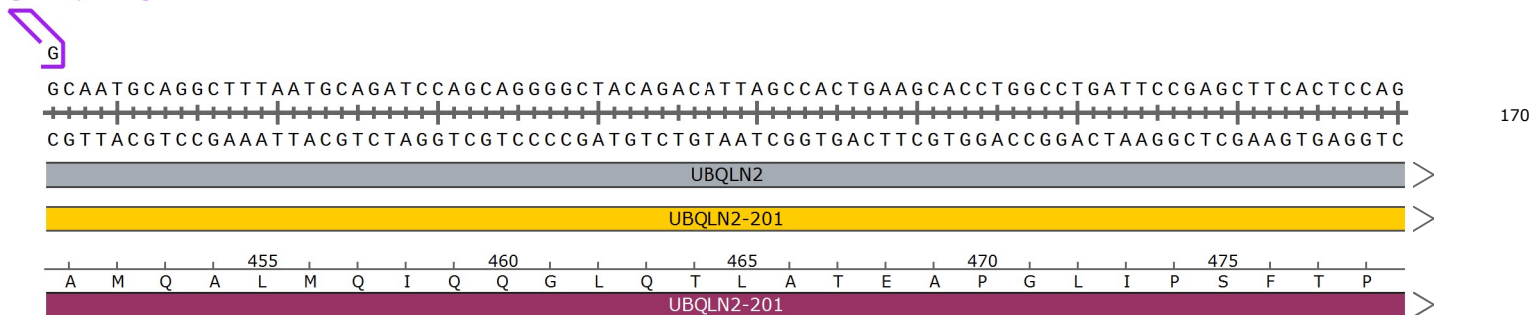
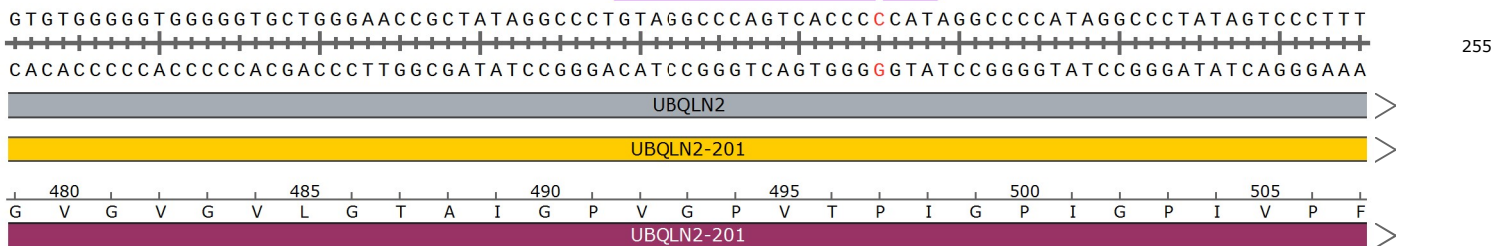


Sanger Sequencing Primer



gRNA Protospacer

GTAGGCCAGTCACCCACAT



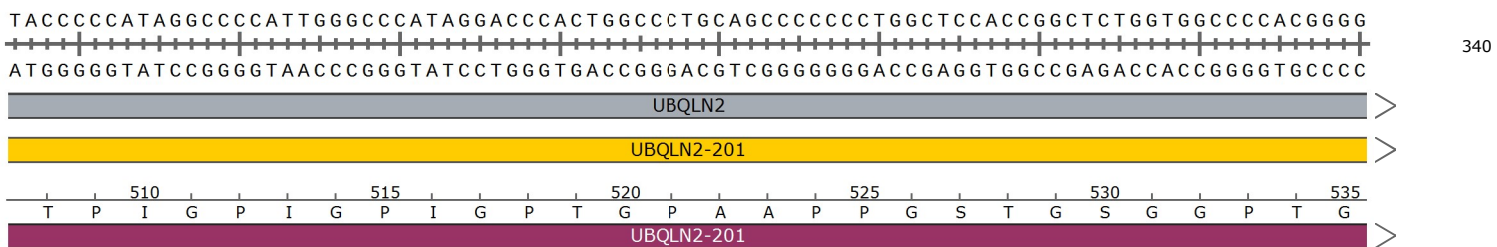
DNA Donor Template Sequence SNV -> Rev

gRNA Protospacer Sequence PAM

SNV

CCTTGGCGATATCCGGGACATCCGGGTGAGTGGGGGTATCCGGGGTATCCGGGATATCAGGGAAA

DNA Donor Template SNV -> Rev



DNA Donor Template Sequence SNV -> Rev

ATGGGGGTATCCGGGGTAACCCGGGTATCCTGGGT

DNA Donor Template SNV -> Rev

Feature	Location	Size		→	Type
✓ UBQLN2	1 .. 698	698 bp	■	→	gene
/note = gene ENSG00000188021 Protein coding					
✓ UBQLN2-201	1 .. 698	698 bp	■	→	prim_transcript
/note = primary transcript ENST00000338222					
✓ UBQLN2-201	1 .. 610	610 bp	■	→	CDS
/codon_start = 1					
/note = coding sequence ENSP00000345195					
/translation = QEQMRPQLPAFLQQMQNPDTLSAMSNPRAMQALMQIQQQLTLATEAPGLIPSFPTPGVGVGLGTAIGPVGPVTPIGPIGPIVPTPIGPIGPIGPTGPAAPPSTGSGGPTG PTVSSAAPSETTSPTSESGPNQQFIQQMVQALAGANAPQLPNPEVRFQQLEQLNAMGFLNREANLQALIATGGDINAAIERLLGSQPS* 202 amino acids = 20.5 kDa					
✓ DNA Donor Template Sequence SNV -> Rev	191 .. 290	100 bp	■	⌊	misc_feature
✓ gRNA Protospacer Sequence	209 .. 228	20 bp	■	⌊	misc_feature
✓ SNV	225 .. 225	1 bp	■	⌊	misc_feature
/note = SNV = A REV = C					
✓ PAM	229 .. 231	3 bp	■	⌊	misc_feature

Primer	Length	Binding Sites	Tm	Date Added
✓ PCR Forward /sequence = AGATGCAGAATCCAGACACACTATC 44% GC / 7628.0 Da	25-mer	42 .. 66	58°C	Aug 15, 2023
✓ Sanger Sequencing Primer /sequence = AGCCATGTCAAACCCAAGAG 50% GC / 6104.0 Da	20-mer	67 .. 86	57°C	Aug 15, 2023
✓ DNA Donor Template SNV -> Rev /sequence = TGGGTCCTATGGGCCAATGGGGCCTATGGGGTAAAAGGGACTATAGGGCCTATGGGGCCTATGGGGGTGACTGGGCCTACAGGGCCTATAGCGGTTCC 61% GC / 31,205.2 Da	100-mer	191 .. 290	82°C	Aug 15, 2023
✓ gRNA Protospacer /sequence = GTAGGCCAGTCACCCACAT 60% GC / 6047.0 Da	20-mer	209 .. 228	58°C	Aug 15, 2023
✓ PCR Reverse /sequence = GGTACAGAAATGTGATTACGATGGC 44% GC / 7770.1 Da	25-mer	601 .. 625	58°C	Aug 15, 2023