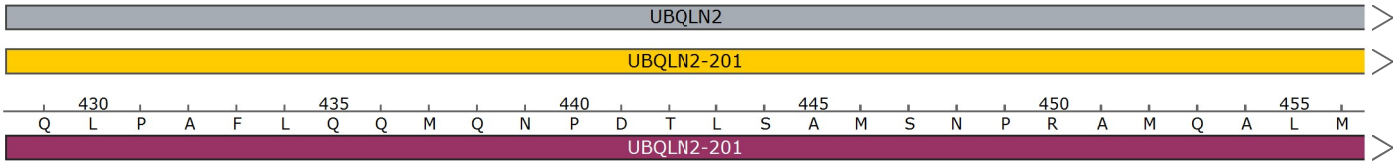


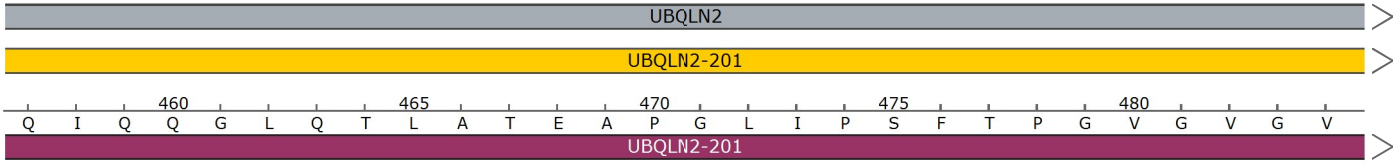
Sanger Sequencing Primer  
**AGCCATGTCAAACCCAAGAG**

PCR Forward  
**AGATGCAGAATCCAGACACACTATC**

5' ACAGCTCCCAGCCTTCTGCAGCAGATGCAGAATCCAGACACACTATCAGCCATGTCAAACCCAAGAGCAATGCAGGCTTTAATG  
 3' TGTCGAGGGTTCGGAAGGACGTCGTCTACGCTCTTAGGCTGTGTGATAGTCGGTACAGTTTGGGTTCTCGTTACGTCGGAATTAC

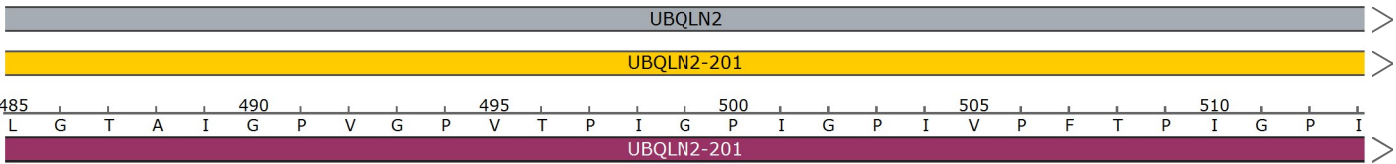


CAGATCCAGCAGGGGCTACAGACATTAGCCACTGAAGCACCTGGCCTGATTCCGAGCTTCACTCCAGGTGTGGGGGTGGGGGTGC  
 GTCTAGGTCGTCCCCGATGTCTGTAATCGGTGACTTCGTGGACCGGACTAAGGCTCGAAGTGAGGTCCACACCCCCACCCCCACG



gRNA Protospacer  
**GTAGGCCAGTCACCCCAT**

TGGGAACCGCTATAGGCCCTGTAGGCCAGTCACCCCATAGGCCCATAGGCCCTATAGTCCCTTTTACCCCATAGGCCCAT  
 ACCCTTGGCGATATCCGGGACATCCGGGTCAGTGGGGGTATCCGGGGTATCCGGGATATCAGGGAAAATGGGGGTATCCGGGGTA



DNA Donor Template Sequence WT -> SNV

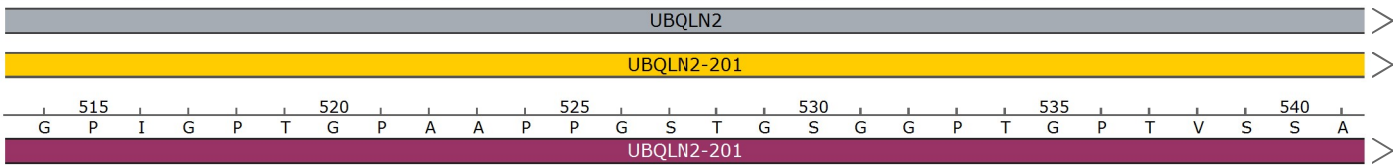
gRNA Protospacer Sequence PAM

SNV

**CCTTGGCGATATCCGGGACATCCGGGTCAGTGGGGTATCCGGGGTATCCGGGATATCAGGGAAAATGGGGGTATCCGGGGTA**

DNA Donor Template WT -> SNV

TGGGCCCATAGGACCCACTGGCCCTGCAGCCCCCCTGGCTCCACCGGCTCTGGTGGCCCCACGGGGCCTACTGTGTCCAGCGCT  
 ACCCGGGTATCCTGGGTGACCGGGACGTCGGGGGGGACCGAGGTGGCCGAGACCACCGGGGTGCCCGGATGACACAGGTTCGCGA



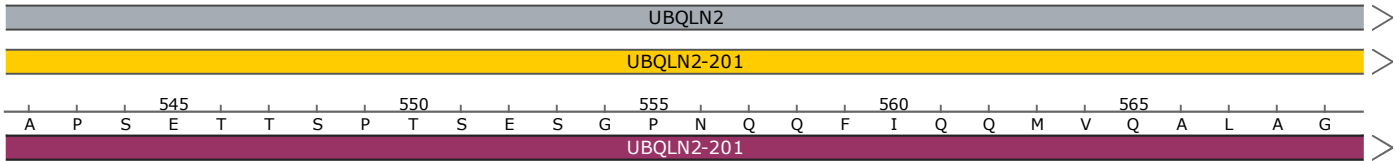
DNA Donor Template Sequence WT -> SNV

**ACCCGGGTATCCTGGGT**

DNA Donor Template WT -> SNV

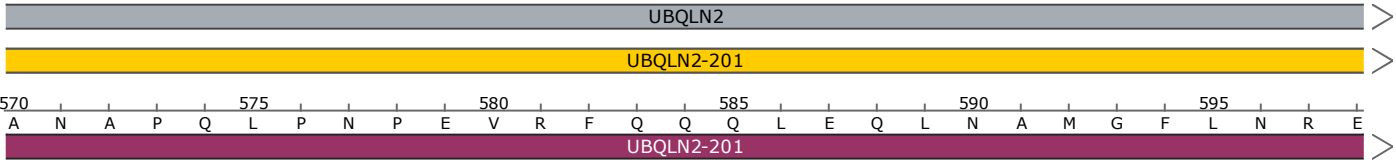
GCACCTAGTGAAACCACGAGTCCTACATCAGAATCTGGACCCAACCAGCAGTTTCATTTCAGCAAATGGTGCAGGCCCTGGCTGGAG  
 CGTGGATCACTTTGGTGCTCAGGATGTAGTCTTAGACCTGGGTTGGTCGTCAAGTAAGTCGTTTACCACGTCCGGGACCGACCTC

425



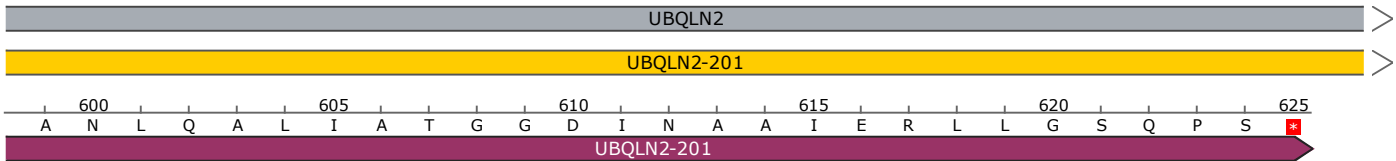
CAAATGCTCCACAGCTGCCGAATCCAGAAGTCAGATTTTCAGCAACAACCTGGAACAGCTCAACGCAATGGGGTTCTTAAACCGTGA  
 GTTTACGAGGTGTCGACGGCTTAGGTCTTCAGTCTAAAGTCGTTGTTGACCTTGTTCGAGTTGCGTTACCCCAAGAATTTGGCACT

510



AGCAAACCTGCAGGCCCTAATAGCAACAGGAGGCGACATCAATGCAGCCATTGAAAGGCTGCTGGGCTCCCAGCCATCGTAATCA  
 TCGTTTGAACGTCCGGGATTATCGTTGTCCTCCGCTGTAGTTACGTCGGTAACTTTCCGACGACCCGAGGGTTCGGTAGCATTAGT

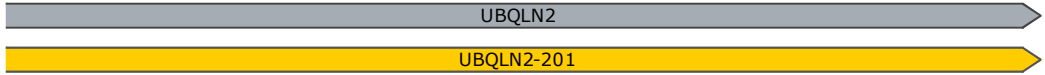
595



CGGTAGCATTAGT  
 PCR Reverse

CATTTCTGTACCTGGAAAAAAAAATGTATCTTATTTTTGATAATGGCTCTTAAATCTTTAAACACA  
 GTAAAGACATGGACCTTTTTTTTACATAGAATAAAAACCTATTACCGAGAATTTAGAAATTTGTGT

3 '  
 660  
 5 '



GTAAAGACATGG  
 PCR Reverse

Feature	Location	Size		→	Type
✓ <b>UBQLN2</b>	1 .. 660	660 bp	■	→	gene
/note = gene <a href="#">ENSG00000188021</a> Protein coding					
✓ <b>UBQLN2-201</b>	1 .. 660	660 bp	■	→	prim_transcript
/note = primary transcript <a href="#">ENST00000338222</a>					
✓ <b>UBQLN2-201</b>	1 .. 592	592 bp	■	→	CDS
/codon_start = 1					
/note = coding sequence <a href="#">ENSP00000345195</a>					
/translation = QLPAFLQQMQNPDTLSAMSNPRAMQALMQIQQGLQTLATEAPGLIPSFTPGVGVGVLTGAIGPVGVPVTPIGPIGPIVPFTPIGPIGPIGPTGPAAPPSTGSGGPTGPTVSSA APSETTSPTSESGPNQQFIQQMVQALAGANAPQLPNPEVRFQQLEQLNAMGFLNREANLQALIATGGDINAAIERLLGSQPS* 196 amino acids = 19.7 kDa					
✓ <b>DNA Donor Template Sequence WT -&gt; SNV</b>	173 .. 272	100 bp	■	⌊	misc_feature
✓ <b>gRNA Protospacer Sequence</b>	191 .. 210	20 bp	■	⌊	misc_feature
✓ <b>SNV</b>	207 .. 207	1 bp	■	⌊	misc_feature
/note = WT = C SNV = A					
✓ <b>PAM</b>	211 .. 213	3 bp	■	⌊	misc_feature

Primer	Length	Binding Sites	Tm	Date Added
✓ <b>PCR Forward</b>  /sequence = AGATGCAGAATCCAGACACACTATC 44% GC / 7628.0 Da	25-mer	24 .. 48	58°C	Aug 15, 2023
✓ <b>Sanger Sequencing Primer</b>  /sequence = AGCCATGTCAAACCCAAGAG 50% GC / 6104.0 Da	20-mer	49 .. 68	57°C	Aug 15, 2023
✓ <b>DNA Donor Template WT -&gt; SNV</b>  /sequence = TGGGTCCTATGGGCCAATGGGGCCTATGGGGTAAAAGGGACTATAGGGCCTATGGGGCCTATGTGGGTGACTGGGCCTACAGGGCCTATAGCGGTTCC 60% GC / 31,180.2 Da	100-mer	173 .. 272	81°C	Aug 15, 2023
✓ <b>gRNA Protospacer</b>  /sequence = GTAGGCCAGTCACCCCAT 65% GC / 6022.9 Da	20-mer	191 .. 210	63°C	Aug 15, 2023
✓ <b>PCR Reverse</b>  /sequence = GGTACAGAAATGTGATTACGATGGC 44% GC / 7770.1 Da	25-mer	583 .. 607	58°C	Aug 15, 2023