

SUPPLEMENTAL RESULTS:

Table S1: List of sgRNA used in the study.

SpCas9 VQR/EQR sgRNAs targeting sequences of 17 to 22 nucleotides tested:	
sgRNA 22bp	ATTCTGCATCCATCTTCACTTC
sgRNA 21bp	TTCTGCATCCATCTTCACTTC
sgRNA 20bp	TCTGCATCCATCTTCACTTC
sgRNA 19bp	CTGCATCCATCTTCACTTC
sgRNA 18bp	TGCATCCATCTTCACTTC
sgRNA 17bp	GCATCCATCTTCACTTC
SaCas9 sgRNAs tested:	
sgRNA 22bp	ATTCTGCATCCATCTTCACTTC
sgRNA 21bp	TTCTGCATCCATCTTCACTTC
sgRNA 20bp	TCTGCATCCATCTTCACTTC
sgRNA 19bp	CTGCATCCATCTTCACTTC
sgRNA 18bp	TGCATCCATCTTCACTTC
sgRNA 17bp	GCATCCATCTTCACTTC

Table S2: Example of Deep-Sequencing analysis.

	Target-AID-SpCas9nVQR 19	BE3_SpCas9nVQR 19
Total reads	100%	100%
Wild-Type	31,8	66,0
C1	3,8	1,0
C2	3,0	0,3
C3	0,1	3,6
C4	0,3	0,0
C5	0,8	0,3
C1+C2	26,2	0,0
C1+C3	0,2	2,1
C1+C4	0,2	0,0
C1+C5	0,2	0,0
C2+C3	0,1	0,2
C2+C4	0,1	0,0
C2+C5	0,2	0,0
C3+C4	0,1	0,2
C3+C5	0,0	0,2
C4+C5	0,0	0,0
C1+C2+C3	2,0	1,0
C1+C2+C4	1,6	0,0
C1+C2+C5	4,2	0,0
C2+C4+C5	0,0	0,0
C2+C3+C5	0,0	0,0
C1+C2+C3+C4	0,1	0,1
C2+C3+C4+C5	0,3	0,2
C1+C3+C4+C5	0,0	0,0
C1+C2+C4+C5	0,0	0,1
C1+C2+C3+C5	0,2	0,0
C1+C2+C3+C4+C5	0,3	0,2
Total	75,60%	75,70%
Mis-sequencing	24,40%	24,30%

Table S3: Percentage of reduction of amyloid- β peptides 40 and 42 induced by the addition of the A673T mutation to wild type APP gene or to an APP gene containing the London mutation or containing a C1 deamination (E674K). The addition of the A673T mutation reduced the production of A β 40 and A β 42 peptides in all 3 situations.

FAD mutation	Wild-Type	V717I (London)	A673T+E674 K (C1+C2)
Abeta42 Decrease (%)	-46	-65	-53
Abeta40 Decrease (%)	-63	-81	-44

Figure S1.

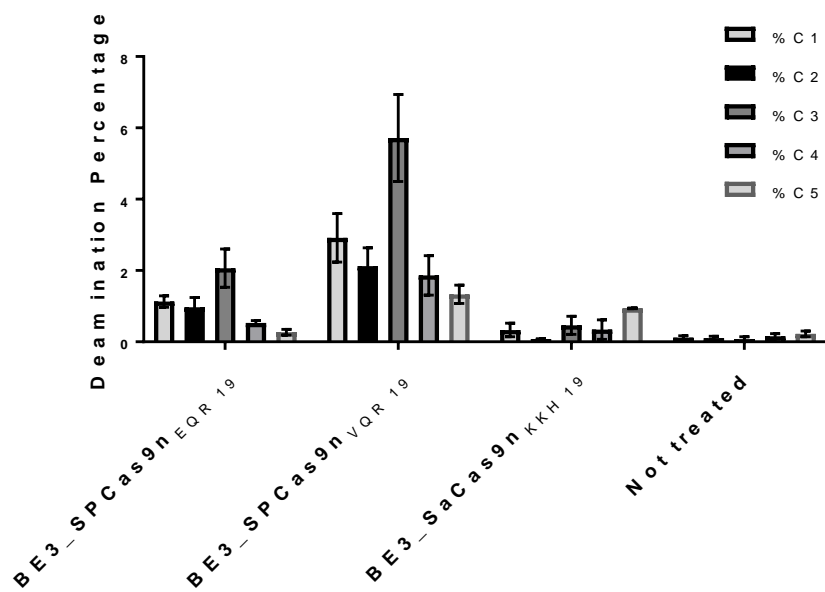


Figure S1: Percentages of cytidine deamination produced by various enzymes and sgRNAs. BE3_SpCas9nEQR, BE3_SpCas9nVQR, BE3_SaCas9nKKH enzymes test in SH-SY5Y cells. The figure illustrates the means +/- SEM (n=4).

Figure S2.

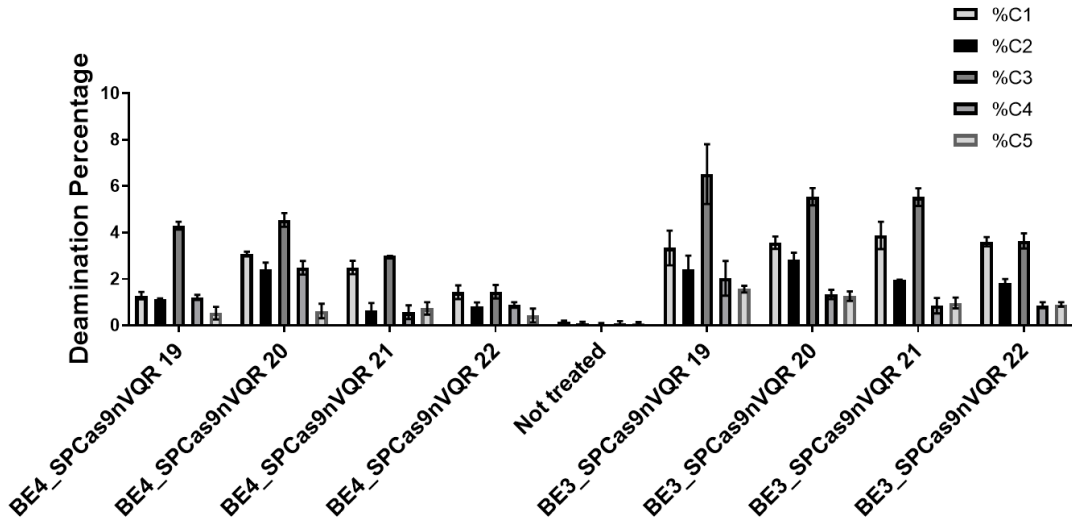


Figure S2: Percentages of cytidine deamination produced by various enzymes and sgRNAs.

BE4_SpCas9nVQR and BE3_SpCas9nVQR enzymes test in SH-SY5Y cells. The figure illustrates the means \pm SEM (n=3).

Figure S3.

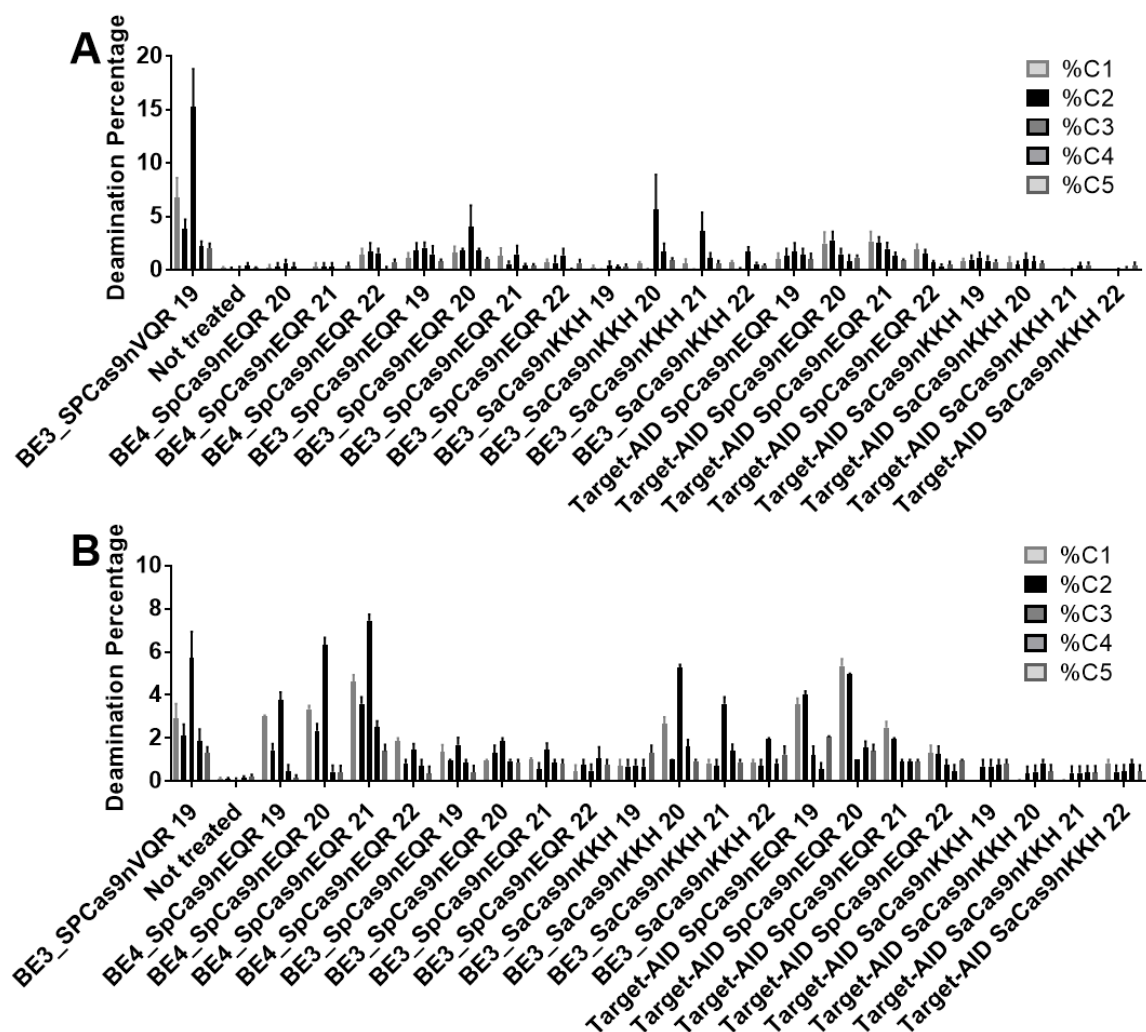


Figure S3: Percentages of cytosine deamination produced by BE3_SpCas9nVQR, BE4_SpCas9nVQR, BE3_SpCas9nVQR, BE3_SaCas9nKKH, Target-AID_SpCas9nVQR, Target-AID_SaCas9nKKH enzymes. In **A**, test in HEK293T cells. In **B**, test in SH-SY5Y. The figure illustrates the means \pm SEM (n=3).

Figure S4.

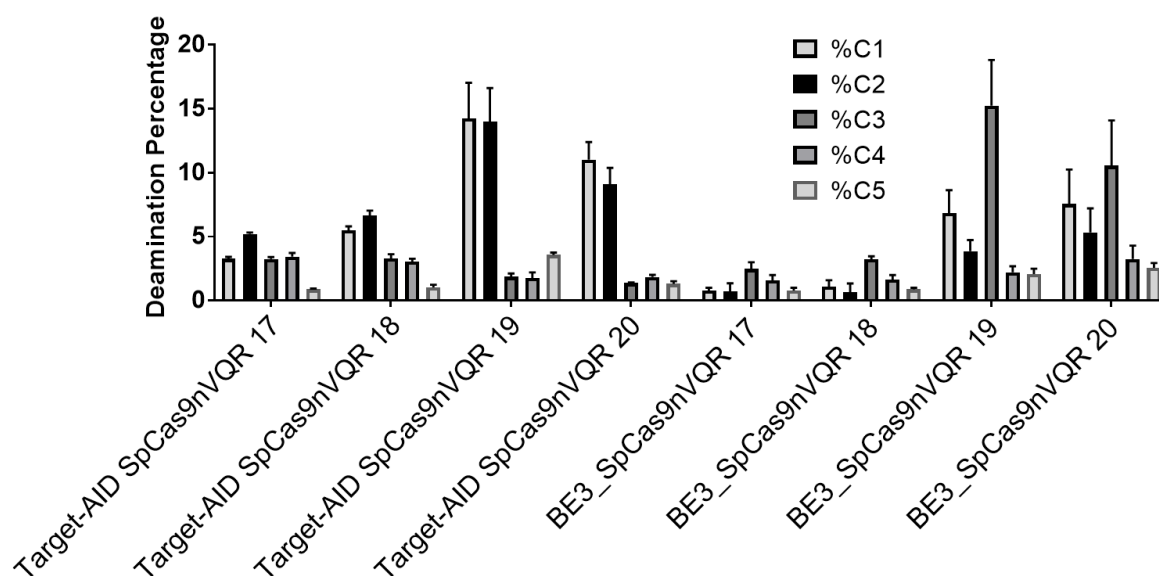


Figure S4: **Deamination efficiencies using various Cas9n-deaminases and sgRNAs targeting various numbers of nucleotides.** The difference of deamination in HEK293T cells of cytidines C1 to C5 produced by the Target-AID-SpCas9nVQR and BE3_SpCas9nVQR enzymes and two copies of a sgRNA targeting 17 to 20 nucleotides. The figure illustrates the means \pm SEM (n=3).

Figure S5

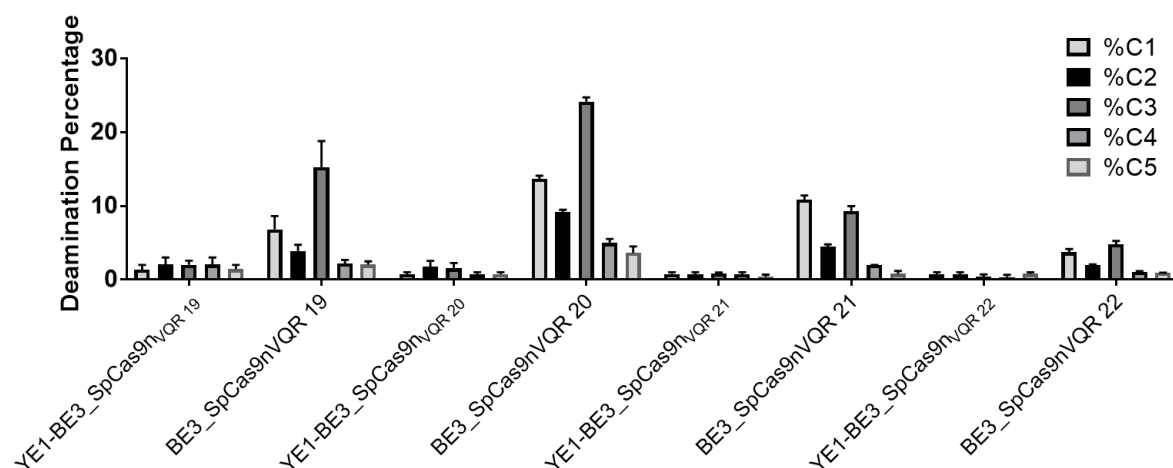


Figure S5: **Deamination efficiencies using various Cas9n-deaminases and sgRNAs targeting various numbers of nucleotides.** Difference between YE1-BE3_SpCas9nVQR and BE3_SpCas9nVQR in HEK293T cells. The figure illustrates the means \pm SEM (n=4).

Figure S6:

Off-Target Sites ×

Copy TSV

Sequence	PAM	Score ▼	Gene	Locus
CTGCATCCATCTTCACTTC	AGAG	100.0	APP (ENSG00000142192)	chr21:+25897633
CTGAAGCCATCTTCACTTC	GGAG	1.4		chr5:-74133250
CTGCCCTCCATCTTCACTG	TGAG	0.5		chr11:+70758797
CTGCTTCCAACCTTCACTTT	GGAG	0.5	SPOCK2 (ENSG00000107742)	chr10:-72059638
CAGGATCCATCTTAACCTTC	TGAG	0.4		chr12:-47654519
CTGCTTCCATCTTCTGTTC	AGAG	0.4		chr3:-36729648
CTGCATCCTTCTCCACTTG	GGAG	0.4		chr8:-67603274
CTGAATCAATCTCCACTTC	AGAG	0.4		chr12:-56572869
ATCCATGCATCTTCACTTC	AGAG	0.3		chr11:-20359838
CTGCCCCACCTTCACTTC	TGAG	0.3		chr9:-85398673
CTGCATCCATCTCTCCTTC	AGAG	0.3		chr3:+176747607
CTATTTCATCTTCACTTC	AGAG	0.3		chr9:-21442550
ATGTATCCATCTTCACTGT	TGAG	0.1		chr14:-35505959
TTTCATCCATCTCCACTTT	AGAG	0.1		chr5:-44542656
TTTCATCCATCTTAACCTAC	AGAG	0.1		chr10:-120808163
ATCCATCCACCTTCACTTG	TGAG	0.1		chr4:+10179124

Figure S6: Off target analysis performed with Benchling.com interface.