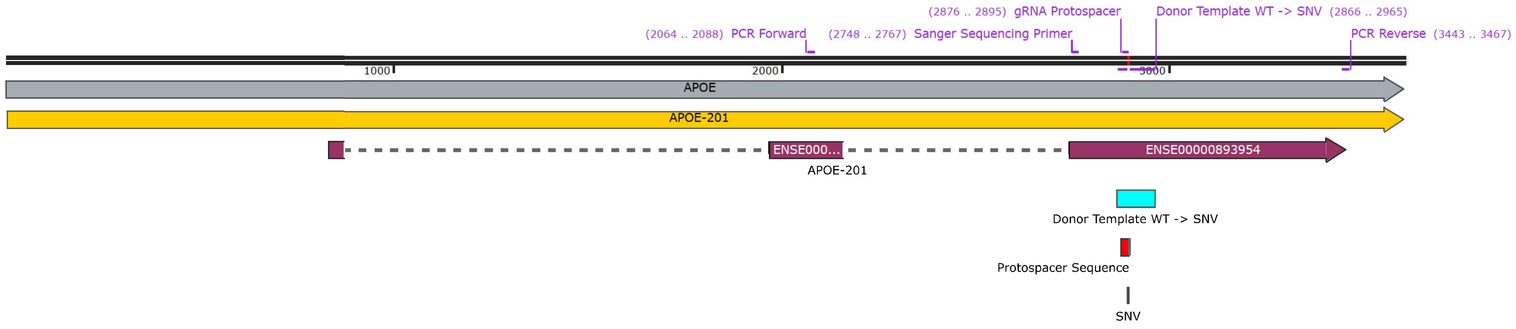

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3603 bp

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135
3′ CTCAGGATGAGTCGGGGTCGCCTCCACTTCCTGCAGGAAGGGGTCCTCGGCCACTCTTCGCGTCAGCCCCCGTGCCCCTACTCGAGTCCCCGGAGATCTTTCTCGACCCTGGGACCCTTGGGGACCGGAGGTCCA
```

APOE

APOE-201

```
AGTCTCAGGAGAGCTACTCGGGGTCGGGCTTGGGGAGAGGAGGAGCGGGGGTGAGGCAAGCAGCAGGGGACTGGACCTGGGAAGGGCTGGGCAGCAGAGACGACCCGACCCGCTAGAAGGTGGGGTGGGGAGAGC
270
TCAGAGTCCTCTCGATGAGCCCCAGCCCGAACCCCTCTCCTCCTCGCCCCCACTCCGTTCGTCGTCCCCTGACCTGGACCCTTCCCGACCCGTCGTCTCTGCTGGGCTGGGCGATCTTCCACCCCACCCCTCTCG
```

APOE

APOE-201

```
AGCTGGACTGGGATGTAAGCCATAGCAGGACTCCACGAGTTGTCACTATCATTTATCGAGCACCTACTGGGTGTCCCCAGTGTCCTCAGATCTCCATAACTGGGGAGCCAGGGGCAGCGACACGGTAGCTAGCCG
405
TCGACCTGACCCTACATTCGGTATCGTCCTGAGGTGCTCAACAGTGATAGTAAATAGCTCGTGGATGACCCACAGGGGTCACAGGAGTCTAGAGGTATTGACCCCTCGGTCCCCGTCGCTGTGCCATCGATCGGC
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APOE

APOE-201

```
TCGATTGGAGAACTTTAAAATGAGGACTGAATTAGCTCATAAATGGAACACGGCGCTTAACTGTGAGGTTGGAGCTTAGAATGTGAAGGGAGAATGAGGAATGCGAGACTGGGACTGAGATGGAACCGGCGGTGG
540
AGCTAACCTCTTGAAATTTTACTCCTGACTTAATCGAGTATTTACCTTGTGCCGCGAATTGACACTCCAACCTCGAATCTTACACTTCCCTCTTACTCCTTACGCTCTGACCCTGACTCTACCTTGGCCGCCACC
```

APOE

APOE-201

```
GGAGGGGGTGGGGGGATGGAATTTGAACCCCGGGAGAGGAAGATGGAATTTTCTATGGAGGCCGACCTGGGGATGGGGAGATAAGAGAAGACCAGGAGGGAGTTAAATAGGGAATGGGTTGGGGGCGGCTTGGTA
675
CCTCCCCCACCCCCCTACCTTAAACTTGGGGCCCTCTCCTTCTACCTTAAAAGATACCTCCGGCTGGACCCCTACCCCTCTATTCTCTTCTGGTCCTCCCTCAATTTATCCCTTACCCAACCCCCGCCGAACCAT
```

APOE

APOE-201

```
AATGTGCTGGGATTAGGCTGTTGCAGATAATGCAACAAGGCTTGGAAGGCTAACCTGGGGTGAGGCCGGGTTGGGGCCGGGCTGGGGGTGGGAGGAGTCCTCACTGGCGGTTGATTGACAGTTTCTCCTTCCCCA
810
TTACACGACCCTAATCCGACAACGTCTATTACGTTGTTCCGAACCTTCCGATTGGACCCCACTCCGGCCCAACCCCGGCCCGACCCCCACCCTCCTCAGGAGTGACCGCCAACTAACTGTCAAAGAGGAAGGGGT
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APOE

APOE-201

```
GACTGGCCAATCACAGGCAGGAAGATGAAGGTTCTGTGGGCTGCGTTGCTGGTCACATTCCTGGCAGGTATGGGGGCGGGGCTTGCTCGGTTCCCCCCGCTCCTCCCCCTCTCATCCTCACCTCAACCTCCTGGC
945
CTGACCGGTTAGTGTCCGTCCTTCTACTTCCAAGACACCCGACGCAACGACCAGTGTAAGGACCGTCCATACCCCCGCCCCGAACGAGCCAAGGGGGGCGAGGAGGGGGAGAGTAGGAGTGGAGTTGGAGGACCG
```

APOE

APOE-201

1 5 10

M K V L W A A L L V T F L A

ENSE00003577086

APOE-201

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1080
GGGTAAGTCCGTCTGGGACCCGGGGGAGAAGACTCCGAAGACACGACGAAGGACCGAGACTTGTCGCTAAACTGCGAGAGACCCGGAGCCAAAGGGGGTAGGAACTCTATCCTCAATCTTCAACAAAACAACAAC
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APOE

APOE-201

APOE-201

```
TTGTTTGTTGTTGTTGTTTTGTTTTTTTGAGATGAAGTCTCGCTCTGTCGCCCAGGCTGGAGTGCAGTGGCGGGATCTCGGCTCACTGCAAGCTCCGCCTCCCAGGTCCACGCCATTCTCCTGCCTCAGCCTCCC
1215
AACAAACAACAACAACAAAACAAAAAAACTCTACTTCAGAGCGAGACAGCGGGTCCGACCTCACGTCACCGCCCTAGAGCCGAGTGACGTTCGAGGCGGAGGGTCCAGGTGCGGTAAGAGGACGGAGTCGGAGGG
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APOE

APOE-201

APOE-201

```
AAGTAGCTGGGACTACAGGCACATGCCACCACACCCGACTAACTTTTTTGTATTTTCAGTAGAGACGGGGTTTCACCATGTTGGCCAGGCTGGTCTGGAACTCCTGACCTCAGGTGATCTGCCCGTTTCGATCTC
1350
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APOE

APOE-201

APOE-201

```
CCAAAGTGCTGGGATTACAGGCGTGAGCCACCGCACCTGGCTGGGAGTTAGAGGTTTCTAATGCATTGCAGGCAGATAGTGAATACCAGACACGGGGCAGCTGTGATCTTTATTCTCCATCACCCCCACACAGCC
1485
GGTTTCACGACCCTAATGTCCGCACTCGGTGGCGTGGACCGACCCTCAATCTCCAAAGATTACGTAACGTCCGTCTATCACTTATGGTCTGTGCCCCGTCGACACTAGAAATAAGAGGTAGTGGGGGTGTGTCGG
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APOE

APOE-201

APOE-201

```
CTGCCTGGGGCACACAAGGACACTCAATACATGCTTTTCCGCTGGGCGCGGTGGCTCACCCCTGTAATCCCAGCACTTTGGGAGGCCAAGGTGGGAGGATCACTTGAGCCCAGGAGTTCAACACCAGCCTGGGCA
1620
GACGGACCCCGTGTGTTCCTGTGAGTTATGTACGAAAAGGCGACCCGCGCCACCGAGTGGGGACATTAGGGTCGTGAAACCCTCCGGTTCCACCCTCCTAGTGAACTCGGGTCCTCAAGTTGTGGTCGGACCCGT
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APOE

APOE-201

APOE-201

```
ACATAGTGAGACCCTGTCTCTACTAAAAATACAAAAATTAGCCAGGCATGGTGCCACACACCTGTGCTCTCAGCTACTCAGGAGGCTGAGGCAGGAGGATCGCTTGAGCCCAGAAGGTCAAGGTTGCAGTGAACC
1755
TGTATCACTCTGGGACAGAGATGATTTTTATGTTTTTAATCGGTCCGTACCACGGTGTGTGGACACGAGAGTCGATGAGTCCTCCGACTCCGTCCTCCTAGCGAACTCGGGTCTTCCAGTTCCAACGTCACTTGG
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APOE

APOE-201

APOE-201


1890
APOE
APOE-201
2025
ENSE00000893952
2160
PCR Forward
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2295
2430
2565
2700
2835
Sanger Sequencing Primer
TGATGGACGAGACCATGAAG
ENSE00000893954
2970
gRNA Protospacer
GCGGACATGGAGGACGTGTG
Donor Template WT -> SNV
Protospacer Sequence
PAM
SNV
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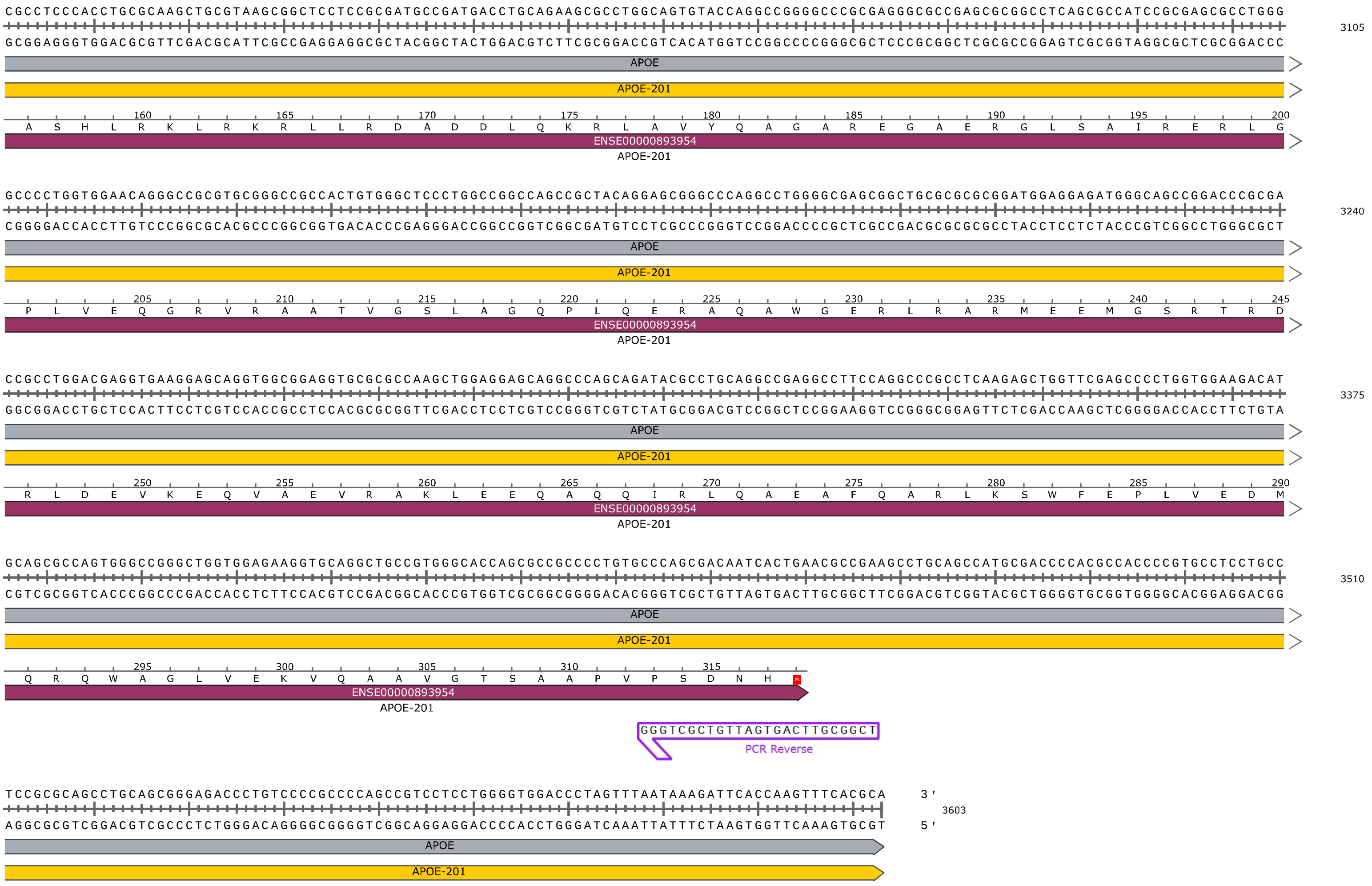
Donor Template WT -> SNV



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CGCCTCCCACCTGCGCAAGCTGCGTAAGCGGCTCCTCCGCGATGCCGATGACCTGCAGAAGCGCCTGGCAGTGTACCAGGCCGGGGCCCGCGAGGGCGCCGAGCGCGGCCTCAGCGCCATCCGCGAGCGCCTGGG   3105
GCGGAGGGTGGACGCGTTCGACGCATTCGCCGAGGAGGCGCTACGGCTACTGGACGTCTTCGCGGACCGTCACATGGTCCGGCCCCGGGCGCTCCCGCGGCTCGCGCCGGAGTCGCGGTAGGCGCTCGCGGACCC
 A  S  H  L  R  K  L  R  K  R  L  L  R  D  A  D  D  L  Q  K  R  L  A  V  Y  Q  A  G  A  R  E  G  A  E  R  G  L  S  A  I  R  E  R  L  G

GCCCCTGGTGGAACAGGGCCGCGTGCGGGCCGCCACTGTGGGCTCCCTGGCCGGCCAGCCGCTACAGGAGCGGGCCCAGGCCTGGGGCGAGCGGCTGCGCGCGCGGATGGAGGAGATGGGCAGCCGGACCCGCGA   3240
CGGGGACCACCTTGTCCCGGCGCACGCCCGGCGGTGACACCCGAGGGACCGGCCGGTCGGCGATGTCCTCGCCCGGGTCCGGACCCCGCTCGCCGACGCGCGCGCCTACCTCCTCTACCCGTCGGCCTGGGCGCT
 P  L  V  E  Q  G  R  V  R  A  A  T  V  G  S  L  A  G  Q  P  L  Q  E  R  A  Q  A  W  G  E  R  L  R  A  R  M  E  E  M  G  S  R  T  R  D

CCGCCTGGACGAGGTGAAGGAGCAGGTGGCGGAGGTGCGCGCCAAGCTGGAGGAGCAGGCCCAGCAGATACGCCTGCAGGCCGAGGCCTTCCAGGCCCGCCTCAAGAGCTGGTTCGAGCCCCTGGTGGAAGACAT   3375
GGCGGACCTGCTCCACTTCCTCGTCCACCGCCTCCACGCGCGGTTCGACCTCCTCGTCCGGGTCGTCTATGCGGACGTCCGGCTCCGGAAGGTCCGGGCGGAGTTCTCGACCAAGCTCGGGGACCACCTTCTGTA
 R  L  D  E  V  K  E  Q  V  A  E  V  R  A  K  L  E  E  Q  A  Q  Q  I  R  L  Q  A  E  A  F  Q  A  R  L  K  S  W  F  E  P  L  V  E  D  M

GCAGCGCCAGTGGGCCGGGCTGGTGGAGAAGGTGCAGGCTGCCGTGGGCACCAGCGCCGCCCCTGTGCCCAGCGACAATCACTGAACGCCGAAGCCTGCAGCCATGCGACCCCACGCCACCCCGTGCCTCCTGCC   3510
CGTCGCGGTCACCCGGCCCGACCACCTCTTCCACGTCCGACGGCACCCGTGGTCGCGGCGGGGACACGGGTCGCTGTTAGTGACTTGCGGCTTCGGACGTCGGTACGCTGGGGTGCGGTGGGGCACGGAGGACGG
 Q  R  Q  W  A  G  L  V  E  K  V  Q  A  A  V  G  T  S  A  A  P  V  P  S  D  N  H  *

TCCGCGCAGCCTGCAGCGGGAGACCCTGTCCCCGCCCCAGCCGTCCTCCTGGGGTGGACCCTAGTTTAATAAAGATTCACCAAGTTTCACGCA   3′
                                                                                             3603
AGGCGCGTCGGACGTCGCCCTCTGGGACAGGGGCGGGGTCGGCAGGAGGACCCCACCTGGGATCAAATTATTTCTAAGTGGTTCAAAGTGCGT   5′
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	Feature		Location	Size		⇄	Type
✓	**APOE**		1 .. 3603	3603 bp	◼	→	gene
	/note	= gene ENSG00000130203 Protein coding					
	APOE-204		1 .. 3154	3154 bp	◼	→	prim_transcript
	/note	= primary transcript ENST00000446996					
✓	**APOE-201**		6 .. 3603	3598 bp	🟨	→	prim_transcript
	/note	= primary transcript ENST00000252486					
	APOE-205		6 .. 1536	1531 bp	◼	→	prim_transcript
	/note	= primary transcript ENST00000485628 Retained intron					
	APOE-203		22 .. 3235	3214 bp	◼	→	prim_transcript
	/note	= primary transcript ENST00000434152					
	APOE-203		79 .. 3235	3157 bp	🟪	→	CDS
	▸ 4 segments = 807 bp						
	/note	= coding sequence ENSP00000413653					
	/translation	= MSSGASRKSWDPGNPWPP,,DWPITGRKMKVLWAALLVTFLA,,GCQAKVEQAVETEPEPELRQQTEWQSGQRWELALGRFWDYLRWVQTLSEQVQEELLSSQVTQELR,,ALMDETMKELKAYKSELEEQLTPVAEETRARLSK ELQAAQARLGADMEDVCGRLVQYRGEVQAMLGQSTEELRVRLASHLRKLRKRLLRDADDLQKRLAVYQAGAREGAERGLSAIRERLGPLVEQGRVRAATVGSLAGQPLQERAQAWGERLRARMEEMGSRT 269 amino acids = 30.6 kDa					
	APOE-202		570 .. 3164	2595 bp	◼	→	prim_transcript
	/note	= primary transcript ENST00000425718					
✓	**APOE-201**		835 .. 3460	2626 bp	🟪	→	CDS
	▸ 3 segments = 954 bp						
	/note	= coding sequence ENSP00000252486					
	/translation	= MKVLWAALLVTFLA,,GCQAKVEQAVETEPEPELRQQTEWQSGQRWELALGRFWDYLRWVQTLSEQVQEELLSSQVTQELR,,ALMDETMKELKAYKSELEEQLTPVAEETRARLSKELQAAQARLGADMEDVCGRLVQYRGE VQAMLGQSTEELRVRLASHLRKLRKRLLRDADDLQKRLAVYQAGAREGAERGLSAIRERLGPLVEQGRVRAATVGSLAGQPLQERAQAWGERLRARMEEMGSRTRDRLDEVKEQVAEVRAKLEEQAQQIRLQAEAFQARLK SWFEPLVEDMQRQWAGLVEKVQAAVGTSAAPVPSDNH* 317 amino acids = 36.2 kDa					
	APOE-202		835 .. 3164	2330 bp	🟪	→	CDS
	▸ 3 segments = 658 bp						
	/note	= coding sequence ENSP00000410423					
	/translation	= MKVLWAALLVTFLA,,GCQAKVEQAVETEPEPELRQQTEWQSGQRWELALGRFWDYLRWVQTLSEQVQEELLSSQVTQELR,,ALMDETMKELKAYKSELEEQLTPVAEETRARLSKELQAAQARLGADMEDVCGRLVQYRGE VQAMLGQSTEELRVRLASHLRKLRKRLLRDADDLQKRLAVYQAGAREGAERGLSAIRERLGPLVEQGRVRAATVGSLAGQ 219 amino acids = 24.9 kDa					
	APOE-204		835 .. 3154	2320 bp	🟪	→	CDS
	▸ 3 segments = 648 bp						
	/note	= coding sequence ENSP00000413135					
	/translation	= MKVLWAALLVTFLA,,GCQAKVEQAVETEPEPELRQQTEWQSGQRWELALGRFWDYLRWVQTLSEQVQEELLSSQVTQELR,,ALMDETMKELKAYKSELEEQLTPVAEETRARLSKELQAAQARLGADMEDVCGRLVQYRGE VQAMLGQSTEELRVRLASHLRKLRKRLLRDADDLQKRLAVYQAGAREGAERGLSAIRERLGPLVEQGRVRAATVGSL 216 amino acids = 24.6 kDa					
✓	**Donor Template WT -> SNV**		2866 .. 2965	100 bp	🟦	⟷	misc_feature
✓	**Protospacer Sequence**		2876 .. 2895	20 bp	🟥	⟷	misc_feature
✓	**SNV**		2894 .. 2894	1 bp	🟧	⟷	misc_feature
	/note	= WT = T SNV = C					
✓	**PAM**		2896 .. 2898	3 bp	🟨	⟷	misc_feature
			3585 .. 9178	5594 bp	◼	→	gene
	/note	= gene ENSG00000280087 TEC					
			3585 .. 9178	5594 bp	◼	→	prim_transcript
	/note	= primary transcript ENST00000623895 TEC					

	Primer	Length		Binding Sites	⇄	Tm	Date Added
✓	**PCR Forward**	25-mer		2064 .. 2088	→	65°C	Oct 12, 2022
	/sequence = GGCACTGGGTCGCTTTTGGGATTAC 56% GC / 7720.1 Da						
✓	**Sanger Sequencing Primer**	20-mer		2748 .. 2767	→	56°C	Oct 12, 2022
	/sequence = TGATGGACGAGACCATGAAG 50% GC / 6215.1 Da						
✓	**Donor Template WT -> SNV**	100-mer		2866 .. 2965	←	88°C	Oct 12, 2022
	/sequence = CACCCGCAGCTCCTCGGTGCTCTGGCCGAGCATGGCCTGCACCTCGCCGCGGTACTGCACCAGGCGGCCGCGCACGTCCTCCATGTCCGCGCCCAGCCGG 75% GC / 30,531.6 Da						
✓	**gRNA Protospacer**	20-mer		2876 .. 2895	→	62°C	Oct 12, 2022
	/sequence = GCGGACATGGAGGACGTGTG 65% GC / 6263.1 Da						
✓	**PCR Reverse**	25-mer		3443 .. 3467	←	67°C	Oct 12, 2022
	/sequence = TCGGCGTTCAGTGATTGTCGCTGGG 60% GC / 7736.1 Da						