

**INK2J00079R\_VAPB\_D130E\_H04\_AA**  
1009 bp

5' CTCCCAAAGTGCTGGGATTACAGGCATAAGCCACCATGCCAGCTGAAAACAAGTTTTATTTTCATAAGGAGAGCCAAGGTTACC 85  
 3' GAGGGTTTCACGACCCTAATGTCCGTATTCCGGTGGTACGGGTCGACTTTTGTTCAAAATAAAAGTATTCCTCTCGGTTCCAATGG

VAPB  
 VAPB-204

PCR Forward  
 CCTGTCCCACAGAGTAGCAACATGG

TGGTCATCCAGACATTGTTTAGAGGATTCCTTTTTGTTTCAGTGGTGGGCCTGTCCCACAGAGTAGCAACATGGGGAGCATTGGTT 170  
 ACCAGTAGGTCTGTAACAAATCTCCTAAGGAAAAACAAGTCAACCACCCGGACAGGGTGTCTCATCGTTGTACCCCTCGTAACCAA

VAPB  
 VAPB-204

GACTTTGAGGAGTCAGGCAGCAAGACTTCAGGGTTTATGAAGTGAATATTTTGGAGAAAGTGAGTGTATTTAATCTTGTTCATGTT 255  
 CTGAAACTCCTCAGTCCGTCGTTCTGAAGTCCCAAATACTTCACATTATAAAACTCTTTCCTCACATAAATTAGAACAGTACAA

VAPB  
 VAPB-204

ACAAATATTGAAATCTCAGCAGACTTCAGTTCTTTATTTGATATACATCAGGGCTTTCTCATTAAGAGTATTTTTCTGAAATTGT 340  
 TGTATAAATTTAGAGTCGTCTGAAGTCAAGAAATAAACTATATGTAGTCCCGAAAGAGTAATTCATAAAAAGACTTTAACA

VAPB  
 VAPB-204

Donor Template SNV -> Rev

GGAAGGAGGCAAA

CAGTTATAGGAAGAAAGTTATTCTTCCAGCAGGTTTATAATATCTTGATTATTAATTTAAATTGTTTTAGTGGAAAGGAGGCAAA 425  
 GTCAATATCCTTCTTTCAATAAGAAGGTCGTCCAAATATTATAGAACTAATAATTTAAATTTAACAAAATCACCTTCTCCGTTT

VAPB  
 VAPB-204

W K E A K  
 ENSE00003576198

VAPB-204

Donor Template Sequence SNV -> Rev

Donor Template SNV -> Rev

ACCGGAAGACCTTATGGATTCAAAACTTAGATGTGTGTTTGAATTGCCAGCAGAGAATGATAAACCCAGTAAGTATATTTATAGTT 510  
 ACCGGAAGACCTTATGGATTCAAAACTTAGATGTGTGTTTGAATTGCCAGCAGAGAATGATAAACCCAGTAAGTATATTTATAGTT  
 TGGCCTTCTGGAATACCTAAGTTTTGAATCTACACACAAACTTAACGGTCTGCTCTTACTATTTGGTTCATTCATATAAATATCAA

VAPB  
 VAPB-204

P E D L M D S K L R C V F E L P A E N D K P V S I F I V  
 ENSE00003576198 (in frame with VAPB-204)

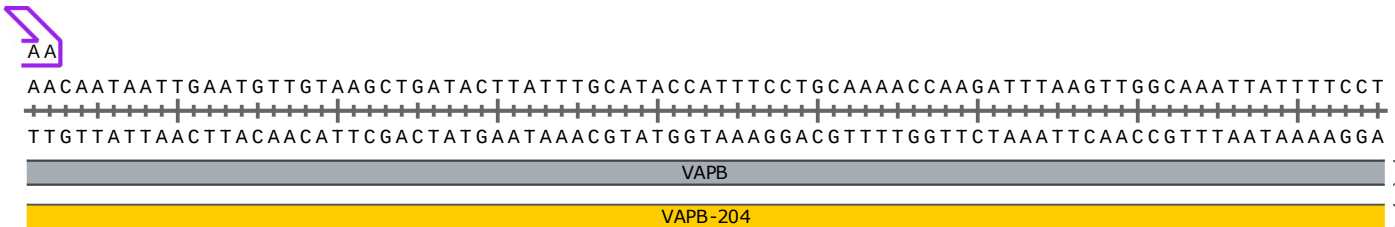
Donor Template Sequence SNV -> Rev

PAM gRNA Protospacer Sequence

SNV

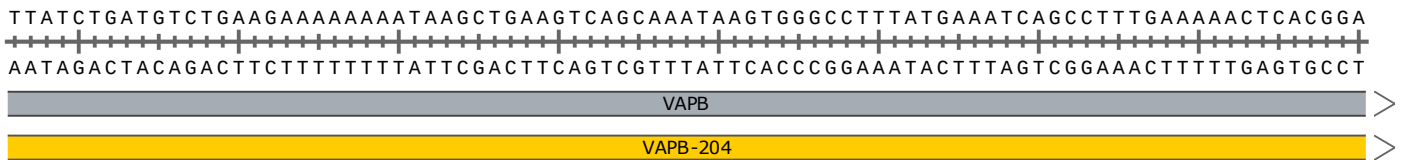
CGTCTTACTTTGGTCA  
 gRNA Protospacer

Donor Template SNV -> Rev

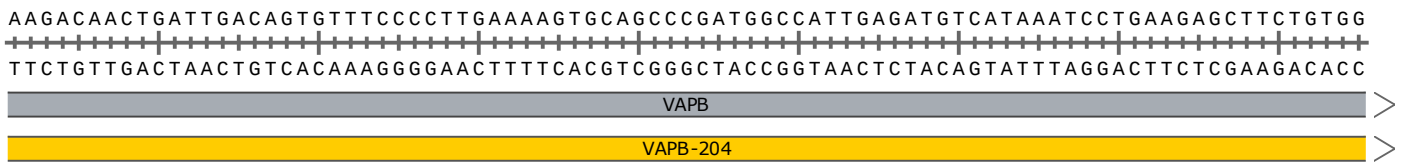


595

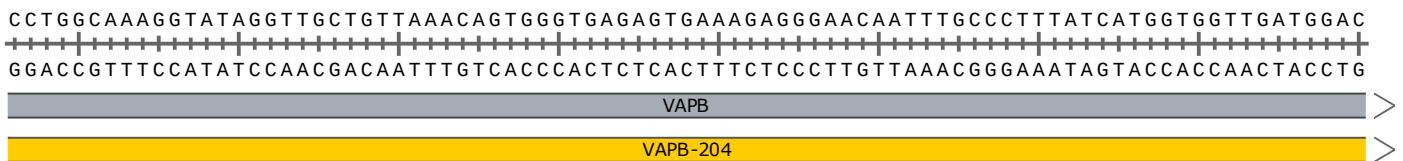
GGA  
Sanger Sequencing Primer



680

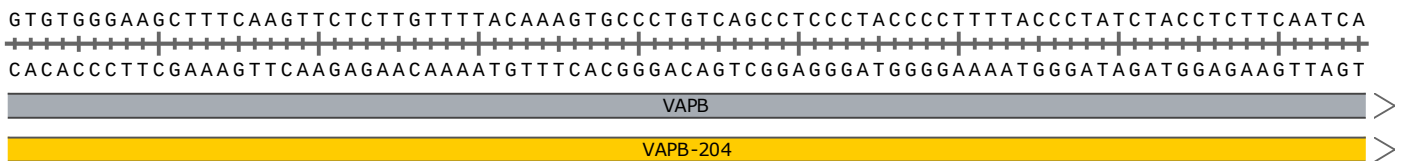


765

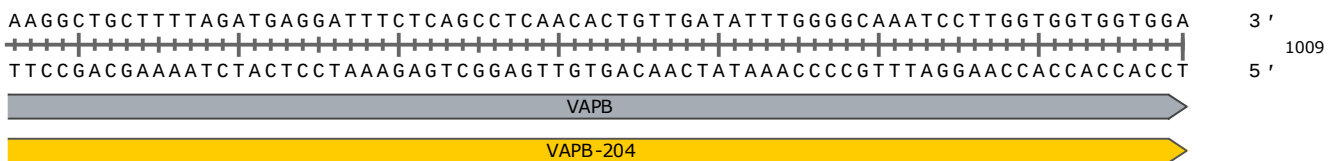


850

CCTG  
PCR Reverse



935



Feature	Location	Size	Start	End	Type
✓ <b>VAPB</b>	1 .. 1009	1009 bp	■	→	gene
/note = gene <a href="#">ENSG00000124164</a> Protein coding					
<b>VAPB-201</b>	1 .. 1009	1009 bp	■	→	prim_transcript
/note = primary transcript <a href="#">ENST00000265619</a> protein_coding_CDS_not_defined					
<b>VAPB-202</b>	1 .. 1009	1009 bp	■	→	prim_transcript
/note = primary transcript <a href="#">ENST00000395802</a>					
<b>VAPB-203</b>	1 .. 1009	1009 bp	■	→	prim_transcript
/note = primary transcript <a href="#">ENST00000463370</a> protein_coding_CDS_not_defined					
✓ <b>VAPB-204</b>	1 .. 1009	1009 bp	■	→	prim_transcript
/note = primary transcript <a href="#">ENST00000475243</a>					
<b>VAPB-206</b>	1 .. 1009	1009 bp	■	→	prim_transcript
/note = primary transcript <a href="#">ENST00000520497</a> Nonsense mediated decay					
✓ <b>VAPB-204</b>	412 .. 492	81 bp	■	→	CDS
/note = coding sequence <a href="#">ENSP00000417175</a>					
/translation = WKEAKPEDLMDSKLRVCFELPAENDKP 27 amino acids = 3.2 kDa					
✓ <b>Donor Template Sequence SNV -&gt; Rev</b>	413 .. 512	100 bp	■		misc_feature
<b>VAPB-205</b>	444 .. 1009	566 bp	■	→	prim_transcript
/note = primary transcript <a href="#">ENST00000476395</a> Retained intron					
✓ <b>PAM</b>	472 .. 474	3 bp	■		misc_feature
✓ <b>gRNA Protospacer Sequence</b>	475 .. 494	20 bp	■		misc_feature
✓ <b>SNV</b>	486 .. 486	1 bp	■		misc_feature
/note = SNV = G WT = T					

Primer	Length	Binding Sites	Tm	Date Added
✓ <b>PCR Forward</b>  /sequence = CCTGTCCCACAGAGTAGCAACATGG 56% GC / 7636.0 Da	25-mer	134 .. 158	62°C	Aug 15, 2023
✓ <b>Donor Template SNV -&gt; Rev</b>  /sequence = GGAAGGAGGCAAAACCGAAGACCTTATGGATTCAAACCTTAGATGTGTGTTTGAATTGCCAGCAGAGAATGATAAACCCAGTAAGTATATTTATAGTTAA 36% GC / 31,120.4 Da	100-mer	413 .. 512	71°C	Aug 15, 2023
✓ <b>gRNA Protospacer</b>  /sequence = ACTGGTTTCTCATTCTCTGC 45% GC / 6025.0 Da	20-mer	475 .. 494	46°C	Aug 15, 2023
✓ <b>Sanger Sequencing Primer</b>  /sequence = CTTCAGACATCAGATAAAGG 40% GC / 6134.1 Da	20-mer	593 .. 612	51°C	Aug 15, 2023
✓ <b>PCR Reverse</b>  /sequence = GAACTTGAAAGCTTCCCACACGTCC 52% GC / 7571.0 Da	25-mer	847 .. 871	61°C	Aug 15, 2023