

**Additional File 10.** Sequences of synthetic parts.

> pU6::HCT\_gRNA1::tNOS

gacaagttgtacaaaaagcaggcttc**GGAGTGATCAAAAGTCCCACATCGATCAGGTGATATATAGCAGCTTAGTTTATA**  
**TAATGATAGAGTCGACATAGCGATTGTCGCTTGAAGAGAGACGATGA****GTTTTAGAGCTAGAAATAGCAAGTTAA**  
**AATAAGGCTAGTCCGTTATCAACTGAAAAAGTGGCACCGAGTCGGTGCCTTTTTTTT**cctaggTTCAAACATTTGGCA  
ATAAAGTTTCTTAAGATTGAATCCTGTTGCCGGTCTTGCATGATTATCATATAATTTCTGTTGAATTACGTTAAGCA  
TGTAATAATTAACATGTAATGCATGACGTTATTTATGAGATGGGTTTTTATGATTAGAGTCCCGCAATTATACATTT  
AATACGCGgaccagcttctgtacaaagtgtc

*HCT\_gRNA1 spacer sequence is shown in bold; U6 promoter sequence is shown in red; sgRNA scaffold sequence is shown in blue; NOS terminator sequence is shown in black; attB1 and attB2 sites are shown in lower case letters on 5' end and 3' end of the sequence.*

> HCT::f<sub>s</sub>STOP::2A::Cys4

ggacaagttgtacaaaaagcaggcttcATGTTAATTAACGGCGCGCCA**AAAATTAACATCAGAGATTCCACCATGGTCCG**  
**GCCTGCCACCGAGACACCAATCACTAATCTTTGAACTCCAACGTCGACCTTGTATCCCCAGATTCCATACCCCTA**  
**GTGTCTACTTCTACAGACCCACCGCGCTTCCAATTTCTTTGACCCTCAGGTCATGAAGGAAGCTCTTTCAAAGCC**  
**CTTGCCCTTTTTACCTATGGCTGGTCGCTTGAAGAGAGACGATGATGGTCGATTGAGATCGATTGTAACGGTG**  
**CTGGTGTCTCTCGTTGTGGCTGATACTCCTTCTGTTATCGATGATTTTGGTGATTTTGTCTCTACCCTTAATCTCCG**  
**TCAGCTTATCCCGAAGTTGATCACTCCGCTGGCATTCACTCTTTCCCGCTTCTCGTTTTGCAGGTGACTTTCTTTAA**  
**ATGTGGGGGAGCTTCACTTGGGGTTGGATGCAACATCACGCGGCAGATGGTTTCTCTGGTCTTCATTTTATCAAC**  
**ACATGGTCTGATATGGCTCGTGGTCTTGACCTAACCATTCCACCTTTTATTGATCGAACACTCCTCCGAGCTAGGGA**  
**CCCGCCACAGCCTGCTTTTTCATCATGTTGAATATCAGCCTGCACCAAGTATGAAGATACCTCTTGATCCGTCTAAAT**  
**CAGGACCTGAGAATACCACTGTCTCTATATCAAATTAACACGAGACCAGCTTGTTGCTCTTAAGGCGAAATCCAA**  
**GGAGGATGGGAACACTGTCAGCTACAGCTCATAAGATGTTGGCAGGGCATGTGTGGAGATCAGTGGGAAAG**  
**GCGCGAGGGCTTCAAACGACCAAGAGACGAACTGTACATTGCAACTGATGGAAGGTCTAGACTACGTCCGCAG**  
**CTGCCTCTGGTTACTTTGGGAATGTGATATTCACTGCAACACCATTGGCTGTTGCAGGGGATTTGTTATCTAAGCC**  
**AACATGGTATGCTGCAGGACAGATTCATGATTTCTGGTTCGTATGGATGATAACTATCTGAGGTGAGCTCTTGAC**  
**TACCTGGAGATGCAGCCTGATCTGTGAGCCCTTGTCCGCGGTGCACATACCTACAAGTGCCCAAATTTGGGAATCA**  
**CAAGCTGGGTTAGATTACCTATTTATGATGCAGACTTTGGTTGGGGTCGTCCTATCTTTATGGGACCTGGTGGAAAT**  
**TCCATACGAGGGTTTGTCTTTTGTGCTACCAAGTCTACTAATGATGGCAGCTTATCCGTTGCCATTGCCCTCCAATC**  
**TGAACACATGAACTGTTTGAAGAAGTTTTTGTGAGATAGCGCTTCAAGGTGCACATGGAGGGGGCGCGCCACG**  
**TGATCTAGGCCGT**CAGCTGTTGAATTTGACCTTCTAAGCTTGCGGGAGACGTGAGTCCAACCCTGGGCCCATG****  
**GCTCCAAAAAAGAAAAGAAAGTTGCTAGCGACCATTACTTAGATATTAGATTGAGACCAGACCCAGAGTTCCCTC**  
**CAGCACAGTTGATGTCAGTGTATTTCGGAAAATTGCACCAAGCTTTGGTTGCACAAGGTGGAGATAGAATTGGTG**  
**TGAGTTTTCTGATTTGGACGAGTCTAGGTCAAGATTGGGAGAAAGTTAAGAATCCATGCTTCAGCAGATGACCT**  
**TAGGGCTTTGTTAGCAAGACCTTGGTTAGAGGGTCTTAGAGATCATTGCAGTTCCGAGAACCAGCTGTTGTGCCA**  
**CACCCTACACCATATAGACAAGTTTCTAGAGTGCAGGCAAAATCAAATCCTGAGAGACTTAGAAGGAGATTGATG**  
**AGGAGACACGACTTATCTGAAGAGGAAGCTAGGAAGAGAATACCTGATACCGTTGCTAGAGCACTTGACTTGCCA**  
**TTTGTGACTTTGAGAAGTCAATCCACTGGTCAGCATTTTAGGTTATTCATTAGACACGGACCACTTCAAGTTACTGC**  
**AGAGGAGGGAGGATTCACCTGTTATGGATTGAGTAAGGGAGGATTCGTGCCTTGGTTCGATCCTGCATTTCTTTAT**  
AAGGTTGTTTGACaccaacttttctatacaagttgtcccc

*HCT gene is shown in yellow; a linker sequence containing a frameshifted stop codon cassette is shown in grey; potential stop codons being activated by genome editing in preceding sequences*

*are underlined; 2A peptide is shown in blue; Csy4 with nuclear localization signal is shown in orange; attB2 tag for immunoblotting is shown in black; attB1 and attB4 sites are shown in lower case letters on 5' end and 3' end of the sequence.*