b> 3'	9/14
	5'AGAATTTGAGTCGCAAGTCG <b>AGG</b> 3'	3/14
	5'AGAATTTGAGTTGCAAGTCG <b>AGG</b> 3'	1/14
	5'AGAACTTGAGTCGTAAGTCG <b>AGG</b> 3'	1/14
AID10-nCas9	5'AGAATTCGAGTCGCAAGTCG <b>AGG</b> 3'	9/12
	5'AGAATTTGAGTCGCAAGTCG <b>AGG</b> 3'	2/12
	5'AGAA <mark>G</mark> TCGAGTCGCAAGTCG <b>AGG</b> 3'	1/12

Supplementary Figure S2. *Xho*I-resistant mutant alleles of the *OsER1-T1* locus generated by APOBEC1-nCas9-UGI, AID10-nCas9-UGI, or AID10-nCas9 in protoplasts.

*Xho*I-resistant amplicons of the *OsER1-T1* locus induced by APOBEC1-nCas9-UGI, AID10-nCas9-UGI, or AID10-nCas9, were cloned and randomly selected for Sanger sequencing. The abundances of individual mutant alleles are indicated by the corresponding clone numbers divided by the total sequenced clone numbers. The target sequence and the PAM are underlined and in bold, respectively. The *Xho*I restriction site is marked in green and the C-to-T substitutions in mutant alleles are highlighted in red.



Supplementary Figure S3. Mutant allele compositions of three target sites edited by APOBEC1-nCas9-UGI, AID10-nCas9-UGI, or AID10-nCas9 in protoplasts.

C-to-T (or G-to-A) conversions are highlighted in bold and the frequencies of individual alleles are shown on the right. Dashed-line box marks the target sequence and the PAM is underlined. The numbers on the top indicate the protospacer positions. Data are collected from one biological replicate in Figure 1C.



Supplementary Figure S4. Base editing at two genomic loci by APOBEC1-nCas9-UGI or AID10-nCas9-UGI in transgenic rice plants.

Sanger sequencing chromatograms evidence cytosine editing at the *OsLAZY1* (**A**) or *OsGL1-1* (**B**) locus by APOBEC1-nCas9-UGI (left) or AID10-nCas9-UGI (right) in transgenic T0 rice lines. Reverse complements of the target sequences and PAMs are underlined and in bold, respectively. Red arrows mark base substitutions.





(A) Two rice *lazy1* mutant alleles (lines #20 and #25) generated by AID10-nCas9-UGI. Sequences of the intron 1 and exon 2 are shown as black lowercase and blue uppercase letters, respectively. Reverse complements of the target sequence and PAM are underlined and in bold, respectively. Base conversions are highlighted in red. (B) Disruption of the intron splice site in *OsLAZY1* leads to increased tiller angle, resembling the reported rice *lazy1* null plants. Two-month-old plants are shown. Scale bar = 20 cm.



Supplementary Figure S6. Base editing at the *AtCER10* locus by APOBEC1-nCas9-UGI or AID10-nCas9-UGI in transgenic *Arabidopsis* plants.

Sanger sequencing chromatograms evidence cytosine editing at the *AtCER10* locus by APOBEC1-nCas9-UGI (left) or AID10-nCas9-UGI (right) in multiple transgenic T1 *Arabidopsis* lines. The expression of base editors was driven by the *ZmUbi* promoter. Reverse complements of the target sequence and PAM are underlined and in bold, respectively. Intronic and exonic sequences are shown as black lowercase and blue uppercase letters, respectively. Red arrows mark base substitutions.



Supplementary Figure S7. Disruption of the intron splice site in *AtCER10* by AID10-nCas9-UGI produces gene knockout phenotype in transgenic *Arabidopsis* plants.

(A) Two T2 homozygous mutant alleles (lines #41-16 and #61-3) generated by AID10-nCas9-UGI. The expression of the base editor was driven by the egg cell-specific *EC1.2en-EC1.1p* promoter. Sequences of the intron 1 and neighboring exons are shown as lowercase and uppercase letters, respectively. Reverse complements of the target sequence and PAM are underlined and in bold, respectively. Base conversions are highlighted in red. (B) Sanger sequencing chromatograms show base substitutions (marked by red arrows) at the *AtCER10* target site in the two T2 mutant lines. (C) Disruption of the intron splice site in *AtCER10* leads to dwarfism, resembling the reported *Arabidopsis cer10* null plants. Four-week-old plants are shown. Scale bar = 1 cm.



# Supplementary Figure S8. Base editing at three genomic loci by AID10-nSpCas9-UGI or AID10-nSaCas9-UGI in transgenic *Arabidopsis* plants.

Sanger sequencing chromatograms for the AtHDC1 (A), AtABI3 (B) or At-eTM166-1 (C) locus in representative transgenic T1 lines edited by AID10-nSpCas9-UGI (left) or AID10-nSaCas9-UGI (right). In (C), AID10-nSaCas9-UGI (not shown) failed to induce any detectable base change at the At-eTM166-1 locus. The expression of base editors was driven by the egg cell-specific EC1.2en-EC1.1p promoter. The target sequences are underlined and the PAMs are in bold. Red arrows mark base substitutions.



Supplementary Figure S9. Amplicon deep sequencing reveals no guide-dependent off-target editing by AID10-nSpCas9-NG-UGI and AID10-nSaCas9-KKH-UGI in protoplasts.

(A) No guide-dependent (no more than 3 base mismatches) off-target editing is detected for AID10-nSpCas9-NG-UGI when targeting the *At-eTM166-1*, *AtFLS2*, and *AtCERK1* sites. No potential off-target site was identified for the *AtTPST* site. (B) No guide-dependent (no more than 3 base mismatches) off-target editing is detected for AID10-nSaCas9-KKH-UGI when targeting the *AtFLS2*, *AtCERK1*, and *AtTPST* sites. No potential off-target site was identified for the *At-eTM166-1* site. Mismatches between off-target and on-target sites are highlighted in red. The PAM is shown in green. Data are shown as mean  $\pm$  s.d. of two biological replicates performed at different times.



Supplementary Figure S10. Base editing at the *OsSLR1* locus by AID10-nCas9-AID10-UGI in transgenic rice plants.

Three transgenic T0 rice lines containing cytosine changes at the *OsSLR1* target site were generated by AID10-nCas9-AID10-UGI. The target sequence is underlined and the PAM is in bold. C-to-T base substitutions are highlighted in red.



# Supplementary Figure S11. Base editing at the *OsCKX2* locus by ABE7.10-nCas9-AID10 in transgenic rice plants.

Two transgenic T0 rice lines edited by the dual base editor ABE7.10-nCas9-AID10 only contained cytosine changes at the *OsCKX2* target site. The line #19 contained chimeric mutations. The target sequence is underlined and the PAM is in bold. Base substitutions or deletions are highlighted in red.



# Supplementary Figure S12. Simultaneous editing of cytosines and adenines at the same target sequence by AID10- and TadA7.10-based dual base editors.

(A) Schematic diagram of four AID10- and TadA7.10-based dual base editors with different protein architectures. (B) Comparison of C-to-T (top) and A-to-G (bottom) editing efficiencies between the four dual base editors at the *AtChr4* locus in *Arabidopsis* protoplasts by amplicon deep sequencing. Data are shown as mean  $\pm$  s.d. of two biological replicates performed at different times. (C) Comparison of editing product compositions at the *AtChr4* locus generated by the four dual base editors. (D) The ten most abundant mutant alleles with concurrent C-to-T/G/A and A-to-G substitutions by individual dual base editors. Data of one replicate in (C) are shown. The A-to-G, C-to-T, C-to-G, and C-to-A substitutions are highlighted in blue, red, orange, and green, respectively.



Supplementary Figure S13. Luciferase reporter assay reflects inhibitory effects of different *AtBAK1* uORF mutations on the primary ORF.

(A) Schematic diagram of six *AtBAK1-LUC* reporters containing no uORF (WT) or different combinations of uORF mutations in the 5' UTR of *AtBAK1*. Two possible target sequences in the 5' UTR that can be used to introduce the uORF mutations through base editing are underlined. Their PAMs are indicated in bold. The native ACG trinucleotides and the artificially created start codons are highlighted in blue and red, respectively, with subscripted numbers indicating the protospacer positions. (B) Inhibition of luciferase expression by different uORFs. The reporter constructs were transiently expressed in *Arabidopsis* protoplasts. Data are presented as means  $\pm$  s.d. of three biological replicates. n.s., not significant (two-tailed Student's *t* test). (C) Sanger sequencing chromatograms evidence the creation of uORF-2 in the 5' UTR of *AtBAK1* by AID10-nCas9-UGI in two transgenic T1 lines. Red arrows mark base substitutions.



Supplementary Figure S14. AID10 cytosine deaminase shows no sequence context bias in plant cells.

C-to-T editing frequencies are shown as mean  $\pm$  s.e.m. with individual data points representing the average editing frequency of a single cytosine within the optimal activity window (positions -1–9) of each protospacer. Data shown in Figures 1C, 2B, and 3B for AID10-nCas9-UGI and in Figure 2D for AID10-nCas9-NG-UGI are used for this analysis. n.s., not statistically significant (one-way ANOVA with Tukey's multiple comparisons test).

Target site	Gene ID or genomic location	Target sequence (5'-3') <sup>1</sup>		
OsER1-T1		AGAACTCGAGTCGCAAGTCG <b>AGG</b>		
OsER1-T2	$LOC_{Os00g10230}$	CACTCGCCTATGACATGTGG <b>TGG</b>		
OsLAZY1	LOC_Os11g29840	GCTGATCATATACACATATG <b>AGG</b>		
OsGL1-1	LOC_Os09g25850	CAAATACCTTGTACGCGCCG		
		ACCGTCACGACTGTAGTTAG <b>GGG</b>		
OsNAL1	LOC_Os04g52479	ACCGTCACGACTGTAGTTAG <b>GGGAGT</b>		
		<b>TTTC</b> CACCGTCACGACTGTAGTTAGGG		
		ACCACACCCTCCCTACGTTG <b>GGG</b>		
OsTCP21	LOC_Os07g05720	TAACCACACCCTCCCTACGT <b>TGGGGT</b>		
		<b>TTTG</b> TTGTAACCACACCCTCCCTACGT		
OsALS	LOC_Os02g30630	ATCTCCATGGACGCGCCGCC		
OsSWEET11	LOC_Os08g42350	TGTACACCACCAAAAGTGGA <b>GGG</b>		
OsSWEET13	LOC_Os12g29220	GGAGTTGTGGTGCTTTATAT <b>AGG</b>		
OsSWEET14	LOC_Os11g31190	GCTTAGCACCTGGTTGGAGG <b>GGG</b>		
OsSLR1	LOC_Os03g49990	CCCCTCGGACCTCTCCTCCT		
OsIPA1	LOC_Os08g39890	AGAGAGAGCACAGCTCGAGT <u>CGG</u>		
OsCKX2	LOC_Os01g10110	GAGCTCAAGCTCCGCGCCGC		
AtCNGC16	At3g48010	CGCCACCCTTCCTCTTCCTC		
AtMTA	At4g10760	CCTCCCCACATCTCAGGTCT <u>CGG</u>		
AtCER10	At3g55360	ACCGAATCGGGGAGGTCAAG <b>GGG</b>		
		CGCCGGAAAAATGTCCATCC <b>TGG</b>		
AtBIK1	At2g39660	CGCCGGAAAAATGTCCATCC		
		<b>TTTC</b> AAGTAAACCGCCGGAAAAATGTC		
		CCTCTCCCGTTCCCACCTAT <b>CGG</b>		
AtHDC1	At5g08450	TCCTCTCCCGTTCCCACCTA <b>TCGGAT</b>		
		<b>TTTC</b> TGCTCCATCCTCTCCCGTTCCCA		
44012	4.2.24(50	TGCACGAGAAGTGGCACACT <u>CGG</u>		
ATABI3	A13g24030	ATGCACGAGAAGTGGCACAC		
	chr5:2532762725327649	TCCCCATTTAGCGTCATCAA <b>TGG</b>		
Al-e1M100-1	chr5:2532762425327649	TCCCCATTTAGCGTCATCAA <b>TGGAAT</b>		
14EL 93	4+5~46220	ATTCTCTGCAGAATCAGACA <b>AGT</b>		
AIF L52	AIJg40330	ATTCTCTGCAGAATCAGACA		
A+CEDV1	4+2~21620	CTTCTACTAGTCTCCAAAGT <b>GGA</b>		
AICEKKI	AI3g21030	CTTCTACTAGTCTCCAAAGT		

# Supplementary Table S1. Summary of editing targets in this study

AtTPST	4+1-09020	GCTCCACATCGAGGTTGTTA <b>AGC</b>	
	Allg08050	GCTCCACATCGAGGTTGTTA <b>AGCGAT</b>	
AtBAK1	444-22420	CCTACACCGCCATCACCTGC <b>AGG</b>	
AtBAK1-5'UTR	Al4g55450	ACGGGTGGATACGCGTTGAG <b>AGG</b>	
AtChr3	Chr3: 951091951113	ACACACACACACTCCTCT	
AtChr4	Chr4: 1830223218302254	CACACACACCGCAACAGA <b>CGG</b>	

<sup>1</sup> The PAM sequence is in bold and underlined. Sequences with a 3'-NGG or 3'-NGN PAM, a 3'-NNGRRT or 3'-NNNRRT PAM, and a 5'-TTTN PAM are targets for base editors derived from SpCas9, SaCas9 and LbCas12a, respectively, where "N" refers to any type of nucleotide and "R" stands for "A" or "G".

Primer name	Primer sequence (5'-3')	Application	
OsER1-T1-F	TTGAATACCGGAGTAAGGTGGTG	RFLP assay analysis of the	
OsER1-T1-R	ATGGCCGATGGACACTGAGA	OsER1-T1 site	
OsER1-T2-NGS-F	NNNNNCAGCTACTCGTAAGCTACACTTAT	Amplicon deep sequencing of the <i>OsER1-T2</i> site	
OsER1-T2-NGS-R	TGGCCGATGGACACTGAGAT		
OsNAL1-NGS-F	NNNNNTTGTACGAGCTGACGGTGCATT	Amplicon deep sequencing	
OsNAL1-NGS-R	GCATAGGCCATCACAGTCCCAG	of the OsNAL1 site	
AtCNGC16-NGS-F	NNNNNGTGAAATCGCTATCCGTTAC	Amplicon deep sequencing	
AtCNGC16-NGS-R	AAACCAAATCATTATCTGAGA	of the AtCNGC16 site	
AtMTA-NGS-F	NNNNNTGCATTCCTCTTGGTGCATTC	Amplicon deep sequencing	
AtMTA-NGS-R	GCCTAGGATCATGGTTTTAGC	of the AtMTA site	
OsTCP21-NGS-F	NNNNNGCCAATCGCACGGTTTCTGC	Amplicon deep sequencing	
OsTCP21-NGS-R	GACATTGGGGGTTAGCGTCC	of the OsTCP21 site	
AtBIK1-NGS-F	NNNNNCACGGTTTGTCCAGAACCGC	Amplicon deep sequencing	
AtBIK1-NGS-R	ACAACAACTTCAGGACAACTTGGGA	of the AtBIK1 site	
AtHDC1-NGS-F	NNNNNAGTCCTTGGCTCTCTTTTGATATGATCTTT	Amplicon deep sequencing	
AtHDC1-NGS-R	AAGGTTGATGCCAAAAACGAAGATAGATT	of the AtHDC1 site	
At-eTM166-NGS-F	NNNNNCGATGATGATGATGATGGAAGCTA	Amplicon deep sequencing	
At-eTM166-NGS-R	TAGTTCGACGATACATTCCATGAG	of the At-eTM166-1 site	
AtFLS2-NGS-F	NNNNNACAAAGGTCAGCTAGAAGATGGG	Amplicon deep sequencing	
AtFLS2-NGS-R	AATGGTGTCCTCCAAGTTTCCAT	of the AtFLS2 site	
AtCERK1-NGS-F	NNNNNTGTCTCTTTTGTTAGCTGGGAGT	Amplicon deep sequencing	
AtCERK1-NGS-R	TGCATAGTAAACAGCCCCAAAAC	of the <i>AtCERK1</i> site Amplicon deep sequencing	
AtTPST-NGS-F	NNNNNTCTAACAGACGCGTACAAGAGTT		
AtTPST-NGS-R	TTAGACTGCACACGAATCATGGA	of the AtTPST site	
AtBAK1-NGS-F	NNNNNGTTTTGCCAACACCAAGTTGACT	Amplicon deep sequencing	
AtBAK1-NGS-R	TAGAAGTGCAGCACCTGCAG	of the AtBAK1 site	
AtC3-NGS-F	NNNNNTCTTAATGTTGAACAAACCATCTAACC	Amplicon deep sequencing	
AtC3-NGS-R	TATTGGCTAAGTGAAGTAATTTGAGATTAT	of the AtChr3 site	
AtC4-NGS-F	NNNNNCATTCAAAACAAAAAGTTTTATCTACATCAC	Amplicon deep sequencing	
AtC4-NGS-R	GAATTGTTTGTTTTTCTTTTTTACATGTACT	of the AtChr4 site	
AtCERK1-Sp-OT1-		Amplicon deep sequencing	
F		of the <i>AtCERK1</i> off-target	
AtCERK1-Sp-OT1-	GCCCGTGATGCATTAAGCTTTTA	site 1 of SpCas9-NG	
R			
AtCERK1-Sp-OT2-	NNNNNCAGTAGCGACGTGGTCTTGA	Amplicon deen sequencing	
F		of the <i>AtCERK1</i> off-target	
AtCERK1-Sp-OT2-	GCCGAATTCGATACGGATTACC	site 2 of SpCas9-NG	
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# Supplementary Table S2. Primers used for amplification of target sites in this study

AtCERK1-Sp-OT3- F	NNNNNTGCTGTAGACTCTTGGTTTGCT	Amplicon deep sequencing of the <i>AtCERK1</i> off-target site 3 of SpCas9-NG	
AtCERK1-Sp-OT3- R	ATTGGTCATGGCGTAGCTGT		
AtCERK1-Sp-OT4- F	NNNNNCAAGACCCAGCCTAGCGAAG	Amplicon deep sequencing of the <i>AtCERK1</i> off-target site 4 of SpCas9-NG	
AtCERK1-Sp-OT4- R	AAGCAGCAGACTCGTCATCTC		
AtCERK1-Sa-OT1- F	NNNNNTCGAGTTTACCCGCATGTTTC	Amplicon deep sequencing	
AtCERK1-Sa-OT1- R	GATGCATAGACTTAAAATTCCACC	of the <i>AtCERK1</i> off-target site 1 of SaCas9-KKH	
AtFLS2-Sa_Sp-OT1 -F	NNNNNTGCCAGAGATTTCCAGAGGC	Amplicon deep sequencing of the <i>AtFLS2</i> off-target	
AtFLS2-Sa_Sp-OT1 -R	TCTCCAGTTTCTTGGAACGCA	site 1of SpCas9-NG and SaCas9-KKH	
AtFLS2-Sp-OT2-F	NNNNNTTATCTTCATCGACTCTTCTCCTT	Amplicon deep sequencing	
AtFLS2-Sp-OT2-R	FLS2-Sp-OT2-R AGCTTCCATCGTTAGTCACAA		
AtFLS2-Sa-OT2-F	NNNNNTCGTTTAGGGGAATCGGACAA	Amplicon deep sequencing	
AtFLS2-Sa-OT2-R	AtFLS2-Sa-OT2-R TCGTCTGTTAATTAGCACCATTCA		
AtFLS2-Sa-OT3-F	NNNNNTGTCATCGAGATCACACTTGAAGA	Amplicon deep sequencing	
AtFLS2-Sa-OT3-R	CACACTCTCATTCTATCTTTCTTCTTCTT	site 3 of SaCas9-KKH	
At-eTM166-Sp-OT1 -F	NNNNNCTGTCCTCACCAACCACCAG	Amplicon deep sequencing of the <i>At-eTM166-1</i>	
At-eTM166-Sp-OT1 -R	GAGAGGTTGCAGGAGAACCC	off-target site 1 of SpCas9-NG	
AtTPST-Sa-OT1-F	NNNNNTGACACAGGTGAGTGAGAGC	Amplicon deep sequencing	
AtTPST-Sa-OT1-R	AtTPST-Sa-OT1-R CGACAAACAGGGACGCAATG		
AtTPST-Sa-OT2-F	NNNNNGTGGATTTGCAGTTTCCGACC	Amplicon deep sequencing	
AtTPST-Sa-OT2-R	AtTPST-Sa-OT2-R AAAGCGACGTCGTCTCGG		
OsLAZY1-HiTOM-	GGAGTGAGTACGGTGTGCCCTCCGGTGTTGAACTCTT		
F	TGAA CACTTCCATCCTCCATCCTACCTCATCATCCTTCTC	HiTOM sequencing of the	
R	GGTTGAAT	OSLAZI I SILE	
OsLAZY1-F	TCTTAGCACGCTAAACCGGC	Sanger sequencing of the	
OsLAZY1-R	AGTGCGTTTGTGTTACGAGC	OsLAZY1 site	

OsGL1-1-HiTOM-F	GGAGTGAGTACGGTGTGCAGGACGAAGAAGAAGATC	HiTOM sequencing of the	
	AAGC		
OsGL1-1-HiTOM-R	JL1-1-HiTOM-R GAGITGGATGCTGGATGGCACATACTGCGCACACACA AT		
OsGL1-1-F	OsGL1-1-F AACATCTTCTAACTCCCCTCACG		
OsGL1-1-R	TGCACCTGCAGTATCAAGAAGTT	OsGL1-1 site	
AtCER10-HiTOM-F	GGAGTGAGTACGGTGTGCTTAAGCACCATGAAGGTCA CCG	HiTOM sequencing of the	
AtCER10-HiTOM-R	GAGTTGGATGCTGGATGGCGAAGCACAAAACACACG AAAT	AtCER10 site	
AtCER10-F	GGTGGATCCTTAAAACAAAGCCA	Sanger sequencing of the	
AtCER10-R	CGAAGCACAAAACACACGAAATC	AtCER10 site	
AtHDC1-HiTOM-F	GGAGTGAGTACGGTGTGCCCCAAAGAGATTGGAAGA GAAG	HiTOM sequencing of the	
AtHDC1-HiTOM-R	GAGTTGGATGCTGGATGGTTCTCAGCACCAGTCCTTG G	<i>AtHDC1</i> site	
AtHDC1-F	CCCAAAGAGATTGGAAGAGAAG	Sanger sequencing of the	
AtHDC1-R	TTCTCAGCACCAGTCCTTGG	AtHDC1 site	
AtABI3-HiTOM-F	GGAGTGAGTACGGTGTGCGTACCAAATAAGAAAGAG GG	HiTOM sequencing of the	
AtABI3-HiTOM-R	GAGTTGGATGCTGGATGGCCAGAAATATAACACAAA CACC	AtABI3 site	
AtABI3-F	GTACCAAATAAGAAAGAGGG	Sanger sequencing of the	
AtABI3-R	ACGATATGTAAGCTCGACTC	AtABI3 site	
At-eTM166-HiTOM	GGAGTGAGTACGGTGTGCATTCCTTTCAAAATAAAGG		
-F	TCG	HiTOM sequencing of the	
At-eTM166-HiTOM	GAGTTGGATGCTGGATGGATGATGATGATGATGGAAG	At-eTM166-1 site	
-R	С		
At-eTM166-F	ATTCCTTTCAAAATAAAGGTCG	Sanger sequencing of the	
At-eTM166-R	ATGATGATGATGATGGAAGC	At-eTM166-1 site	
OsALS-HiTOM-F	GGAGTGAGTACGGTGTGCCGCCGCGAACGCCTCG	HiTOM sequencing of the	
OsALS-HiTOM-R	GAGTTGGATGCTGGATGGCGGCCGAGCCCCGCAA	OsALS site	
OsALS-F	CACCACCACCATGGCT	Sanger sequencing of the	
OsALS-R	GTCCTCCACATCAAGGACAAGGTAA	OsALS site	
OsSWEET11-HiTO	GGAGTGAGTACGGTGTGCAGTGAAAAAGAAATATCA		
M-F	AGCACAAG	HiTOM sequencing of the	
OsSWEET11-HiTO	GAGTTGGATGCTGGATGGAGGTGTTAATCAGTGAGAA	OsSWEET11 site	
M-R	GGTTAG		
OsSWEET11-F	TGATTGCACACGAACTACTCTG	Sanger sequencing of the	
OsSWEET11-R	GCAATGGTGCAGACAACAACTA	OsSWEET11 site	

OsSWEET13-HiTO	GGAGTGAGTACGGTGTGCAGGGAGAGCTCTGAAGAA		
M-F	GA	HiTOM sequencing of the	
OsSWEET13-HiTO GAGTTGGATGCTGGATGGCTCCAACAGTAGCTCATTT		OsSWEET13 site	
M-R	TTGTAA		
OsSWEET13-F	TCGATCTCTACTGACAATGCAC	Sanger sequencing of the	
OsSWEET13-R	AGTACCATCCATATTGCCTTCG	OsSWEET13 site	
OsSWEET14-HiTO	GGAGTGAGTACGGTGTGCGCTGGTAATCAGCTACTAC		
M-F	TTG	HiTOM sequencing of the	
OsSWEET14-HiTO	GAGTTGGATGCTGGATGGCTGATGATTATCTT	OsSWEET14 site	
M-R			
OsSWEET14-F	GCCCAACTCTAGATCCCTTAAC	Sanger sequencing of the	
OsSWEET14-R	CCACTCACAATTGCATCCAAAA	OsSWEET14 site	
OsSLR1-HiTOM-F	GGAGTGAGTACGGTGTGCCGCGGATGACGGGTTCG	HiTOM sequencing of the	
OsSLR1-HiTOM-R	GAGTTGGATGCTGGATGGTGGAAGCATGGCGGG	OsSLR1 site	
OsSLR1-F	DsSLR1-F GCCGCGGATGACGGGTTCG		
OsSLR1-R	DsSLR1-R GGTGGAAGCATGGCGGG		
OsCKX2-HiTOM-F	GGAGTGAGTACGGTGTGCGCGCGGGGTTCGTGTTCG	HiTOM sequencing of the	
OsCKX2-HiTOM-R	GAGTTGGATGCTGGATGGGATGCCGTGGAAGACGC	OsCKX2 site	
OsCKX2-F	ATCCGAGAAGATCCTGCATG	Sanger sequencing of the	
OsCKX2-R	CTTAGTACACACTATTTGCAGTCAAA	OsCKX2 site	
	GGAGTGAGTACGGTGTGCGCTGGTGGTAGTGGACATG		
USIPAT-HITUM-F	G	HiTOM sequencing of the	
OcIDA 1 HITOM P	GAGTTGGATGCTGGATGGTCGCTGGCCCAAATCTCCC	OsIPA1 site	
OSIFAT-HITOWI-K	Т		
OsIPA1-F	GTTCCAAGCAGCGTAAGGAATG	Sanger sequencing of the	
OsIPA1-R	AGACTTCATGTGGTAGCTGGTG	OsIPA1 site	
AtBAK1-5'UTR-Hi	GGAGTGAGTACGGTGTGCGGAAATATCTTCTTCCTCC		
TOM-F	TTTCG	HiTOM sequencing of the	
AtBAK1-5'UTR-Hi	GAGTTGGATGCTGGATGGTCGATTAAGCTACCACCCA	AtBAK1-5'UTR site	
TOM-R	СА		
AtBAK1-5'UTR-F	GGAAATATCTTCTTCCTCCTTTCG	Sanger sequencing of the	
AtBAK1-5'UTR-R TATCCTCAAGAGATTAAAAAACAAACCC		AtBAK1 site	

Primer name	Primer sequence (5'-3')
AtPHV-qPCR-F	CTGAGTGTTGACAAGCTCGA
AtPHV-qPCR-R	TCTTCTCTCGATTGCGGA
AtPHB-qPCR-F	AGGAGCATACATCTGCGTGT
AtPHB-qPCR-R	TTTCTATAGCAGAGGAGGCCC
AtCNA-qPCR-F	CAGCCAGAAATCGCGTGGT
AtCNA-qPCR-R	CAGCACCAATTGGCATCTCAA

Supplementary Table S3. Primers used for qPCR analysis in this study

Target gene	Base editor (BE)	Promoter for BE expression	No. of transgenic plants	Genotype and No. <sup>1</sup>	Base editing efficiency <sup>2</sup>	Indel ratio <sup>3</sup>
Oct 47V1	APOBEC1-nCas9-UGI	ZmUbi	40	11WT/7Homo/3Het/15Bi/4Chi	65.0% (26/40)	42.5% (17/40)
<i>OSLAL11</i>	AID10-nCas9-UGI	ZmUbi	33	8WT/4Homo/1Het/14Bi/6Chi	72.7% (24/33)	21.2% (7/33)
0-011	APOBEC1-nCas9-UGI	ZmUbi	27	12WT/4Homo/2Het/8Bi/1Chi	51.9% (14/27)	11.1% (3/27)
USGL1-1	AID10-nCas9-UGI	ZmUbi	47	7WT/5Homo/16Het/18Bi/1Chi	83.0% (39/47)	14.9% (7/47)
	APOBEC1-nCas9-UGI	ZmUbi	42	14WT/28Chi	64.3% (27/42)	4.8% (2/42)
AtCER10	AID10-nCas9-UGI	ZmUbi	34	2WT/32Chi	94.1% (32/34)	0% (0/34)
	AID10-nCas9-UGI	EC1.2en-EC1.1p	157	153WT/4Het	2.5% (4/157)	0% (0/157)
AtHDC1	AID10-nCas9-UGI	EC1.2en-EC1.1p	118	108WT/8Het/2Bi	8.5% (10/118)	0% (0/118)
	AID10-nSaCas9-UGI	EC1.2en-EC1.1p	57	54WT/1Homo/2Het	5.3% (3/57)	0% (0/57)
444012	AID10-nCas9-UGI	EC1.2en-EC1.1p	107	103WT/4Het	3.7% (4/107)	0% (0/107)
AtABI3	AID10-nSaCas9-UGI	EC1.2en-EC1.1p	101	75WT/2Homo/16Het/6Bi/2Chi	17.8% (18/101)	9.9% (10/101)
	AID10-nCas9-UGI	EC1.2en-EC1.1p	71	63WT/3Het/5Bi	11.3% (8/71)	0% (0/71)
At-eTM166-1	AID10-nSaCas9-UGI	EC1.2en-EC1.1p	139	139WT	0% (0/139)	0% (0/139)
OsALS	nCas9-AID10-UGI	ZmUbi	39	33WT/5Het/1Bi	12.8% (5/39)	2.6% (1/39)
OsSWEET11	AID10-nCas9-AID10-UGI	ZmUbi	77	38WT/3Homo/14Het/20Bi/2Chi	36.4% (28/77)	26.0% (20/77)
OsSWEET13	AID10-nCas9-AID10-UGI	ZmUbi	77	69WT/1Homo/6Het/1Bi	9.1% (7/77)	0% (0/77)

Supplementary Table S4. Summary of base editing outcomes in transgenic Arabidopsis T1 and rice T0 plants

OsSWEET14	AID10-nCas9-AID10-UGI	ZmUbi	77	38WT/2Homo/11Het/21Bi/5Chi	41.6% (32/77)	26.0% (20/77)
OsSLR1	AID10-nCas9-AID10-UGI	ZmUbi	56	52WT/4Het	5.4% (3/56)	1.8% (1/56)
OsIPA 1	ABE7.10-nCas9-AID10	ZmUbi	29	29WT	0% (0/29)	0% (0/29)
OsCKX2	ABE7.10-nCas9-AID10	ZmUbi	23	21WT/1Het/1Chi	8.7% (2/23)	4.3% (1/23)
AtBAK1-5'UTR	AID10-nCas9-UGI	EC1.2en-EC1.1p	325	320WT/5Het	1.5% (5/325)	0% (0/325)

<sup>1</sup> Homo, Het, Bi, and Chi represent homozygote, heterozygote, biallele, and chimera, respectively.

<sup> $^{2}$ </sup> Base editing efficiency is defined as the ratio of the number of transgenic lines with base substitution(s) to the number of total lines.

<sup>3</sup> Indel ratio is the percentage of transgenic lines with nucleotide insertion or deletion (indel). Numbers of plants with indels to total transgenic plants are given in parentheses.

Base editor	Cas effector	РАМ	Optimal editing window <sup>1</sup>	Possible editing window <sup>1</sup>
AID10-nCas9-UGI	nSpCas9	NGG	C <sub>-1</sub> C <sub>9</sub>	C-5-C15
AID10-nCas9-NG-UGI	nSpCas9-NG	NG	C <sub>-1</sub> –C <sub>9</sub>	C-5-C15
AID10-nSaCas9-UGI	nSaCas9	NNGRRT	C <sub>-2</sub> C <sub>12</sub>	C-5-C16
AID10-nSaCas9-KKH-UGI	nSaCas9-KKH	NNNRRT	C <sub>-2</sub> C <sub>12</sub>	C-5-C16
nCas9-AID10-UGI	nSpCas9	NGG	$C_{12} - C_{14}$	$C_{12}$ - $C_{14}$
AID10-nCas9-AID10-UGI	nSpCas9	NGG	C-1-C14	$C_{-1}-C_{14}$
ABE8e-nCas9-AID10-UGI	nSpCas9	NGG	A <sub>4</sub> -A <sub>8</sub> , C <sub>11</sub> -C <sub>14</sub>	$A_2 - A_8, C_9 - C_{14}$
AID10-ABE8e-nCas9-UGI	nSpCas9	NGG	$A_4 - A_8, C_{-1} - C_{11}$	A <sub>2</sub> -A <sub>8</sub> , C <sub>-1</sub> -C <sub>14</sub>

Supplementary Table S5. Summary of base editors developed in this study

<sup>1</sup> The subscripted numbers represent the protospacer positions, counting the PAM as 21–23.

Supplementary Sequence. Amino acid sequences of base editors tested in this study.

# AID10-dCas9

(NLS-AID10-XTEN-dCas9-NLS-2×FLAG)

MAPKKKRKVGIHGVPAADSLLMNRREFLYQFKNVRWAKGRRETYLCYVVERRDCATSFSLDF GYLRNKNGCHVELLFLRYISDWDLDPGRCYRVTWFISWSPCYDCARHVADFLRGNPNLSLRI FAARLYFCEDRKAEPEGLRRLRRAGVQIAIMTFKDYFYCWNTFAENHGRTFKAWEGLHENSV RLSGOLRRILSGSETPGTSESATPESDKKYSIGLAIGTNSVGWAVITDEYKVPSKKFKVLGN TDRHSIKKNLIGALLFDSGETAEATRLKRTARRRYTRRKNRICYLQEIFSNEMAKVDDSFFH RLEESFLVEEDKKHERHPIFGNIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRLIYLALAHM IKFRGHFLIEGDLNPDNSDVDKLFIOLVOTYNOLFEENPINASGVDAKAILSARLSKSRRLE NLIAQLPGEKKNGLFGNLIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDNLLAQIGDQ YADLFLAAKNLSDAILLSDILRVNTEITKAPLSASMIKRYDEHHQDLTLLKALVRQQLPEKY KEIFFDQSKNGYAGYIDGGASQEEFYKFIKPILEKMDGTEELLVKLNREDLLRKQRTFDNGS IPHOIHLGELHAILRROEDFYPFLKDNREKIEKILTFRIPYYVGPLARGNSRFAWMTRKSEE TITPWNFEEVVDKGASAQSFIERMTNFDKNLPNEKVLPKHSLLYEYFTVYNELTKVKYVTEG MRKPAFLSGEQKKAIVDLLFKTNRKVTVKQLKEDYFKKIECFDSVEISGVEDRFNASLGTYH DLLKIIKDKDFLDNEENEDILEDIVLTLTLFEDREMIEERLKTYAHLFDDKVMKQLKRRRYT GWGRLSRKLINGIRDKQSGKTILDFLKSDGFANRNFMQLIHDDSLTFKEDIQKAQVSGQGDS LHEHIANLAGSPAIKKGILQTVKVVDELVKVMGRHKPENIVIEMARENQTTQKGQKNSRERM KRIEEGIKELGSQILKEHPVENTQLQNEKLYLYYLQNGRDMYVDQELDINRLSDYDVD<mark>A</mark>IVP QSFLKDDSIDNKVLTRSDKNRGKSDNVPSEEVVKKMKNYWRQLLNAKLITQRKFDNLTKAER GGLSELDKAGFIKRQLVETRQITKHVAQILDSRMNTKYDENDKLIREVKVITLKSKLVSDFR KDFQFYKVREINNYHHAHDAYLNAVVGTALIKKYPKLESEFVYGDYKVYDVRKMIAKSEQEI GKATAKYFFYSNIMNFFKTEITLANGEIRKRPLIETNGETGEIVWDKGRDFATVRKVLSMPQ VNIVKKTEVQTGGFSKESILPKRNSDKLIARKKDWDPKKYGGFDSPTVAYSVLVVAKVEKGK SKKLKSVKELLGITIMERSSFEKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELENGRKRMLA SAGELQKGNELALPSKYVNFLYLASHYEKLKGSPEDNEQKQLFVEQHKHYLDEIIEQISEFS KRVILADANLDKVLSAYNKHRDKPIREQAENIIHLFTLTNLGAPAAFKYFDTTIDRKRYTST KEVLDATLIHQSITGLYETRIDLSQLGGDKRPAATKKAGQAKKKKRPPPSDYKDDDDKDYKD DDDK\*

# AID10-dCas9-UGI

# (NLS-AID10-XTEN-dCas9-(GGS)<sub>6</sub> linker-UGI-NLS-2×FLAG)

MAPKKKRKVGIHGVPAADSLLMNRREFLYQFKNVRWAKGRRETYLCYVVERRDCATSFSLDF GYLRNKNGCHVELLFLRYISDWDLDPGRCYRVTWFISWSPCYDCARHVADFLRGNPNLSLRI FAARLYFCEDRKAEPEGLRRLRRAGVQIAIMTFKDYFYCWNTFAENHGRTFKAWEGLHENSV RLSGQLRRILSGSETPGTSESATPESDKKYSIGL<mark>A</mark>IGTNSVGWAVITDEYKVPSKKFKVLGN TDRHSIKKNLIGALLFDSGETAEATRLKRTARRRYTRRKNRICYLQEIFSNEMAKVDDSFFH RLEESFLVEEDKKHERHPIFGNIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRLIYLALAHM IKFRGHFLIEGDLNPDNSDVDKLFIQLVQTYNQLFEENPINASGVDAKAILSARLSKSRRLE NLIAQLPGEKKNGLFGNLIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDNLLAQIGDQ YADLFLAAKNLSDAILLSDILRVNTEITKAPLSASMIKRYDEHHQDLTLLKALVRQQLPEKY KEIFFDQSKNGYAGYIDGGASQEEFYKFIKPILEKMDGTEELLVKLNREDLLRKQRTFDNGS IPHQIHLGELHAILRRQEDFYPFLKDNREKIEKILTFRIPYYVGPLARGNSRFAWMTRKSEE TITPWNFEEVVDKGASAQSFIERMTNFDKNLPNEKVLPKHSLLYEYFTVYNELTKVKYVTEG MRKPAFLSGEQKKAIVDLLFKTNRKVTVKQLKEDYFKKIECFDSVEISGVEDRFNASLGTYH DLLKIIKDKDFLDNEENEDILEDIVLTLTLFEDREMIEERLKTYAHLFDDKVMKOLKRRRYT GWGRLSRKLINGIRDKQSGKTILDFLKSDGFANRNFMQLIHDDSLTFKEDIQKAQVSGQGDS LHEHIANLAGSPAIKKGILQTVKVVDELVKVMGRHKPENIVIEMARENQTTQKGQKNSRERM KRIEEGIKELGSQILKEHPVENTQLQNEKLYLYYLQNGRDMYVDQELDINRLSDYDVD<mark>A</mark>IVP QSFLKDDSIDNKVLTRSDKNRGKSDNVPSEEVVKKMKNYWRQLLNAKLITQRKFDNLTKAER GGLSELDKAGFIKRQLVETRQITKHVAQILDSRMNTKYDENDKLIREVKVITLKSKLVSDFR KDFQFYKVREINNYHHAHDAYLNAVVGTALIKKYPKLESEFVYGDYKVYDVRKMIAKSEQEI GKATAKYFFYSNIMNFFKTEITLANGEIRKRPLIETNGETGEIVWDKGRDFATVRKVLSMPQ VNIVKKTEVQTGGFSKESILPKRNSDKLIARKKDWDPKKYGGFDSPTVAYSVLVVAKVEKGK SKKLKSVKELLGITIMERSSFEKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELENGRKRMLA SAGELQKGNELALPSKYVNFLYLASHYEKLKGSPEDNEQKQLFVEQHKHYLDEIIEQISEFS KRVILADANLDKVLSAYNKHRDKPIREOAENIIHLFTLTNLGAPAAFKYFDTTIDRKRYTST SDIIEKETGKQLVIQESILMLPEEVEEVIGNKPESDILVHTAYDESTDENVMLLTSDAPEYK PWALVIODSNGENKIKMLSGGSKRPAATKKAGOAKKKKRPPPSDYKDDDDKDYKDDDDK\*

# AID10-nCas9

#### (NLS-AID10-XTEN-nCas9-NLS-2×FLAG)

MAPKKKRKVGIHGVPAADSLLMNRREFLYQFKNVRWAKGRRETYLCYVVERRDCATSFSLDF GYLRNKNGCHVELLFLRYISDWDLDPGRCYRVTWFISWSPCYDCARHVADFLRGNPNLSLRI FAARLYFCEDRKAEPEGLRRLRRAGVQIAIMTFKDYFYCWNTFAENHGRTFKAWEGLHENSV RLSGOLRRILSGSETPGTSESATPESDKKYSIGLAIGTNSVGWAVITDEYKVPSKKFKVLGN TDRHSIKKNLIGALLFDSGETAEATRLKRTARRRYTRRKNRICYLOEIFSNEMAKVDDSFFH RLEESFLVEEDKKHERHPIFGNIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRLIYLALAHM IKFRGHFLIEGDLNPDNSDVDKLFIQLVQTYNQLFEENPINASGVDAKAILSARLSKSRRLE NLIAQLPGEKKNGLFGNLIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDNLLAQIGDQ YADLFLAAKNLSDAILLSDILRVNTEITKAPLSASMIKRYDEHHQDLTLLKALVRQQLPEKY KEIFFDQSKNGYAGYIDGGASQEEFYKFIKPILEKMDGTEELLVKLNREDLLRKQRTFDNGS IPHQIHLGELHAILRRQEDFYPFLKDNREKIEKILTFRIPYYVGPLARGNSRFAWMTRKSEE TITPWNFEEVVDKGASAOSFIERMTNFDKNLPNEKVLPKHSLLYEYFTVYNELTKVKYVTEG MRKPAFLSGEQKKAIVDLLFKTNRKVTVKQLKEDYFKKIECFDSVEISGVEDRFNASLGTYH DLLKIIKDKDFLDNEENEDILEDIVLTLTLFEDREMIEERLKTYAHLFDDKVMKQLKRRRYT GWGRLSRKLINGIRDKØSGKTILDFLKSDGFANRNFMØLIHDDSLTFKEDIØKAØVSGØGDS LHEHIANLAGSPAIKKGILQTVKVVDELVKVMGRHKPENIVIEMARENQTTQKGQKNSRERM KRIEEGIKELGSQILKEHPVENTQLQNEKLYLYYLQNGRDMYVDQELDINRLSDYDVD<mark>H</mark>IVP QSFLKDDSIDNKVLTRSDKNRGKSDNVPSEEVVKKMKNYWRQLLNAKLITQRKFDNLTKAER GGLSELDKAGFIKRQLVETRQITKHVAQILDSRMNTKYDENDKLIREVKVITLKSKLVSDFR KDFQFYKVREINNYHHAHDAYLNAVVGTALIKKYPKLESEFVYGDYKVYDVRKMIAKSEQEI GKATAKYFFYSNIMNFFKTEITLANGEIRKRPLIETNGETGEIVWDKGRDFATVRKVLSMPQ VNIVKKTEVQTGGFSKESILPKRNSDKLIARKKDWDPKKYGGFDSPTVAYSVLVVAKVEKGK SKKLKSVKELLGITIMERSSFEKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELENGRKRMLA SAGELQKGNELALPSKYVNFLYLASHYEKLKGSPEDNEQKQLFVEQHKHYLDEIIEQISEFS KRVILADANLDKVLSAYNKHRDKPIREQAENIIHLFTLTNLGAPAAFKYFDTTIDRKRYTST KEVLDATLIHQSITGLYETRIDLSQLGGDKRPAATKKAGQAKKKKRPPPSDYKDDDDKDYKD DDDK\*

# AID10-nCas9-UGI

(NLS-AID10-XTEN-nCas9-(GGS)<sub>6</sub> linker-UGI-NLS-2×FLAG)

MAPKKKRKVGIHGVPAADSLLMNRREFLYQFKNVRWAKGRRETYLCYVVERRDCATSFSLDF GYLRNKNGCHVELLFLRYISDWDLDPGRCYRVTWFISWSPCYDCARHVADFLRGNPNLSLRI FAARLYFCEDRKAEPEGLRRLRRAGVQIAIMTFKDYFYCWNTFAENHGRTFKAWEGLHENSV RLSGOLRRILSGSETPGTSESATPESDKKYSIGLAIGTNSVGWAVITDEYKVPSKKFKVLGN TDRHSIKKNLIGALLFDSGETAEATRLKRTARRRYTRRKNRICYLQEIFSNEMAKVDDSFFH RLEESFLVEEDKKHERHPIFGNIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRLIYLALAHM IKFRGHFLIEGDLNPDNSDVDKLFIQLVQTYNQLFEENPINASGVDAKAILSARLSKSRRLE NLIAQLPGEKKNGLFGNLIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDNLLAQIGDQ YADLFLAAKNLSDAILLSDILRVNTEITKAPLSASMIKRYDEHHODLTLLKALVROOLPEKY KEIFFDQSKNGYAGYIDGGASQEEFYKFIKPILEKMDGTEELLVKLNREDLLRKQRTFDNGS IPHQIHLGELHAILRRQEDFYPFLKDNREKIEKILTFRIPYYVGPLARGNSRFAWMTRKSEE TITPWNFEEVVDKGASAQSFIERMTNFDKNLPNEKVLPKHSLLYEYFTVYNELTKVKYVTEG MRKPAFLSGEQKKAIVDLLFKTNRKVTVKQLKEDYFKKIECFDSVEISGVEDRFNASLGTYH DLLKIIKDKDFLDNEENEDILEDIVLTLTLFEDREMIEERLKTYAHLFDDKVMKQLKRRRYT GWGRLSRKLINGIRDKQSGKTILDFLKSDGFANRNFMQLIHDDSLTFKEDIQKAQVSGQGDS LHEHIANLAGSPAIKKGILQTVKVVDELVKVMGRHKPENIVIEMARENQTTQKGQKNSRERM KRIEEGIKELGSQILKEHPVENTQLQNEKLYLYYLQNGRDMYVDQELDINRLSDYDVD<mark>H</mark>IVP OSFLKDDSIDNKVLTRSDKNRGKSDNVPSEEVVKKMKNYWROLLNAKLITORKFDNLTKAER GGLSELDKAGFIKRQLVETRQITKHVAQILDSRMNTKYDENDKLIREVKVITLKSKLVSDFR KDFQFYKVREINNYHHAHDAYLNAVVGTALIKKYPKLESEFVYGDYKVYDVRKMIAKSEQEI GKATAKYFFYSNIMNFFKTEITLANGEIRKRPLIETNGETGEIVWDKGRDFATVRKVLSMPQ VNIVKKTEVQTGGFSKESILPKRNSDKLIARKKDWDPKKYGGFDSPTVAYSVLVVAKVEKGK SKKLKSVKELLGITIMERSSFEKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELENGRKRMLA SAGELOKGNELALPSKYVNFLYLASHYEKLKGSPEDNEOKOLFVEOHKHYLDEIIEOISEFS KRVILADANLDKVLSAYNKHRDKPIREOAENIIHLFTLTNLGAPAAFKYFDTTIDRKRYTST SDIIEKETGKQLVIQESILMLPEEVEEVIGNKPESDILVHTAYDESTDENVMLLTSDAPEYK PWALVIODSNGENKIKMLSGGSKRPAATKKAGOAKKKKRPPPSDYKDDDDKDYKDDDDK\*

# AtCDA1-nCas9-UGI

(NLS-AtCDA1-XTEN-nCas9-NLS-UGI-NLS)

MPKKKRKVMDKPSFVIQSKEAESAAKQLGVSVIQLLPSLVKPAQSYARTPISKFNVAVVGLG SSGRIFLGVNVEFPNLPLHHSIHAEQFLVTNLTLNGERHLNFFAVSAAPCGHCRQFLQEIRD APEIKILITDPNNSADSDSAADSDGFLRLGSFLPHRFGPDDLLGKDHPLLLESHDNHLKISD LDSICNGNTDSSADLKOTALAAANRSYAPYSLCPSGVSLVDCDGKVYRGWYMESAAYNPSMG PVQAALVDYVANGGGGGYERIVGAVLVEKEDAVVRQEHTARLLLETISPKCEFKVFHCYEAS GSETPGTSESATPELKDKKYSIGLAIGTNSVGWAVITDEYKVPSKKFKVLGNTDRHSIKKNL IGALLFDSGETAEATRLKRTARRRYTRRKNRICYLQEIFSNEMAKVDDSFFHRLEESFLVEE DKKHERHPIFGNIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRLIYLALAHMIKFRGHFLIE GDLNPDNSDVDKLFIOLVOTYNOLFEENPINASGVDAKAILSARLSKSRRLENLIAOLPGEK KNGLFGNLIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDNLLAQIGDQYADLFLAAKN LSDAILLSDILRVNTEITKAPLSASMIKRYDEHHQDLTLLKALVRQQLPEKYKEIFFDQSKN GYAGYIDGGASQEEFYKFIKPILEKMDGTEELLVKLNREDLLRKQRTFDNGSIPHQIHLGEL HAILRRQEDFYPFLKDNREKIEKILTFRIPYYVGPLARGNSRFAWMTRKSEETITPWNFEEV VDKGASAQSFIERMTNFDKNLPNEKVLPKHSLLYEYFTVYNELTKVKYVTEGMRKPAFLSGE QKKAIVDLLFKTNRKVTVKQLKEDYFKKIECFDSVEISGVEDRFNASLGTYHDLLKIIKDKD FLDNEENEDILEDIVLTLTLFEDREMIEERLKTYAHLFDDKVMKQLKRRRYTGWGRLSRKLI NGIRDKOSGKTILDFLKSDGFANRNFMOLIHDDSLTFKEDIOKAOVSGOGDSLHEHIANLAG SPAIKKGILQTVKVVDELVKVMGRHKPENIVIEMARENQTTQKGQKNSRERMKRIEEGIKEL GSQILKEHPVENTQLQNEKLYLYYLQNGRDMYVDQELDINRLSDYDVD<mark>H</mark>IVPQSFLKDDSID NKVLTRSDKNRGKSDNVPSEEVVKKMKNYWRQLLNAKLITQRKFDNLTKAERGGLSELDKAG FIKRQLVETRQITKHVAQILDSRMNTKYDENDKLIREVKVITLKSKLVSDFRKDFQFYKVRE INNYHHAHDAYLNAVVGTALIKKYPKLESEFVYGDYKVYDVRKMIAKSEQEIGKATAKYFFY SNIMNFFKTEITLANGEIRKRPLIETNGETGEIVWDKGRDFATVRKVLSMPQVNIVKKTEVQ TGGFSKESILPKRNSDKLIARKKDWDPKKYGGFDSPTVAYSVLVVAKVEKGKSKKLKSVKEL LGITIMERSSFEKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELENGRKRMLASAGELOKGNE LALPSKYVNFLYLASHYEKLKGSPEDNEOKOLFVEOHKHYLDEIIEOISEFSKRVILADANL DKVLSAYNKHRDKPIREQAENIIHLFTLTNLGAPAAFKYFDTTIDRKRYTSTKEVLDATLIH QSITGLYETRIDLSQLGGDKRPAATKKAGQAKKKKTRDSGGSTNLSDIIEKETGKQLVIQES ILMLPEEVEEVIGNKPESDILVHTAYDESTDENVMLLTSDAPEYKPWALVIODSNGENKIKM LSGGSPKKKRKV\*

# APOBEC1-nCas9-UGI

# (NLS-APOBEC1-XTEN-nCas9-NLS-UGI-NLS)

MPKKKRKVSSETGPVAVDPTLRRRIEPHEFEVFFDPRELRKETCLLYEINWGGRHSIWRHTS QNTNKHVEVNFIEKFTTERYFCPNTRCSITWFLSWSPCGECSRAITEFLSRYPHVTLFIYIA RLYHHADPRNRQGLRDLISSGVTIQIMTEQESGYCWRNFVNYSPSNEAHWPRYPHLWVRLYV LELYCIILGLPPCLNILRRKQPQLTFFTIALQSCHYQRLPPHILWATGLKSGSETPGTSESA TPELKDKKYSIGLAIGTNSVGWAVITDEYKVPSKKFKVLGNTDRHSIKKNLIGALLFDSGET AEATRLKRTARRRYTRRKNRICYLQEIFSNEMAKVDDSFFHRLEESFLVEEDKKHERHPIFG NIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRLIYLALAHMIKFRGHFLIEGDLNPDNSDVD KLFIQLVQTYNQLFEENPINASGVDAKAILSARLSKSRRLENLIAQLPGEKKNGLFGNLIAL SLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDNLLAQIGDQYADLFLAAKNLSDAILLSDIL RVNTEITKAPLSASMIKRYDEHHQDLTLLKALVRQQLPEKYKEIFFDQSKNGYAGYIDGGAS QEEFYKFIKPILEKMDGTEELLVKLNREDLLRKQRTFDNGSIPHQIHLGELHAILRRQEDFY PFLKDNREKIEKILTFRIPYYVGPLARGNSRFAWMTRKSEETITPWNFEEVVDKGASAQSFI TNRKVTVKOLKEDYFKKIECFDSVEISGVEDRFNASLGTYHDLLKIIKDKDFLDNEENEDIL EDIVLTLTLFEDREMIEERLKTYAHLFDDKVMKQLKRRRYTGWGRLSRKLINGIRDKQSGKT ILDFLKSDGFANRNFMQLIHDDSLTFKEDIQKAQVSGQGDSLHEHIANLAGSPAIKKGILQT VKVVDELVKVMGRHKPENIVIEMARENQTTQKGQKNSRERMKRIEEGIKELGSQILKEHPVE NTQLQNEKLYLYYLQNGRDMYVDQELDINRLSDYDVD<mark>H</mark>IVPQSFLKDDSIDNKVLTRSDKNR GKSDNVPSEEVVKKMKNYWROLLNAKLITORKFDNLTKAERGGLSELDKAGFIKROLVETRO ITKHVAQILDSRMNTKYDENDKLIREVKVITLKSKLVSDFRKDFQFYKVREINNYHHAHDAY LNAVVGTALIKKYPKLESEFVYGDYKVYDVRKMIAKSEQEIGKATAKYFFYSNIMNFFKTEI TLANGEIRKRPLIETNGETGEIVWDKGRDFATVRKVLSMPQVNIVKKTEVQTGGFSKESILP KRNSDKLIARKKDWDPKKYGGFDSPTVAYSVLVVAKVEKGKSKKLKSVKELLGITIMERSSF EKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELENGRKRMLASAGELQKGNELALPSKYVNFL YLASHYEKLKGSPEDNEQKQLFVEQHKHYLDEIIEQISEFSKRVILADANLDKVLSAYNKHR DKPIREQAENIIHLFTLTNLGAPAAFKYFDTTIDRKRYTSTKEVLDATLIHQSITGLYETRI DLSQLGGDKRPAATKKAGQAKKKKTRDSGGSTNLSDIIEKETGKQLVIQESILMLPEEVEEV IGNKPESDILVHTAYDESTDENVMLLTSDAPEYKPWALVIODSNGENKIKMLSGGSPKKKRK <u>\</u>7\*

### AID10-nSaCas9-UGI

(NLS-AID10-XTEN-nSaCas9-(GGS)<sub>6</sub> linker-UGI-NLS-2×FLAG)

MAPKKKRKVGIHGVPAADSLLMNRREFLYQFKNVRWAKGRRETYLCYVVERRDCATSFSLDF GYLRNKNGCHVELLFLRYISDWDLDPGRCYRVTWFISWSPCYDCARHVADFLRGNPNLSLRI FAARLYFCEDRKAEPEGLRRLRRAGVQIAIMTFKDYFYCWNTFAENHGRTFKAWEGLHENSV RLSGQLRRILSGSETPGTSESATPESMKRNYILGLAIGITSVGYGIIDYETRDVIDAGVRLF KEANVENNEGRRSKRGARRLKRRRRHRIORVKKLLFDYNLLTDHSELSGINPYEARVKGLSO KLSEEEFSAALLHLAKRRGVHNVNEVEEDTGNELSTKEQISRNSKALEEKYVAELQLERLKK DGEVRGSINRFKTSDYVKEAKQLLKVQKAYHQLDQSFIDTYIDLLETRRTYYEGPGEGSPFG WKDIKEWYEMLMGHCTYFPEELRSVKYAYNADLYNALNDLNNLVITRDENEKLEYYEKF0II ENVFKOKKKPTLKOIAKEILVNEEDIKGYRVTSTGKPEFTNLKVYHDIKDITARKEIIENAE LLDQIAKILTIYQSSEDIQEELTNLNSELTQEEIEQISNLKGYTGTHNLSLKAINLILDELW HTNDNQIAIFNRLKLVPKKVDLSQQKEIPTTLVDDFILSPVVKRSFIQSIKVINAIIKKYGL PNDIIIELAREKNSKDAOKMINEMOKRNROTNERIEEIIRTTGKENAKYLIEKIKLHDMOEG KCLYSLEAIPLEDLLNNPFNYEVDHIIPRSVSFDNSFNNKVLVKQEE<mark>N</mark>SKKGNRTPFQYLSS SDSKISYETFKKHILNLAKGKGRISKTKKEYLLEERDINRFSVOKDFINRNLVDTRYATRGL MNLLRSYFRVNNLDVKVKSINGGFTSFLRRKWKFKKERNKGYKHHAEDALIIANADFIFKEW KKLDKAKKVMENQMFEEKQAESMPEIETEQEYKEIFITPHQIKHIKDFKDYKYSHRVDKKPN RELINDTLYSTRKDDKGNTLIVNNLNGLYDKDNDKLKKLINKSPEKLLMYHHDPQTYQKLKL IMEQYGDEKNPLYKYYEETGNYLTKYSKKDNGPVIKKIKYYGNKLNAHLDITDDYPNSRNKV VKLSLKPYRFDVYLDNGVYKFVTVKNLDVIKKENYYEVNSKCYEEAKKLKKISNQAEFIASF YNNDLIKINGELYRVIGVNNDLLNRIEVNMIDITYREYLENMNDKRPPRIIKTIASKTOSIK KYSTDILGNLYEVKSKKHPQIIKKGSRADGGGGSGGGGSGGGGSGGGGSGGGGSGGGSTNL SDIIEKETGKQLVIQESILMLPEEVEEVIGNKPESDILVHTAYDESTDENVMLLTSDAPEYK PWALVIODSNGENKIKMLSGGSKRPAATKKAGOAKKKKRPPPSDYKDDDDKDYKDDDDK\*

### AID10-dLbCas12a-UGI

(NLS-AID10-XTEN-dLbCas12a-(GGS)<sub>6</sub> linker-UGI-NLS-2×FLAG)

MAPKKKRKVGIHGVPAADSLLMNRREFLYQFKNVRWAKGRRETYLCYVVERRDCATSFSLDF GYLRNKNGCHVELLFLRYISDWDLDPGRCYRVTWFISWSPCYDCARHVADFLRGNPNLSLRI FAARLYFCEDRKAEPEGLRRLRRAGVQIAIMTFKDYFYCWNTFAENHGRTFKAWEGLHENSV **RLSGOLRRIL**SGSETPGTSESATPESMSKLEKFTNCYSLSKTLRFKAIPVGKTOENIDNKRL LVEDEKRAEDYKGVKKLLDRYYLSFINDVLHSIKLKNLNNYISLFRKKTRTEKENKELENLE INLRKEIAKAFKGNEGYKSLFKKDIIETILPEFLDDKDEIALVNSFNGFTTAFTGFFDNREN MFSEEAKSTSIAFRCINENLTRYISNMDIFEKVDAIFDKHEVQEIKEKILNSDYDVEDFFEG EFFNFVLTOEGIDVYNAIIGGFVTESGEKIKGLNEYINLYNOKTKOKLPKFKPLYKOVLSDR ESLSFYGEGYTSDEEVLEVFRNTLNKNSEIFSSIKKLEKLFKNFDEYSSAGIFVKNGPAIST ISKDIFGEWNVIRDKWNAEYDDIHLKKKAVVTEKYEDDRRKSFKKIGSFSLEQLQEYADADL SVVEKLKEIIIQKVDEIYKVYGSSEKLFDADFVLEKSLKKNDAVVAIMKDLLDSVKSFENYI KAFFGEGKETNRDESFYGDFVLAYDILLKVDHIYDAIRNYVTOKPYSKDKFKLYFONPOFMG GWDKDKETDYRATILRYGSKYYLAIMDKKYAKCLOKIDKDDVNGNYEKINYKLLPGPNKMLP KVFFSKKWMAYYNPSEDIOKIYKNGTFKKGDMFNLNDCHKLIDFFKDSISRYPKWSNAYDFN FSETEKYKDIAGFYREVEEQGYKVSFESASKKEVDKLVEEGKLYMFQIYNKDFSDKSHGTPN LHTMYFKLLFDENNHGQIRLSGGAELFMRRASLKKEELVVHPANSPIANKNPDNPKKTTTLS YDVYKDKRFSEDQYELHIPIAINKCPKNIFKINTEVRVLLKHDDNPYVIGI<mark>A</mark>RGERNLLYIV VVDGKGNIVEOYSLNEIINNFNGIRIKTDYHSLLDKKEKERFEARONWTSIENIKELKAGYI SQVVHKICELVEKYDAVIALEDLNSGFKNSRVKVEKQVYQKFEKMLIDKLNYMVDKKSNPCA TGGALKGYOITNKFESFKSMSTONGFIFYIPAWLTSKIDPSTGFVNLLKTKYTSIADSKKFI SSFDRIMYVPEEDLFEFALDYKNFSRTDADYIKKWKLYSYGNRIRIFRNPKKNNVFDWEEVC LTSAYKELFNKYGINYQQGDIRALLCEQSDKAFYSSFMALMSLMLQMRNSITGRTDVDFLIS PVKNSDGIFYDSRNYEAQENAILPKNADANGAYNIARKVLWAIGQFKKAEDEKLDKVKIAIS ESILMLPEEVEEVIGNKPESDILVHTAYDESTDENVMLLTSDAPEYKPWALVIODSNGENKI KMLSGGSKRPAATKKAGQAKKKKRPPPSDYKDDDDKDYKDDDDK\*

#### AID10-nSpCas9-NG-UGI

# (NLS-AID10-XTEN-nSpCas9-NG-(GGS)<sub>6</sub> linker-UGI-NLS-2×FLAG)

MAPKKKRKVGIHGVPAADSLLMNRREFLYQFKNVRWAKGRRETYLCYVVERRDCATSFSLDF GYLRNKNGCHVELLFLRYISDWDLDPGRCYRVTWFISWSPCYDCARHVADFLRGNPNLSLRI FAARLYFCEDRKAEPEGLRRLRRAGVQIAIMTFKDYFYCWNTFAENHGRTFKAWEGLHENSV RLSGQLRRILSGSETPGTSESATPESDKKYSIGL<mark>A</mark>IGTNSVGWAVITDEYKVPSKKFKVLGN TDRHSIKKNLIGALLFDSGETAEATRLKRTARRRYTRRKNRICYLQEIFSNEMAKVDDSFFH RLEESFLVEEDKKHERHPIFGNIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRLIYLALAHM IKFRGHFLIEGDLNPDNSDVDKLFIQLVQTYNQLFEENPINASGVDAKAILSARLSKSRRLE NLIAQLPGEKKNGLFGNLIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDNLLAQIGDQ YADLFLAAKNLSDAILLSDILRVNTEITKAPLSASMIKRYDEHHQDLTLLKALVRQQLPEKY KEIFFDQSKNGYAGYIDGGASQEEFYKFIKPILEKMDGTEELLVKLNREDLLRKQRTFDNGS IPHQIHLGELHAILRRQEDFYPFLKDNREKIEKILTFRIPYYVGPLARGNSRFAWMTRKSEE TITPWNFEEVVDKGASAQSFIERMTNFDKNLPNEKVLPKHSLLYEYFTVYNELTKVKYVTEG MRKPAFLSGEQKKAIVDLLFKTNRKVTVKQLKEDYFKKIECFDSVEISGVEDRFNASLGTYH DLLKIIKDKDFLDNEENEDILEDIVLTLTLFEDREMIEERLKTYAHLFDDKVMKQLKRRRYT GWGRLSRKLINGIRDKQSGKTILDFLKSDGFANRNFMQLIHDDSLTFKEDIQKAQVSGQGDS LHEHIANLAGSPAIKKGILQTVKVVDELVKVMGRHKPENIVIEMARENQTTQKGQKNSRERM KRIEEGIKELGSQILKEHPVENTQLQNEKLYLYYLQNGRDMYVDQELDINRLSDYDVD<mark>H</mark>IVP OSFLKDDSIDNKVLTRSDKNRGKSDNVPSEEVVKKMKNYWROLLNAKLITORKFDNLTKAER GGLSELDKAGFIKROLVETROITKHVAQILDSRMNTKYDENDKLIREVKVITLKSKLVSDFR KDFQFYKVREINNYHHAHDAYLNAVVGTALIKKYPKLESEFVYGDYKVYDVRKMIAKSEQEI GKATAKYFFYSNIMNFFKTEITLANGEIRKRPLIETNGETGEIVWDKGRDFATVRKVLSMPO VNIVKKTEVQTGGFSKESIRPKRNSDKLIARKKDWDPKKYGGFVSPTVAYSVLVVAKVEKGK SKKLKSVKELLGITIMERSSFEKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELENGRKRMLA SARFLQKGNELALPSKYVNFLYLASHYEKLKGSPEDNEQKQLFVEQHKHYLDEIIEQISEFS KRVILADANLDKVLSAYNKHRDKPIREQAENIIHLFTLTNLGAPRAFKYFDTTIDRKVYRST SDIIEKETGKOLVIOESILMLPEEVEEVIGNKPESDILVHTAYDESTDENVMLLTSDAPEYK PWALVIODSNGENKIKMLSGGSKRPAATKKAGOAKKKKRPPPSDYKDDDDKDYKDDDDK\*

#### AID10-nSaCas9-KKH-UGI

# (NLS-AID10-XTEN-nSaCas9-KKH-(GGS)<sub>6</sub> linker-UGI-NLS-2×FLAG)

MAPKKKRKVGIHGVPAADSLLMNRREFLYQFKNVRWAKGRRETYLCYVVERRDCATSFSLDF GYLRNKNGCHVELLFLRYISDWDLDPGRCYRVTWFISWSPCYDCARHVADFLRGNPNLSLRI FAARLYFCEDRKAEPEGLRRLRRAGVQIAIMTFKDYFYCWNTFAENHGRTFKAWEGLHENSV RLSGQLRRILSGSETPGTSESATPESMKRNYILGLAIGITSVGYGIIDYETRDVIDAGVRLF KEANVENNEGRRSKRGARRLKRRRRHRIORVKKLLFDYNLLTDHSELSGINPYEARVKGLSO KLSEEEFSAALLHLAKRRGVHNVNEVEEDTGNELSTKEQISRNSKALEEKYVAELQLERLKK DGEVRGSINRFKTSDYVKEAKQLLKVQKAYHQLDQSFIDTYIDLLETRRTYYEGPGEGSPFG WKDIKEWYEMLMGHCTYFPEELRSVKYAYNADLYNALNDLNNLVITRDENEKLEYYEKF0II ENVFKOKKKPTLKOIAKEILVNEEDIKGYRVTSTGKPEFTNLKVYHDIKDITARKEIIENAE LLDQIAKILTIYQSSEDIQEELTNLNSELTQEEIEQISNLKGYTGTHNLSLKAINLILDELW HTNDNQIAIFNRLKLVPKKVDLSQQKEIPTTLVDDFILSPVVKRSFIQSIKVINAIIKKYGL PNDIIIELAREKNSKDAQKMINEMQKRNRQTNERIEEIIRTTGKENAKYLIEKIKLHDMQEG KCLYSLEAIPLEDLLNNPFNYEVDHIIPRSVSFDNSFNNKVLVKQEE<mark>N</mark>SKKGNRTPFQYLSS SDSKISYETFKKHILNLAKGKGRISKTKKEYLLEERDINRFSVOKDFINRNLVDTRYATRGL MNLLRSYFRVNNLDVKVKSINGGFTSFLRRKWKFKKERNKGYKHHAEDALIIANADFIFKEW KKLDKAKKVMENQMFEEKQAESMPEIETEQEYKEIFITPHQIKHIKDFKDYKYSHRVDKKPN RKLINDTLYSTRKDDKGNTLIVNNLNGLYDKDNDKLKKLINKSPEKLLMYHHDPQTYQKLKL IMEQYGDEKNPLYKYYEETGNYLTKYSKKDNGPVIKKIKYYGNKLNAHLDITDDYPNSRNKV VKLSLKPYRFDVYLDNGVYKFVTVKNLDVIKKENYYEVNSKCYEEAKKLKKISNQAEFIASF YKNDLIKINGELYRVIGVNNDLLNRIEVNMIDITYREYLENMNDKRPPHIIKTIASKTOSIK KYSTDILGNLYEVKSKKHPQIIKKGSRADGGGGSGGGGSGGGGSGGGGSGGGGSGGGSTNL SDIIEKETGKOLVIOESILMLPEEVEEVIGNKPESDILVHTAYDESTDENVMLLTSDAPEYK PWALVIODSNGENKIKMLSGGSKRPAATKKAGOAKKKKRPPPSDYKDDDDKDYKDDDDK\*

#### nCas9-AID10-UGI

# (NLS-nCas9-NLS-XTEN-AID10-NLS-UGI-2×FLAG)

MAPKKKRKVGIHGVPAADKKYSIGLAIGTNSVGWAVITDEYKVPSKKFKVLGNTDRHSIKKN LIGALLFDSGETAEATRLKRTARRRYTRRKNRICYLOEIFSNEMAKVDDSFFHRLEESFLVE EDKKHERHPIFGNIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRLIYLALAHMIKFRGHFLI EGDLNPDNSDVDKLFIOLVOTYNOLFEENPINASGVDAKAILSARLSKSRRLENLIAOLPGE KKNGLFGNLIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDNLLAQIGDQYADLFLAAK NLSDAILLSDILRVNTEITKAPLSASMIKRYDEHHODLTLLKALVROOLPEKYKEIFFDOSK NGYAGYIDGGASQEEFYKFIKPILEKMDGTEELLVKLNREDLLRKQRTFDNGSIPHQIHLGE LHAILRROEDFYPFLKDNREKIEKILTFRIPYYVGPLARGNSRFAWMTRKSEETITPWNFEE VVDKGASAOSFIERMTNFDKNLPNEKVLPKHSLLYEYFTVYNELTKVKYVTEGMRKPAFLSG EQKKAIVDLLFKTNRKVTVKQLKEDYFKKIECFDSVEISGVEDRFNASLGTYHDLLKIIKDK DFLDNEENEDILEDIVLTLTLFEDREMIEERLKTYAHLFDDKVMKQLKRRRYTGWGRLSRKL INGIRDKQSGKTILDFLKSDGFANRNFMQLIHDDSLTFKEDIQKAQVSGQGDSLHEHIANLA GSPAIKKGILOTVKVVDELVKVMGRHKPENIVIEMARENOTTOKGOKNSRERMKRIEEGIKE LGSQILKEHPVENTQLQNEKLYLYLQNGRDMYVDQELDINRLSDYDVDHIVPQSFLKDDSI DNKVLTRSDKNRGKSDNVPSEEVVKKMKNYWRQLLNAKLITQRKFDNLTKAERGGLSELDKA GFIKRQLVETRQITKHVAQILDSRMNTKYDENDKLIREVKVITLKSKLVSDFRKDFQFYKVR EINNYHHAHDAYLNAVVGTALIKKYPKLESEFVYGDYKVYDVRKMIAKSEQEIGKATAKYFF YSNIMNFFKTEITLANGEIRKRPLIETNGETGEIVWDKGRDFATVRKVLSMPOVNIVKKTEV QTGGFSKESILPKRNSDKLIARKKDWDPKKYGGFDSPTVAYSVLVVAKVEKGKSKKLKSVKE LLGITIMERSSFEKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELENGRKRMLASAGELOKGN ELALPSKYVNFLYLASHYEKLKGSPEDNEQKQLFVEQHKHYLDEIIEQISEFSKRVILADAN LDKVLSAYNKHRDKPIREQAENIIHLFTLTNLGAPAAFKYFDTTIDRKRYTSTKEVLDATLI HQSITGLYETRIDLSQLGGDKRPAATKKAGQAKKKKRPSPSGSETPGTSESATPESDSLLMN RREFLYQFKNVRWAKGRRETYLCYVVERRDCATSFSLDFGYLRNKNGCHVELLFLRYISDWD LDPGRCYRVTWFISWSPCYDCARHVADFLRGNPNLSLRIFAARLYFCEDRKAEPEGLRRLRR AGVQIAIMTFKDYFYCWNTFAENHGRTFKAWEGLHENSVRLSGQLRRILGPKKKRKVGSTNL SDIIEKETGKOLVIOESILMLPEEVEEVIGNKPESDILVHTAYDESTDENVMLLTSDAPEYK PWALVIQDSNGENKIKMLRPPPSDYKDDDDKDYKDDDDK\*

# AID10-nCas9-AID10-UGI

#### (NLS-AID10-XTEN-nCas9-NLS-XTEN-AID10-NLS-UGI-2×FLAG)

MAPKKKRKVGIHGVPAADSLLMNRREFLYQFKNVRWAKGRRETYLCYVVERRDCATSFSLDF GYLRNKNGCHVELLFLRYISDWDLDPGRCYRVTWFISWSPCYDCARHVADFLRGNPNLSLRI FAARLYFCEDRKAEPEGLRRLRRAGVQIAIMTFKDYFYCWNTFAENHGRTFKAWEGLHENSV RLSGQLRRILSGSETPGTSESATPESDKKYSIGL<mark>A</mark>IGTNSVGWAVITDEYKVPSKKFKVLGN TDRHSIKKNLIGALLFDSGETAEATRLKRTARRRYTRRKNRICYLQEIFSNEMAKVDDSFFH RLEESFLVEEDKKHERHPIFGNIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRLIYLALAHM IKFRGHFLIEGDLNPDNSDVDKLFIQLVQTYNQLFEENPINASGVDAKAILSARLSKSRRLE NLIAQLPGEKKNGLFGNLIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDNLLAQIGDQ YADLFLAAKNLSDAILLSDILRVNTEITKAPLSASMIKRYDEHHQDLTLLKALVRQQLPEKY KEIFFDQSKNGYAGYIDGGASQEEFYKFIKPILEKMDGTEELLVKLNREDLLRKQRTFDNGS

IPHQIHLGELHAILRRQEDFYPFLKDNREKIEKILTFRIPYYVGPLARGNSRFAWMTRKSEE TITPWNFEEVVDKGASAQSFIERMTNFDKNLPNEKVLPKHSLLYEYFTVYNELTKVKYVTEG MRKPAFLSGEQKKAIVDLLFKTNRKVTVKQLKEDYFKKIECFDSVEISGVEDRFNASLGTYH DLLKIIKDKDFLDNEENEDILEDIVLTLTLFEDREMIEERLKTYAHLFDDKVMKQLKRRRYT GWGRLSRKLINGIRDKQSGKTILDFLKSDGFANRNFMQLIHDDSLTFKEDIQKAQVSGQGDS LHEHIANLAGSPAIKKGILOTVKVVDELVKVMGRHKPENIVIEMARENOTTOKGOKNSRERM KRIEEGIKELGSQILKEHPVENTQLQNEKLYLYYLQNGRDMYVDQELDINRLSDYDVD<mark>H</mark>IVP QSFLKDDSIDNKVLTRSDKNRGKSDNVPSEEVVKKMKNYWRQLLNAKLITQRKFDNLTKAER GGLSELDKAGFIKROLVETROITKHVAQILDSRMNTKYDENDKLIREVKVITLKSKLVSDFR KDFQFYKVREINNYHHAHDAYLNAVVGTALIKKYPKLESEFVYGDYKVYDVRKMIAKSEQEI GKATAKYFFYSNIMNFFKTEITLANGEIRKRPLIETNGETGEIVWDKGRDFATVRKVLSMPQ VNIVKKTEVQTGGFSKESILPKRNSDKLIARKKDWDPKKYGGFDSPTVAYSVLVVAKVEKGK SKKLKSVKELLGITIMERSSFEKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELENGRKRMLA SAGELQKGNELALPSKYVNFLYLASHYEKLKGSPEDNEQKQLFVEQHKHYLDEIIEQISEFS KRVILADANLDKVLSAYNKHRDKPIREQAENIIHLFTLTNLGAPAAFKYFDTTIDRKRYTST KEVLDATLIHOSITGLYETRIDLSOLGGDKRPAATKKAGOAKKKKRPSPSGSETPGTSESAT **PESDSLLMNRREFLYQFKNVRWAKGRRETYLCYVVERRDCATSFSLDFGYLRNKNGCHVELL** FLRYISDWDLDPGRCYRVTWFISWSPCYDCARHVADFLRGNPNLSLRIFAARLYFCEDRKAE PEGLRRLRRAGVQIAIMTFKDYFYCWNTFAENHGRTFKAWEGLHENSVRLSGQLRRILGPKK KRKVGSTNLSDIIEKETGKQLVIQESILMLPEEVEEVIGNKPESDILVHTAYDESTDENVML LTSDAPEYKPWALVIQDSNGENKIKMLRPPPSDYKDDDDKDYKDDDDK\*

# ABE7.10-nCas9

# (TadA-32aa linker-TadA7.10-32aa linker-nCas9-NLS-NLS-2×FLAG)

MGSEVEFSHEYWMRHALTLAKRAWDEREVPVGAVLVHNNRVIGEGWNRPIGRHDPTAHAEIM ALROGGLVMONYRLIDATLYVTLEPCVMCAGAMIHSRIGRVVFGARDAKTGAAGSLMDVLHH PGMNHRVEITEGILADECAALLSDFFRMRROEIKAOKKAOSSTDSGGSSGGSSGSETPGTSE SATPESSGGSSGGSSEVEFSHEYWMRHALTLAKRARDEREVPVGAVLVLNNRVIGEGWNRAI GLHDPTAHAEIMALRQGGLVMQNYRLIDATLYVTFEPCVMCAGAMIHSRIGRVVFGVRNAKT GAAGSLMDVLHYPGMNHRVEITEGILADECAALLCYFFRMPRQVFNAQKKAQSSTDSGGSSG GSSGSETPGTSESATPESSGGSSGGSDKKYSIGLAIGTNSVGWAVITDEYKVPSKKFKVLGN TDRHSIKKNLIGALLFDSGETAEATRLKRTARRRYTRRKNRICYLQEIFSNEMAKVDDSFFH RLEESFLVEEDKKHERHPIFGNIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRLIYLALAHM IKFRGHFLIEGDLNPDNSDVDKLFIOLVOTYNOLFEENPINASGVDAKAILSARLSKSRRLE NLIAQLPGEKKNGLFGNLIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDNLLAQIGDQ YADLFLAAKNLSDAILLSDILRVNTEITKAPLSASMIKRYDEHHQDLTLLKALVRQQLPEKY KEIFFDQSKNGYAGYIDGGASQEEFYKFIKPILEKMDGTEELLVKLNREDLLRKQRTFDNGS IPHQIHLGELHAILRRQEDFYPFLKDNREKIEKILTFRIPYYVGPLARGNSRFAWMTRKSEE TITPWNFEEVVDKGASAOSFIERMTNFDKNLPNEKVLPKHSLLYEYFTVYNELTKVKYVTEG MRKPAFLSGEQKKAIVDLLFKTNRKVTVKQLKEDYFKKIECFDSVEISGVEDRFNASLGTYH DLLKIIKDKDFLDNEENEDILEDIVLTLTLFEDREMIEERLKTYAHLFDDKVMKQLKRRRYT GWGRLSRKLINGIRDKQSGKTILDFLKSDGFANRNFMQLIHDDSLTFKEDIQKAQVSGQGDS LHEHIANLAGSPAIKKGILQTVKVVDELVKVMGRHKPENIVIEMARENQTTQKGQKNSRERM KRIEEGIKELGSQILKEHPVENTQLQNEKLYLYYLQNGRDMYVDQELDINRLSDYDVDHIVP QSFLKDDSIDNKVLTRSDKNRGKSDNVPSEEVVKKMKNYWRQLLNAKLITQRKFDNLTKAER GGLSELDKAGFIKRQLVETRQITKHVAQILDSRMNTKYDENDKLIREVKVITLKSKLVSDFR KDFQFYKVREINNYHHAHDAYLNAVVGTALIKKYPKLESEFVYGDYKVYDVRKMIAKSEQEI GKATAKYFFYSNIMNFFKTEITLANGEIRKRPLIETNGETGEIVWDKGRDFATVRKVLSMPQ VNIVKKTEVQTGGFSKESILPKRNSDKLIARKKDWDPKKYGGFDSPTVAYSVLVVAKVEKGK SKKLKSVKELLGITIMERSSFEKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELENGRKRMLA SAGELQKGNELALPSKYVNFLYLASHYEKLKGSPEDNEQKQLFVEQHKHYLDEIIEQISEFS KRVILADANLDKVLSAYNKHRDKPIREQAENIIHLFTLTNLGAPAAFKYFDTTIDRKRYTST KEVLDATLIHQSITGLYETRIDLSQLGGDKRPAATKKAGQAKKKKSGGSPKKKRKVRPPPSD YKDDDDKDYKDDDDK\*

#### nCas9-AID10

# (NLS-nCas9-NLS-XTEN-AID10-2×FLAG)

MAPKKKRKVGIHGVPAADKKYSIGL<mark>A</mark>IGTNSVGWAVITDEYKVPSKKFKVLGNTDRHSIKKN LIGALLFDSGETAEATRLKRTARRRYTRRKNRICYLQEIFSNEMAKVDDSFFHRLEESFLVE EDKKHERHPIFGNIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRLIYLALAHMIKFRGHFLI EGDLNPDNSDVDKLFIQLVQTYNQLFEENPINASGVDAKAILSARLSKSRRLENLIAQLPGE KKNGLFGNLIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDNLLAQIGDQYADLFLAAK NLSDAILLSDILRVNTEITKAPLSASMIKRYDEHHODLTLLKALVROOLPEKYKEIFFDOSK NGYAGYIDGGASQEEFYKFIKPILEKMDGTEELLVKLNREDLLRKQRTFDNGSIPHQIHLGE LHAILRRQEDFYPFLKDNREKIEKILTFRIPYYVGPLARGNSRFAWMTRKSEETITPWNFEE VVDKGASAQSFIERMTNFDKNLPNEKVLPKHSLLYEYFTVYNELTKVKYVTEGMRKPAFLSG EOKKAIVDLLFKTNRKVTVKOLKEDYFKKIECFDSVEISGVEDRFNASLGTYHDLLKIIKDK DFLDNEENEDILEDIVLTLTLFEDREMIEERLKTYAHLFDDKVMKQLKRRRYTGWGRLSRKL INGIRDKQSGKTILDFLKSDGFANRNFMQLIHDDSLTFKEDIQKAQVSGQGDSLHEHIANLA GSPAIKKGILOTVKVVDELVKVMGRHKPENIVIEMARENOTTOKGOKNSRERMKRIEEGIKE LGSQILKEHPVENTQLQNEKLYLYLQNGRDMYVDQELDINRLSDYDVDHIVPQSFLKDDSI DNKVLTRSDKNRGKSDNVPSEEVVKKMKNYWRQLLNAKLITQRKFDNLTKAERGGLSELDKA GFIKRQLVETRQITKHVAQILDSRMNTKYDENDKLIREVKVITLKSKLVSDFRKDFQFYKVR EINNYHHAHDAYLNAVVGTALIKKYPKLESEFVYGDYKVYDVRKMIAKSEQEIGKATAKYFF YSNIMNFFKTEITLANGEIRKRPLIETNGETGEIVWDKGRDFATVRKVLSMPQVNIVKKTEV OTGGFSKESILPKRNSDKLIARKKDWDPKKYGGFDSPTVAYSVLVVAKVEKGKSKKLKSVKE LLGITIMERSSFEKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELENGRKRMLASAGELOKGN ELALPSKYVNFLYLASHYEKLKGSPEDNEOKOLFVEOHKHYLDEIIEQISEFSKRVILADAN LDKVLSAYNKHRDKPIREQAENIIHLFTLTNLGAPAAFKYFDTTIDRKRYTSTKEVLDATLI HQSITGLYETRIDLSQLGGDKRPAATKKAGQAKKKKRPSGSETPGTSESATPESDSLLMNRR EFLYQFKNVRWAKGRRETYLCYVVERRDCATSFSLDFGYLRNKNGCHVELLFLRYISDWDLD PGRCYRVTWFISWSPCYDCARHVADFLRGNPNLSLRIFAARLYFCEDRKAEPEGLRRLRRAG VQIAIMTFKDYFYCWNTFAENHGRTFKAWEGLHENSVRLSGQLRRILRPPPSDYKDDDDKDY KDDDDK\*

#### ABE7.10-nCas9-AID10

(TadA-32aa linker-TadA7.10-32aa linker-nCas9-NLS-NLS-XTEN-AID10-2×FLAG)

MGSEVEFSHEYWMRHALTLAKRAWDEREVPVGAVLVHNNRVIGEGWNRPIGRHDPTAHAEIM ALROGGLVMONYRLIDATLYVTLEPCVMCAGAMIHSRIGRVVFGARDAKTGAAGSLMDVLHH PGMNHRVEITEGILADECAALLSDFFRMRRQEIKAQKKAQSSTDSGGSSGGSSGSETPGTSE **SATPESSGGSSGGSSEVEFSHEYWMRHALTLAKRARDEREVPVGAVLVLNNRVIGEGWNRAI** GLHDPTAHAEIMALRQGGLVMQNYRLIDATLYVTFEPCVMCAGAMIHSRIGRVVFGVRNAKT GAAGSLMDVLHYPGMNHRVEITEGILADECAALLCYFFRMPRQVFNAQKKAQSSTDSGGSSG GSSGSETPGTSESATPESSGGSSGGSDKKYSIGLAIGTNSVGWAVITDEYKVPSKKFKVLGN TDRHSIKKNLIGALLFDSGETAEATRLKRTARRRYTRRKNRICYLOEIFSNEMAKVDDSFFH RLEESFLVEEDKKHERHPIFGNIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRLIYLALAHM IKFRGHFLIEGDLNPDNSDVDKLFIQLVQTYNQLFEENPINASGVDAKAILSARLSKSRRLE NLIAQLPGEKKNGLFGNLIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDNLLAQIGDQ YADLFLAAKNLSDAILLSDILRVNTEITKAPLSASMIKRYDEHHQDLTLLKALVRQQLPEKY KEIFFDOSKNGYAGYIDGGASOEEFYKFIKPILEKMDGTEELLVKLNREDLLRKORTFDNGS IPHQIHLGELHAILRRQEDFYPFLKDNREKIEKILTFRIPYYVGPLARGNSRFAWMTRKSEE TITPWNFEEVVDKGASAQSFIERMTNFDKNLPNEKVLPKHSLLYEYFTVYNELTKVKYVTEG MRKPAFLSGEQKKAIVDLLFKTNRKVTVKQLKEDYFKKIECFDSVEISGVEDRFNASLGTYH DLLKIIKDKDFLDNEENEDILEDIVLTLTLFEDREMIEERLKTYAHLFDDKVMKQLKRRRYT GWGRLSRKLINGIRDKOSGKTILDFLKSDGFANRNFMOLIHDDSLTFKEDIOKAOVSGOGDS LHEHIANLAGSPAIKKGILQTVKVVDELVKVMGRHKPENIVIEMARENQTTQKGQKNSRERM KRIEEGIKELGSQILKEHPVENTQLQNEKLYLYYLQNGRDMYVDQELDINRLSDYDVD<mark>H</mark>IVP QSFLKDDSIDNKVLTRSDKNRGKSDNVPSEEVVKKMKNYWRQLLNAKLITQRKFDNLTKAER GGLSELDKAGFIKROLVETROITKHVAOILDSRMNTKYDENDKLIREVKVITLKSKLVSDFR KDFQFYKVREINNYHHAHDAYLNAVVGTALIKKYPKLESEFVYGDYKVYDVRKMIAKSEQEI GKATAKYFFYSNIMNFFKTEITLANGEIRKRPLIETNGETGEIVWDKGRDFATVRKVLSMPQ VNIVKKTEVOTGGFSKESILPKRNSDKLIARKKDWDPKKYGGFDSPTVAYSVLVVAKVEKGK SKKLKSVKELLGITIMERSSFEKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELENGRKRMLA SAGELQKGNELALPSKYVNFLYLASHYEKLKGSPEDNEQKQLFVEQHKHYLDEIIEQISEFS KRVILADANLDKVLSAYNKHRDKPIREQAENIIHLFTLTNLGAPAAFKYFDTTIDRKRYTST KEVLDATLIHOSITGLYETRIDLSOLGGDKRPAATKKAGOAKKKKSGGSPKKKRKVRPSGSE **TPGTSESATPESDSLLMNRREFLYOFKNVRWAKGRRETYLCYVVERRDCATSFSLDFGYLRN** KNGCHVELLFLRYISDWDLDPGRCYRVTWFISWSPCYDCARHVADFLRGNPNLSLRIFAARL YFCEDRKAEPEGLRRLRRAGVQIAIMTFKDYFYCWNTFAENHGRTFKAWEGLHENSVRLSGQ LRRILRPPPSDYKDDDDKDYKDDDDK\*

# TadA7.10-nCas9-AID10

(TadA7.10–32aa linker–nCas9–NLS–NLS–XTEN–AID10–2×FLAG)

MGSEVEFSHEYWMRHALTLAKRARDEREVPVGAVLVLNNRVIGEGWNRAIGLHDPTAHAEIM ALRQGGLVMQNYRLIDATLYVTFEPCVMCAGAMIHSRIGRVVFGVRNAKTGAAGSLMDVLHY PGMNHRVEITEGILADECAALLCYFFRMPRQVFNAQKKAQSSTDSGGSSGGSSGSETPGTSE SATPESSGGSSGGSDKKYSIGL<mark>A</mark>IGTNSVGWAVITDEYKVPSKKFKVLGNTDRHSIKKNLIG ALLFDSGETAEATRLKRTARRRYTRRKNRICYLQEIFSNEMAKVDDSFFHRLEESFLVEEDK KHERHPIFGNIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRLIYLALAHMIKFRGHFLIEGD LNPDNSDVDKLFIQLVQTYNQLFEENPINASGVDAKAILSARLSKSRRLENLIAQLPGEKKN GLFGNLIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDNLLAQIGDQYADLFLAAKNLS DAILLSDILRVNTEITKAPLSASMIKRYDEHHQDLTLLKALVRQQLPEKYKEIFFDQSKNGY AGYIDGGASQEEFYKFIKPILEKMDGTEELLVKLNREDLLRKQRTFDNGSIPHQIHLGELHA ILRROEDFYPFLKDNREKIEKILTFRIPYYVGPLARGNSRFAWMTRKSEETITPWNFEEVVD KGASAQSFIERMTNFDKNLPNEKVLPKHSLLYEYFTVYNELTKVKYVTEGMRKPAFLSGEQK KAIVDLLFKTNRKVTVKQLKEDYFKKIECFDSVEISGVEDRFNASLGTYHDLLKIIKDKDFL DNEENEDILEDIVLTLTLFEDREMIEERLKTYAHLFDDKVMKQLKRRRYTGWGRLSRKLING IRDKQSGKTILDFLKSDGFANRNFMQLIHDDSLTFKEDIQKAQVSGQGDSLHEHIANLAGSP AIKKGILQTVKVVDELVKVMGRHKPENIVIEMARENQTTQKGQKNSRERMKRIEEGIKELGS QILKEHPVENTQLQNEKLYLYLQNGRDMYVDQELDINRLSDYDVDHIVPQSFLKDDSIDNK VLTRSDKNRGKSDNVPSEEVVKKMKNYWRQLLNAKLITQRKFDNLTKAERGGLSELDKAGFI KRQLVETRQITKHVAQILDSRMNTKYDENDKLIREVKVITLKSKLVSDFRKDFQFYKVREIN NYHHAHDAYLNAVVGTALIKKYPKLESEFVYGDYKVYDVRKMIAKSEOEIGKATAKYFFYSN IMNFFKTEITLANGEIRKRPLIETNGETGEIVWDKGRDFATVRKVLSMPOVNIVKKTEVOTG GFSKESILPKRNSDKLIARKKDWDPKKYGGFDSPTVAYSVLVVAKVEKGKSKKLKSVKELLG ITIMERSSFEKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELENGRKRMLASAGELQKGNELA LPSKYVNFLYLASHYEKLKGSPEDNEQKQLFVEQHKHYLDEIIEQISEFSKRVILADANLDK VLSAYNKHRDKPIREQAENIIHLFTLTNLGAPAAFKYFDTTIDRKRYTSTKEVLDATLIHQS ITGLYETRIDLSQLGGDKRPAATKKAGQAKKKKSGGSPKKKRKVRPSGSETPGTSESATPES DSLLMNRREFLYOFKNVRWAKGRRETYLCYVVERRDCATSFSLDFGYLRNKNGCHVELLFLR YISDWDLDPGRCYRVTWFISWSPCYDCARHVADFLRGNPNLSLRIFAARLYFCEDRKAEPEG LRRLRRAGVQIAIMTFKDYFYCWNTFAENHGRTFKAWEGLHENSVRLSGQLRRILRPPPSDY KDDDDKDYKDDDDK\*

#### AID10-ABE7.10-nCas9

(AID10–XTEN–TadA–32aa linker–TadA7.10–32aa linker–nCas9–NLS–NLS–2×FLAG)

MDSLLMNRREFLYQFKNVRWAKGRRETYLCYVVERRDCATSFSLDFGYLRNKNGCHVELLFL RYISDWDLDPGRCYRVTWFISWSPCYDCARHVADFLRGNPNLSLRIFAARLYFCEDRKAEPE GLRRLRRAGVQIAIMTFKDYFYCWNTFAENHGRTFKAWEGLHENSVRLSGQLRRILSGSETP GTSESATPESSEVEFSHEYWMRHALTLAKRAWDEREVPVGAVLVHNNRVIGEGWNRPIGRHD PTAHAEIMALRQGGLVMQNYRLIDATLYVTLEPCVMCAGAMIHSRIGRVVFGARDAKTGAAG SLMDVLHHPGMNHRVEITEGILADECAALLSDFFRMRRQEIKAQKKAQSSTDSGGSSGGSSG SETPGTSESATPESSGGSSGGSSEVEFSHEYWMRHALTLAKRARDEREVPVGAVLVLNNRVI GEGWNRAIGLHDPTAHAEIMALRQGGLVMQNYRLIDATLYVTFEPCVMCAGAMIHSRIGRVV FGVRNAKTGAAGSLMDVLHYPGMNHRVEITEGILADECAALLCYFFRMPRQVFNAQKKAQSS TDSGGSSGGSSGSSETPGTSESATPESSGGSSGGSDKKYSIGL<mark>A</mark>IGTNSVGWAVITDEYKVPS KKFKVLGNTDRHSIKKNLIGALLFDSGETAEATRLKRTARRRYTRRKNRICYLQEIFSNEMA KVDDSFFHRLEESFLVEEDKKHERHPIFGNIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRL IYLALAHMIKFRGHFLIEGDLNPDNSDVDKLFIQLVQTYNQLFEENPINASGVDAKAILSAR LSKSRRLENLIAQLPGEKKNGLFGNLIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDN LLAQIGDQYADLFLAAKNLSDAILLSDILRVNTEITKAPLSASMIKRYDEHHQDLTLLKALV RQQLPEKYKEIFFDQSKNGYAGYIDGGASQEEFYKFIKPILEKMDGTEELLVKLNREDLLRK QRTFDNGSIPHQIHLGELHAILRRQEDFYPFLKDNREKIEKILTFRIPYYVGPLARGNSRFA WMTRKSEETITPWNFEEVVDKGASAQSFIERMTNFDKNLPNEKVLPKHSLLYEYFTVYNELT KVKYVTEGMRKPAFLSGEQKKAIVDLLFKTNRKVTVKQLKEDYFKKIECFDSVEISGVEDRF NASLGTYHDLLKIIKDKDFLDNEENEDILEDIVLTLTLFEDREMIEERLKTYAHLFDDKVMK QLKRRRYTGWGRLSRKLINGIRDKQSGKTILDFLKSDGFANRNFMQLIHDDSLTFKEDIQKA OVSGOGDSLHEHIANLAGSPAIKKGILOTVKVVDELVKVMGRHKPENIVIEMARENOTTOKG QKNSRERMKRIEEGIKELGSQILKEHPVENTQLQNEKLYLYYLQNGRDMYVDQELDINRLSD YDVD<mark>H</mark>IVPQSFLKDDSIDNKVLTRSDKNRGKSDNVPSEEVVKKMKNYWRQLLNAKLITQRKF DNLTKAERGGLSELDKAGFIKRQLVETRQITKHVAQILDSRMNTKYDENDKLIREVKVITLK SKLVSDFRKDFQFYKVREINNYHHAHDAYLNAVVGTALIKKYPKLESEFVYGDYKVYDVRKM IAKSEQEIGKATAKYFFYSNIMNFFKTEITLANGEIRKRPLIETNGETGEIVWDKGRDFATV RKVLSMPQVNIVKKTEVQTGGFSKESILPKRNSDKLIARKKDWDPKKYGGFDSPTVAYSVLV VAKVEKGKSKKLKSVKELLGITIMERSSFEKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELE NGRKRMLASAGELOKGNELALPSKYVNFLYLASHYEKLKGSPEDNEOKOLFVEOHKHYLDEI IEQISEFSKRVILADANLDKVLSAYNKHRDKPIREOAENIIHLFTLTNLGAPAAFKYFDTTI DRKRYTSTKEVLDATLIHQSITGLYETRIDLSQLGGDKRPAATKKAGQAKKKKSGGSPKKKR KVRPPPSDYKDDDDKDYKDDDDK\*

# AID10-TadA7.10-nCas9

# (AID10-XTEN-TadA7.10-32aa linker-nCas9-NLS-NLS-2×FLAG)

MDSLLMNRREFLYQFKNVRWAKGRRETYLCYVVERRDCATSFSLDFGYLRNKNGCHVELLFL RYISDWDLDPGRCYRVTWFISWSPCYDCARHVADFLRGNPNLSLRIFAARLYFCEDRKAEPE GLRRLRRAGVOIAIMTFKDYFYCWNTFAENHGRTFKAWEGLHENSVRLSGOLRRILSGSETP GTSESATPESSEVEFSHEYWMRHALTLAKRARDEREVPVGAVLVLNNRVIGEGWNRAIGLHD PTAHAEIMALROGGLVMONYRLIDATLYVTFEPCVMCAGAMIHSRIGRVVFGVRNAKTGAAG SLMDVLHYPGMNHRVEITEGILADECAALLCYFFRMPROVFNAOKKAOSSTDSGGSSGGSSG SETPGTSESATPESSGGSSGGSDKKYSIGLAIGTNSVGWAVITDEYKVPSKKFKVLGNTDRH SIKKNLIGALLFDSGETAEATRLKRTARRRYTRRKNRICYLQEIFSNEMAKVDDSFFHRLEE SFLVEEDKKHERHPIFGNIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRLIYLALAHMIKFR GHFLIEGDLNPDNSDVDKLFIQLVQTYNQLFEENPINASGVDAKAILSARLSKSRRLENLIA **QLPGEKKNGLFGNLIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDNLLAQIGDQYADL** FLAAKNLSDAILLSDILRVNTEITKAPLSASMIKRYDEHHQDLTLLKALVRQQLPEKYKEIF FDOSKNGYAGYIDGGASOEEFYKFIKPILEKMDGTEELLVKLNREDLLRKORTFDNGSIPHO IHLGELHAILRRQEDFYPFLKDNREKIEKILTFRIPYYVGPLARGNSRFAWMTRKSEETITP WNFEEVVDKGASAQSFIERMTNFDKNLPNEKVLPKHSLLYEYFTVYNELTKVKYVTEGMRKP AFLSGEQKKAIVDLLFKTNRKVTVKQLKEDYFKKIECFDSVEISGVEDRFNASLGTYHDLLK IIKDKDFLDNEENEDILEDIVLTLTLFEDREMIEERLKTYAHLFDDKVMKQLKRRRYTGWGR LSRKLINGIRDKOSGKTILDFLKSDGFANRNFMOLIHDDSLTFKEDIOKAOVSGOGDSLHEH IANLAGSPAIKKGILQTVKVVDELVKVMGRHKPENIVIEMARENQTTQKGQKNSRERMKRIE EGIKELGSQILKEHPVENTQLQNEKLYLYYLQNGRDMYVDQELDINRLSDYDVDHIVPQSFL KDDSIDNKVLTRSDKNRGKSDNVPSEEVVKKMKNYWRQLLNAKLITQRKFDNLTKAERGGLS ELDKAGFIKRQLVETRQITKHVAQILDSRMNTKYDENDKLIREVKVITLKSKLVSDFRKDFQ

FYKVREINNYHHAHDAYLNAVVGTALIKKYPKLESEFVYGDYKVYDVRKMIAKSEQEIGKAT AKYFFYSNIMNFFKTEITLANGEIRKRPLIETNGETGEIVWDKGRDFATVRKVLSMPQVNIV KKTEVQTGGFSKESILPKRNSDKLIARKKDWDPKKYGGFDSPTVAYSVLVVAKVEKGKSKKL KSVKELLGITIMERSSFEKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELENGRKRMLASAGE LQKGNELALPSKYVNFLYLASHYEKLKGSPEDNEQKQLFVEQHKHYLDEIIEQISEFSKRVI LADANLDKVLSAYNKHRDKPIREQAENIIHLFTLTNLGAPAAFKYFDTTIDRKRYTSTKEVL DATLIHQSITGLYETRIDLSQLGGDKRPAATKKAGQAKKKKSGGSPKKKRKVRPPPSDYKDD DDKDYKDDDDK\*

# ABE7.10-nCas9-AID10-UGI

(TadA-32aa linker-TadA7.10-32aa linker-nCas9-NLS-NLS-XTEN-AID10-(GGS)<sub>6</sub> linker -UGI-2×FLAG)

MGSEVEFSHEYWMRHALTLAKRAWDEREVPVGAVLVHNNRVIGEGWNRPIGRHDPTAHAEIM ALROGGLVMONYRLIDATLYVTLEPCVMCAGAMIHSRIGRVVFGARDAKTGAAGSLMDVLHH PGMNHRVEITEGILADECAALLSDFFRMRRQEIKAQKKAQSSTDSGGSSGGSSGSETPGTSE SATPESSGGSSGGSSEVEFSHEYWMRHALTLAKRARDEREVPVGAVLVLNNRVIGEGWNRAI GLHDPTAHAEIMALRQGGLVMQNYRLIDATLYVTFEPCVMCAGAMIHSRIGRVVFGVRNAKT GAAGSLMDVLHYPGMNHRVEITEGILADECAALLCYFFRMPROVFNAOKKAOSSTDSGGSSG GSSGSETPGTSESATPESSGGSSGGSDKKYSIGLAIGTNSVGWAVITDEYKVPSKKFKVLGN TDRHSIKKNLIGALLFDSGETAEATRLKRTARRRYTRRKNRICYLQEIFSNEMAKVDDSFFH RLEESFLVEEDKKHERHPIFGNIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRLIYLALAHM IKFRGHFLIEGDLNPDNSDVDKLFIQLVQTYNQLFEENPINASGVDAKAILSARLSKSRRLE NLIAQLPGEKKNGLFGNLIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDNLLAQIGDQ YADLFLAAKNLSDAILLSDILRVNTEITKAPLSASMIKRYDEHHQDLTLLKALVRQQLPEKY KEIFFDQSKNGYAGYIDGGASQEEFYKFIKPILEKMDGTEELLVKLNREDLLRKQRTFDNGS IPHQIHLGELHAILRRQEDFYPFLKDNREKIEKILTFRIPYYVGPLARGNSRFAWMTRKSEE TITPWNFEEVVDKGASAOSFIERMTNFDKNLPNEKVLPKHSLLYEYFTVYNELTKVKYVTEG MRKPAFLSGEOKKAIVDLLFKTNRKVTVKOLKEDYFKKIECFDSVEISGVEDRFNASLGTYH DLLKIIKDKDFLDNEENEDILEDIVLTLTLFEDREMIEERLKTYAHLFDDKVMKQLKRRRYT GWGRLSRKLINGIRDKØSGKTILDFLKSDGFANRNFMØLIHDDSLTFKEDIØKAØVSGØGDS LHEHIANLAGSPAIKKGILQTVKVVDELVKVMGRHKPENIVIEMARENQTTQKGQKNSRERM KRIEEGIKELGSQILKEHPVENTQLQNEKLYLYYLQNGRDMYVDQELDINRLSDYDVD<mark>H</mark>IVP QSFLKDDSIDNKVLTRSDKNRGKSDNVPSEEVVKKMKNYWRQLLNAKLITQRKFDNLTKAER GGLSELDKAGFIKROLVETROITKHVAOILDSRMNTKYDENDKLIREVKVITLKSKLVSDFR KDFQFYKVREINNYHHAHDAYLNAVVGTALIKKYPKLESEFVYGDYKVYDVRKMIAKSEQEI GKATAKYFFYSNIMNFFKTEITLANGEIRKRPLIETNGETGEIVWDKGRDFATVRKVLSMPQ VNIVKKTEVQTGGFSKESILPKRNSDKLIARKKDWDPKKYGGFDSPTVAYSVLVVAKVEKGK SKKLKSVKELLGITIMERSSFEKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELENGRKRMLA SAGELOKGNELALPSKYVNFLYLASHYEKLKGSPEDNEOKOLFVEOHKHYLDEIIEOISEFS KRVILADANLDKVLSAYNKHRDKPIREQAENIIHLFTLTNLGAPAAFKYFDTTIDRKRYTST KEVLDATLIHQSITGLYETRIDLSQLGGDKRPAATKKAGQAKKKKSGGSPKKKRKVRPSGSE **TPGTSESATPESDSLLMNRREFLYQFKNVRWAKGRRETYLCYVVERRDCATSFSLDFGYLRN** KNGCHVELLFLRYISDWDLDPGRCYRVTWFISWSPCYDCARHVADFLRGNPNLSLRIFAARL 

### ABE8e-nCas9-AID10-UGI

(TadA-32aa linker-TadA8e-32aa linker-nCas9-NLS-NLS-XTEN-AID10-(GGS)<sub>6</sub> linker -UGI-2×FLAG)

MGSEVEFSHEYWMRHALTLAKRAWDEREVPVGAVLVHNNRVIGEGWNRPIGRHDPTAHAEIM ALROGGLVMONYRLIDATLYVTLEPCVMCAGAMIHSRIGRVVFGARDAKTGAAGSLMDVLHH PGMNHRVEITEGILADECAALLSDFFRMRRQEIKAQKKAQSSTDSGGSSGGSSGSETPGTSE SATPESSGGSSGGSSEVEFSHEYWMRHALTLAKRARDEREVPVGAVLVLNNRVIGEGWNRAI GLHDPTAHAEIMALROGGLVMONYRLIDATLYVTFEPCVMCAGAMIHSRIGRVVFGVRNSKR GAAGSLMNVLNYPGMNHRVEITEGILADECAALLCDFYRMPROVFNAOKKAOSSINSGGSSG GSSGSETPGTSESATPESSGGSSGGSDKKYSIGLAIGTNSVGWAVITDEYKVPSKKFKVLGN TDRHSIKKNLIGALLFDSGETAEATRLKRTARRRYTRRKNRICYLQEIFSNEMAKVDDSFFH RLEESFLVEEDKKHERHPIFGNIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRLIYLALAHM IKFRGHFLIEGDLNPDNSDVDKLFIQLVQTYNQLFEENPINASGVDAKAILSARLSKSRRLE NLIAQLPGEKKNGLFGNLIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDNLLAQIGDQ YADLFLAAKNLSDAILLSDILRVNTEITKAPLSASMIKRYDEHHQDLTLLKALVRQQLPEKY KEIFFDQSKNGYAGYIDGGASQEEFYKFIKPILEKMDGTEELLVKLNREDLLRKQRTFDNGS IPHQIHLGELHAILRRQEDFYPFLKDNREKIEKILTFRIPYYVGPLARGNSRFAWMTRKSEE TITPWNFEEVVDKGASAQSFIERMTNFDKNLPNEKVLPKHSLLYEYFTVYNELTKVKYVTEG MRKPAFLSGEOKKAIVDLLFKTNRKVTVKOLKEDYFKKIECFDSVEISGVEDRFNASLGTYH DLLKIIKDKDFLDNEENEDILEDIVLTLTLFEDREMIEERLKTYAHLFDDKVMKQLKRRRYT GWGRLSRKLINGIRDKØSGKTILDFLKSDGFANRNFMØLIHDDSLTFKEDIØKAØVSGØGDS LHEHIANLAGSPAIKKGILQTVKVVDELVKVMGRHKPENIVIEMARENQTTQKGQKNSRERM KRIEEGIKELGSQILKEHPVENTQLQNEKLYLYYLQNGRDMYVDQELDINRLSDYDVD<mark>H</mark>IVP QSFLKDDSIDNKVLTRSDKNRGKSDNVPSEEVVKKMKNYWRQLLNAKLITQRKFDNLTKAER GGLSELDKAGFIKROLVETROITKHVAQILDSRMNTKYDENDKLIREVKVITLKSKLVSDFR KDFQFYKVREINNYHHAHDAYLNAVVGTALIKKYPKLESEFVYGDYKVYDVRKMIAKSEQEI GKATAKYFFYSNIMNFFKTEITLANGEIRKRPLIETNGETGEIVWDKGRDFATVRKVLSMPQ VNIVKKTEVOTGGFSKESILPKRNSDKLIARKKDWDPKKYGGFDSPTVAYSVLVVAKVEKGK SKKLKSVKELLGITIMERSSFEKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELENGRKRMLA SAGELQKGNELALPSKYVNFLYLASHYEKLKGSPEDNEQKQLFVEQHKHYLDEIIEQISEFS KRVILADANLDKVLSAYNKHRDKPIREQAENIIHLFTLTNLGAPAAFKYFDTTIDRKRYTST KEVLDATLIHQSITGLYETRIDLSQLGGDKRPAATKKAGQAKKKKSGGSPKKKRKVRPSGSE **TPGTSESATPESDSLLMNRREFLYQFKNVRWAKGRRETYLCYVVERRDCATSFSLDFGYLRN** KNGCHVELLFLRYISDWDLDPGRCYRVTWFISWSPCYDCARHVADFLRGNPNLSLRIFAARL YFCEDRKAEPEGLRRLRRAGVQIAIMTFKDYFYCWNTFAENHGRTFKAWEGLHENSVRLSGQ VEEVIGNKPESDILVHTAYDESTDENVMLLTSDAPEYKPWALVIODSNGENKIKMLSGGSRP **PPS**DYKDDDDKDYKDDDDK\*

### AID10-ABE7.10-nCas9-UGI

(AID10–XTEN–TadA–32aa linker–TadA7.10–32aa linker–nCas9–(GGS)<sub>6</sub> linker–UGI–NLS –NLS–2×FLAG)

MDSLLMNRREFLYOFKNVRWAKGRRETYLCYVVERRDCATSFSLDFGYLRNKNGCHVELLFL RYISDWDLDPGRCYRVTWFISWSPCYDCARHVADFLRGNPNLSLRIFAARLYFCEDRKAEPE GLRRLRRAGVOIAIMTFKDYFYCWNTFAENHGRTFKAWEGLHENSVRLSGOLRRILSGSETP **GTSESATPES**SEVEFSHEYWMRHALTLAKRAWDEREVPVGAVLVHNNRVIGEGWNRPIGRHD PTAHAEIMALROGGLVMONYRLIDATLYVTLEPCVMCAGAMIHSRIGRVVFGARDAKTGAAG SLMDVLHHPGMNHRVEITEGILADECAALLSDFFRMRRQEIKAQKKAQSSTDSGGSSGGSSG **SETPGTSESATPESSGGSSGGSSEVEFSHEYWMRHALTLAKRARDEREVPVGAVLVLNNRVI** GEGWNRAIGLHDPTAHAEIMALROGGLVMONYRLIDATLYVTFEPCVMCAGAMIHSRIGRVV FGVRNAKTGAAGSLMDVLHYPGMNHRVEITEGILADECAALLCYFFRMPRQVFNAQKKAQSS TDSGGSSGGSSGSETPGTSESATPESSGGSSGGSDKKYSIGLAIGTNSVGWAVITDEYKVPS KKFKVLGNTDRHSIKKNLIGALLFDSGETAEATRLKRTARRRYTRRKNRICYLQEIFSNEMA KVDDSFFHRLEESFLVEEDKKHERHPIFGNIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRL IYLALAHMIKFRGHFLIEGDLNPDNSDVDKLFIQLVQTYNQLFEENPINASGVDAKAILSAR LSKSRRLENLIAQLPGEKKNGLFGNLIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDN LLAQIGDQYADLFLAAKNLSDAILLSDILRVNTEITKAPLSASMIKRYDEHHQDLTLLKALV RQQLPEKYKEIFFDQSKNGYAGYIDGGASQEEFYKFIKPILEKMDGTEELLVKLNREDLLRK ORTFDNGSIPHOIHLGELHAILRROEDFYPFLKDNREKIEKILTFRIPYYVGPLARGNSRFA WMTRKSEETITPWNFEEVVDKGASAQSFIERMTNFDKNLPNEKVLPKHSLLYEYFTVYNELT KVKYVTEGMRKPAFLSGEOKKAIVDLLFKTNRKVTVKOLKEDYFKKIECFDSVEISGVEDRF NASLGTYHDLLKIIKDKDFLDNEENEDILEDIVLTLTLFEDREMIEERLKTYAHLFDDKVMK QLKRRRYTGWGRLSRKLINGIRDKQSGKTILDFLKSDGFANRNFMQLIHDDSLTFKEDIQKA QVSGQGDSLHEHIANLAGSPAIKKGILQTVKVVDELVKVMGRHKPENIVIEMARENQTTQKG QKNSRERMKRIEEGIKELGSQILKEHPVENTQLQNEKLYLYYLQNGRDMYVDQELDINRLSD YDVDHIVPOSFLKDDSIDNKVLTRSDKNRGKSDNVPSEEVVKKMKNYWROLLNAKLITORKF DNLTKAERGGLSELDKAGFIKRQLVETRQITKHVAQILDSRMNTKYDENDKLIREVKVITLK SKLVSDFRKDFOFYKVREINNYHHAHDAYLNAVVGTALIKKYPKLESEFVYGDYKVYDVRKM IAKSEQEIGKATAKYFFYSNIMNFFKTEITLANGEIRKRPLIETNGETGEIVWDKGRDFATV RKVLSMPOVNIVKKTEVOTGGFSKESILPKRNSDKLIARKKDWDPKKYGGFDSPTVAYSVLV VAKVEKGKSKKLKSVKELLGITIMERSSFEKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELE NGRKRMLASAGELQKGNELALPSKYVNFLYLASHYEKLKGSPEDNEQKQLFVEQHKHYLDEI IEQISEFSKRVILADANLDKVLSAYNKHRDKPIREQAENIIHLFTLTNLGAPAAFKYFDTTI **GGGGGSTNLSDIIEKETGKOLVIOESILMLPEEVEEVIGNKPESDILVHTAYDESTDENVMLL** TSDAPEYKPWALVIQDSNGENKIKMLSGGSKRPAATKKAGQAKKKKSGGSPKKKRKVRPPPS DYKDDDDKDYKDDDDK\*

# AID10-ABE8e-nCas9-UGI

(AID10–XTEN–TadA–32aa linker–TadA8e–32aa linker–nCas9–(GGS)<sub>6</sub> linker–UGI–NLS –NLS–2×FLAG)

MDSLLMNRREFLYQFKNVRWAKGRRETYLCYVVERRDCATSFSLDFGYLRNKNGCHVELLFL

RYISDWDLDPGRCYRVTWFISWSPCYDCARHVADFLRGNPNLSLRIFAARLYFCEDRKAEPE GLRRLRRAGVQIAIMTFKDYFYCWNTFAENHGRTFKAWEGLHENSVRLSGQLRRILSGSETP **GTSESATPES**SEVEFSHEYWMRHALTLAKRAWDEREVPVGAVLVHNNRVIGEGWNRPIGRHD PTAHAEIMALRQGGLVMQNYRLIDATLYVTLEPCVMCAGAMIHSRIGRVVFGARDAKTGAAG SLMDVLHHPGMNHRVEITEGILADECAALLSDFFRMRROEIKAOKKAOSSTDSGGSSGGSSG SETPGTSESATPESSGGSSGGSSEVEFSHEYWMRHALTLAKRARDEREVPVGAVLVLNNRVI GEGWNRAIGLHDPTAHAEIMALROGGLVMONYRLIDATLYVTFEPCVMCAGAMIHSRIGRVV FGVRNSKRGAAGSLMNVLNYPGMNHRVEITEGILADECAALLCDFYRMPROVFNAQKKAQSS INSGGSSGGSSGSETPGTSESATPESSGGSSGGSDKKYSIGLAIGTNSVGWAVITDEYKVPS KKFKVLGNTDRHSIKKNLIGALLFDSGETAEATRLKRTARRRYTRRKNRICYLOEIFSNEMA KVDDSFFHRLEESFLVEEDKKHERHPIFGNIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRL IYLALAHMIKFRGHFLIEGDLNPDNSDVDKLFIQLVQTYNQLFEENPINASGVDAKAILSAR LSKSRRLENLIAQLPGEKKNGLFGNLIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDN LLAQIGDQYADLFLAAKNLSDAILLSDILRVNTEITKAPLSASMIKRYDEHHQDLTLLKALV RQQLPEKYKEIFFDQSKNGYAGYIDGGASQEEFYKFIKPILEKMDGTEELLVKLNREDLLRK ORTFDNGSIPHOIHLGELHAILRROEDFYPFLKDNREKIEKILTFRIPYYVGPLARGNSRFA WMTRKSEETITPWNFEEVVDKGASAQSFIERMTNFDKNLPNEKVLPKHSLLYEYFTVYNELT KVKYVTEGMRKPAFLSGEQKKAIVDLLFKTNRKVTVKQLKEDYFKKIECFDSVEISGVEDRF NASLGTYHDLLKIIKDKDFLDNEENEDILEDIVLTLTLFEDREMIEERLKTYAHLFDDKVMK QLKRRRYTGWGRLSRKLINGIRDKQSGKTILDFLKSDGFANRNFMQLIHDDSLTFKEDIQKA QVSGQGDSLHEHIANLAGSPAIKKGILQTVKVVDELVKVMGRHKPENIVIEMARENQTTQKG QKNSRERMKRIEEGIKELGSQILKEHPVENTQLQNEKLYLYYLQNGRDMYVDQELDINRLSD YDVDHIVPQSFLKDDSIDNKVLTRSDKNRGKSDNVPSEEVVKKMKNYWRQLLNAKLITQRKF DNLTKAERGGLSELDKAGFIKRQLVETRQITKHVAQILDSRMNTKYDENDKLIREVKVITLK SKLVSDFRKDFQFYKVREINNYHHAHDAYLNAVVGTALIKKYPKLESEFVYGDYKVYDVRKM IAKSEOEIGKATAKYFFYSNIMNFFKTEITLANGEIRKRPLIETNGETGEIVWDKGRDFATV RKVLSMPOVNIVKKTEVOTGGFSKESILPKRNSDKLIARKKDWDPKKYGGFDSPTVAYSVLV VAKVEKGKSKKLKSVKELLGITIMERSSFEKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELE NGRKRMLASAGELQKGNELALPSKYVNFLYLASHYEKLKGSPEDNEQKQLFVEQHKHYLDEI IEQISEFSKRVILADANLDKVLSAYNKHRDKPIREQAENIIHLFTLTNLGAPAAFKYFDTTI GGGGGSTNLSDIIEKETGKQLVIQESILMLPEEVEEVIGNKPESDILVHTAYDESTDENVMLL TSDAPEYKPWALVIODSNGENKIKMLSGGSKRPAATKKAGOAKKKKSGGSPKKKRKVRPPPS DYKDDDDKDYKDDDDK\*

### ABE8e-nCas9

#### (TadA-32aa linker-TadA8e-32aa linker-nCas9-NLS-NLS-2×FLAG)

MGSEVEFSHEYWMRHALTLAKRAWDEREVPVGAVLVHNNRVIGEGWNRPIGRHDPTAHAEIM ALRQGGLVMQNYRLIDATLYVTLEPCVMCAGAMIHSRIGRVVFGARDAKTGAAGSLMDVLHH PGMNHRVEITEGILADECAALLSDFFRMRRQEIKAQKKAQSSTDSGGSSGGSSGSSGSETPGTSE SATPESSGGSSGGSSEVEFSHEYWMRHALTLAKRARDEREVPVGAVLVLNNRVIGEGWNRAI GLHDPTAHAEIMALRQGGLVMQNYRLIDATLYVTFEPCVMCAGAMIHSRIGRVVFGVRNSKR GAAGSLMNVLNYPGMNHRVEITEGILADECAALLCDFYRMPRQVFNAQKKAQSSINSGGSSG GSSGSETPGTSESATPESSGGSSGGSDKKYSIGL<mark>A</mark>IGTNSVGWAVITDEYKVPSKKFKVLGN TDRHSIKKNLIGALLFDSGETAEATRLKRTARRRYTRRKNRICYLQEIFSNEMAKVDDSFFH RLEESFLVEEDKKHERHPIFGNIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRLIYLALAHM IKFRGHFLIEGDLNPDNSDVDKLFIQLVQTYNQLFEENPINASGVDAKAILSARLSKSRRLE NLIAQLPGEKKNGLFGNLIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDNLLAQIGDQ YADLFLAAKNLSDAILLSDILRVNTEITKAPLSASMIKRYDEHHQDLTLLKALVRQQLPEKY KEIFFDQSKNGYAGYIDGGASQEEFYKFIKPILEKMDGTEELLVKLNREDLLRKQRTFDNGS IPHQIHLGELHAILRRQEDFYPFLKDNREKIEKILTFRIPYYVGPLARGNSRFAWMTRKSEE TITPWNFEEVVDKGASAQSFIERMTNFDKNLPNEKVLPKHSLLYEYFTVYNELTKVKYVTEG MRKPAFLSGEOKKAIVDLLFKTNRKVTVKOLKEDYFKKIECFDSVEISGVEDRFNASLGTYH DLLKIIKDKDFLDNEENEDILEDIVLTLTLFEDREMIEERLKTYAHLFDDKVMKQLKRRRYT GWGRLSRKLINGIRDKQSGKTILDFLKSDGFANRNFMQLIHDDSLTFKEDIQKAQVSGQGDS LHEHIANLAGSPAIKKGILQTVKVVDELVKVMGRHKPENIVIEMARENQTTQKGQKNSRERM KRIEEGIKELGSQILKEHPVENTQLQNEKLYLYYLQNGRDMYVDQELDINRLSDYDVDHIVP QSFLKDDSIDNKVLTRSDKNRGKSDNVPSEEVVKKMKNYWRQLLNAKLITQRKFDNLTKAER GGLSELDKAGFIKROLVETROITKHVAOILDSRMNTKYDENDKLIREVKVITLKSKLVSDFR KDFQFYKVREINNYHHAHDAYLNAVVGTALIKKYPKLESEFVYGDYKVYDVRKMIAKSEQEI GKATAKYFFYSNIMNFFKTEITLANGEIRKRPLIETNGETGEIVWDKGRDFATVRKVLSMPQ VNIVKKTEVQTGGFSKESILPKRNSDKLIARKKDWDPKKYGGFDSPTVAYSVLVVAKVEKGK SKKLKSVKELLGITIMERSSFEKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELENGRKRMLA SAGELQKGNELALPSKYVNFLYLASHYEKLKGSPEDNEQKQLFVEQHKHYLDEIIEQISEFS KRVILADANLDKVLSAYNKHRDKPIREOAENIIHLFTLTNLGAPAAFKYFDTTIDRKRYTST KEVLDATLIHQSITGLYETRIDLSQLGGDKRPAATKKAGQAKKKKSGGSPKKKRKVRPPPSD YKDDDDKDYKDDDDK\*