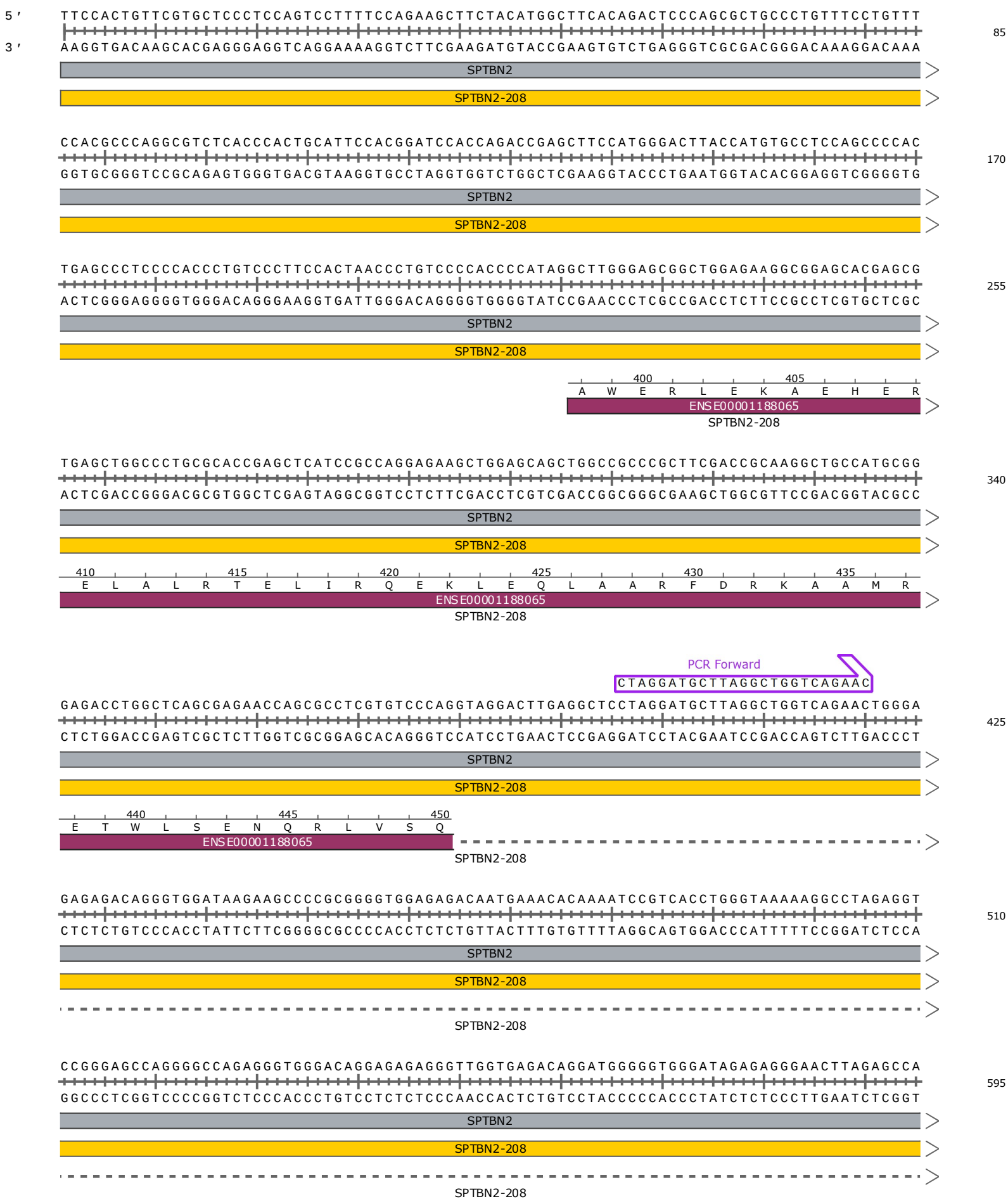
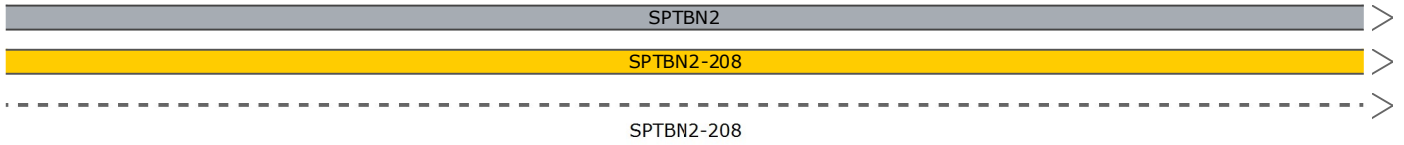


INK2J00076_SPTBN2_R480W_E06.1_AB
1577 bp

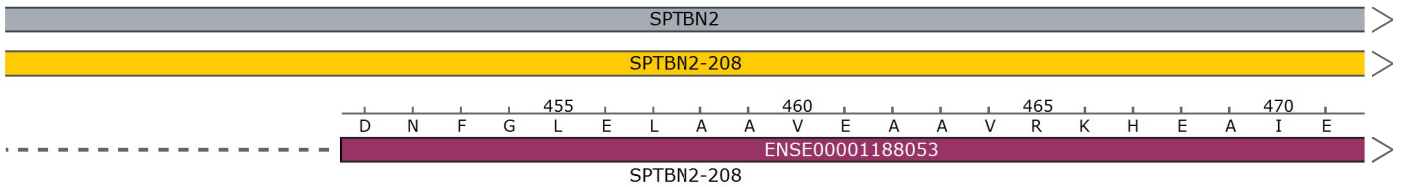


CCGCATGGAGCTGGGATCTTGCAGGCCAAGTGCCTGCAGGACAGAGGAGCAGGCAGAGGCACTGTCCCTTGGTCCCCACACCTC
 GCGGTACCTCGACCCTAGAACGTCCGGTTCACGGGACGTCTGTCTCCTCGTCCGTCTCCGTGACAGGGAACCAGGGGTGTGGAG



Sanger Sequencing Primer
 CAGGACAACCTTTGGGCTGG

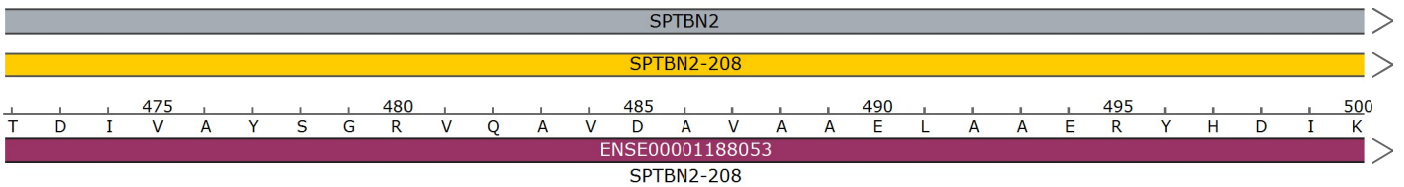
CTCTCCCTGCCCCGACACAGGACAACCTTTGGGCTGGAGCTGGCAGCTGTGAGGCAGCAGTACGGAAGCACGAAGCCATTGAGA
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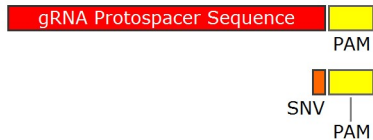
Donor Sequence WT -> SNV
 GCTTCGGTAACTCT
 Donor Template WT -> SNV

gRNA Protospacer
 CATCGTGGCCTACAGCGGCC

CGGACATCGTGGCCTACAGCGGCCGGGTCAGGCAGTGGACGCCGTGGCTGCAGAGCTGGCCGCCGAGCGCTACCACGACATCAA
 GCCTGTAGCACCGGATGTCGCCGCCCCACGTCCGTACCTGCGGCACCGACGTCTCGACCGCGGCTCGCGATGGTGTGTAGTT

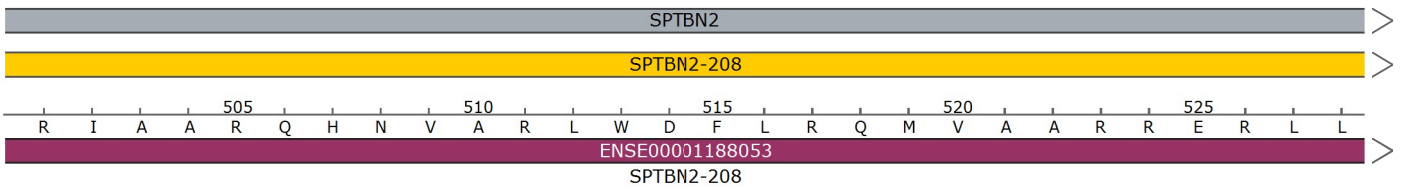


Donor Sequence WT -> SNV



GCCTGTAGCACCGGATGTCGCCGCCCCACGTCCGTACCTGCGGCACCGACGTCTCGACCGCGGCTCGCGATGGTGTGTAGTT
 Donor Template WT -> SNV

GCGCATCGCCGCTCGGCAGCACAACGTGGCAGGGCTCTGGGACTTCTTGCGGCAGATGGTGGCCGCCGGCGGGAGCGGCTCCTC
 CGCGTAGCGGCGAGCCGTGTTGCACCGTCCGAGACCCTGAAGAACGCCGTCTACCACCGCGGGCCGCCCTCGCCGAGGAG



Donor Sequence WT -> SNV

Donor Template WT -> SNV

CTCAACCTGGAGCTGCAGAAGGTGTTCCAGGACCTGCTCTACCTCATGGACTGGATGGAAGAGATGAAGGTACCAGTGAGGCGTG
 GAGTTGGACCTCGACGTCTTCCACAAGGTCTGGACGAGATGGAGTACCTGACCTACCTTCTCTACTTCCATGGTCACTCCGCAC

SPTBN2
 SPTBN2-208

530 535 540 545 550
 L N L E L Q K V F Q D L L Y L M D W M E E M K V P V R R

ENSE00001188053
 SPTBN2-208

CTGGGTGGGGTAAGAGTGATCAAGAGTGCAGGGGGCCCCACAGTGGGTGCGTCCGCCCGTCTGCTCGGCCGATCTCTGTGGAGTG
 GACCCACCCCATTTCTCACTAGTTCTCAGCTCCCCGGGGTGTCAACCACGCAGGCGGGCAGACGAGCCGGCTAGAGACACCTCAC

SPTBN2
 SPTBN2-208

A G W G K S D Q E S R G P H S G C V R P S A R P I S V E C
 (in frame with SPTBN2-208)

TGAACCAGCACAGGGCCCTGTCCCCAGTTGCAGGGAACAAAGTAAACAGGGCCCTGTCCACATGGGGTTTCGTGTCCACATGGAGG
 ACTTGGTTCGTGTCCCGGGACAGGGGTCAACGTCCCTTGTTCATTTGTCCCGGGACAGGTGTACCCCAAGCACAGGTGTACCTCC

SPTBN2
 SPTBN2-208

E P A Q G P V P S C R E Q S K Q G P V H M G F V S T W R
 (in frame with SPTBN2-208)

AGGCTGATGACAAAACCCACGGACCTTCCGTAACAAAAGAGCAGGGGCCATAAAACCACTGATGAAACTGGAACAGGGTGGTG
 TCCGACTACTGTTTTGGGTGCCTGGAAAGGCATTTGTTTTCTCGTCCCGGTATTTGGTGACTACTTTGACCTTGTCACCAC

SPTBN2
 SPTBN2-208

R L M T K P T D L S V N K R A G A I K P L M K L E Q G G
 (in frame with SPTBN2-208)

TGAGGGGCTGGTCAGACAAGGCCTCTGAGAAAATACCATTTAAGCCGAGACCAGAATAGTGCCAGGAGCCAGCACAGAGAGCTCT
 ACTCCCCGACCAGTCTGTTCCGGAGACTCTTTTATGGTAAATTCGGCTCTGGTCTTATCACGGTCTCGGTTCGTCTCTCGAGA

SPTBN2
 SPTBN2-208

V R G W S D K A S E K I P F K P R P E

CTCTTTTATGGTAAATTCGGCTCTG
 PCR Reverse

CAGGAGAGAATGTTCCGGGCAGAAGGAACAGCCGGCACAGGCCCTCGGCCTGGATCAGCATGGTGTGTTTCAGGATCAGAAGGAAG
 GTCCTCTTTACAAGGCCCGTCTTCTTGTGCGCCGTGTCCGGGAGCCGGACCTAGTCGTACCACACAAGTCTAGTCTTCTTCT

SPTBN2
 SPTBN2-208

GCCGGTGGGCTGTGAGCATGAGGAGTGGGGGCCATGGACTGAGATTTGGGAGGGCACGGCCAGCCCACTAGGGCCTTACAGGCCT
 CGGCCACCCGACACTCGTACTCCTCACCCCGGTACCTGACTCTAAACCCTCCCGTGCCGGTTCGGGTGATCCCGGAATGTCCGGA

SPTBN2
 SPTBN2-208

GGTCAGGGGAGTTTGGATTTTATTCAAAGTAACATGGATCCTTTAAAG

3'

|||||

|||||



1577

CCAGTCCCTCAAACCTAAAATAAGTTTCATTGTACCTAGGAAATTC

5'



Feature	Location	Size	Start	End	Type
✓ SPTBN2	1 .. 1577	1577 bp	■	→	gene
/note = gene ENSG00000173898 Protein coding					
C11orf80-216	1 .. 1577	1577 bp	■	←	prim_transcript
/note = primary transcript ENST00000534325 Protein coding					
SPTBN2-201	1 .. 1577	1577 bp	■	→	prim_transcript
/note = primary transcript ENST00000309996					
SPTBN2-203	1 .. 1577	1577 bp	■	→	prim_transcript
/note = primary transcript ENST00000529997					
✓ SPTBN2-208	1 .. 1577	1577 bp	■	→	prim_transcript
/note = primary transcript ENST00000533211					
SPTBN2-209	1 .. 1577	1577 bp	■	→	prim_transcript
/note = primary transcript ENST00000611817					
SPTBN2-210	1 .. 1577	1577 bp	■	→	prim_transcript
/note = primary transcript ENST00000617502					
SPTBN2-211	1 .. 1577	1577 bp	■	→	prim_transcript
/note = primary transcript ENST00000647510 Nonsense mediated decay					
SPTBN2-201	221 .. 1004	784 bp	■	→	CDS
▶ 2 segments = 462 bp					
/note = coding sequence ENSP00000311489					
/translation = AWERLEKAEHERELALRTELIRQEKLEQLAARFDRKAAMRETWLSENQRLVSQ, ,DNFGLELAAVEAAVRKHEAIETDIVAYSGRVQAVDAVAELAERYHDIKRIAARQHN VARLWDFLRQMVAARRERLLLNLLELQKVFQDLLYLMDWMEEMK 154 amino acids = 18.2 kDa					
SPTBN2-203	221 .. 1004	784 bp	■	→	CDS
▶ 2 segments = 462 bp					
/note = coding sequence ENSP00000433593					
/translation = AWERLEKAEHERELALRTELIRQEKLEQLAARFDRKAAMRETWLSENQRLVSQ, ,DNFGLELAAVEAAVRKHEAIETDIVAYSGRVQAVDAVAELAERYHDIKRIAARQHN VARLWDFLRQMVAARRERLLLNLLELQKVFQDLLYLMDWMEEMK 154 amino acids = 18.2 kDa					
✓ SPTBN2-208	221 .. 1004	784 bp	■	→	CDS
▶ 2 segments = 462 bp					
/note = coding sequence ENSP00000432568					
/translation = AWERLEKAEHERELALRTELIRQEKLEQLAARFDRKAAMRETWLSENQRLVSQ, ,DNFGLELAAVEAAVRKHEAIETDIVAYSGRVQAVDAVAELAERYHDIKRIAARQHN VARLWDFLRQMVAARRERLLLNLLELQKVFQDLLYLMDWMEEMK 154 amino acids = 18.2 kDa					
SPTBN2-209	221 .. 1004	784 bp	■	→	CDS
▶ 2 segments = 462 bp					
/note = coding sequence ENSP00000480692					
/translation = AWERLEKAEHERELALRTELIRQEKLEQLAARFDRKAAMRETWLSENQRLVSQ, ,DNFGLELAAVEAAVRKHEAIETDIVAYSGRVQAVDAVAELAERYHDIKRIAARQHN VARLWDFLRQMVAARRERLLLNLLELQKVFQDLLYLMDWMEEMK 154 amino acids = 18.2 kDa					
SPTBN2-210	221 .. 1004	784 bp	■	→	CDS
▶ 2 segments = 462 bp					
/note = coding sequence ENSP00000482000					
/translation = AWERLEKAEHERELALRTELIRQEKLEQLAARFDRKAAMRETWLSENQRLVSQ, ,DNFGLELAAVEAAVRKHEAIETDIVAYSGRVQAVDAVAELAERYHDIKRIAARQHN VARLWDFLRQMVAARRERLLLNLLELQKVFQDLLYLMDWMEEMK 154 amino acids = 18.2 kDa					
✓ Donor Sequence WT -> SNV	752 .. 851	100 bp	■		misc_feature
✓ gRNA Protospacer Sequence	770 .. 789	20 bp	■		misc_feature
✓ SNV	789 .. 789	1 bp	■		misc_feature
/note = WT = A SNV = T					
✓ PAM	790 .. 792	3 bp	■		misc_feature
✓ PAM	790 .. 792	3 bp	■		misc_feature

Feature	Location	Size		Type
C11orf80	38,248 .. 45,932,203 bp	15,794 bp		gene
/note	= gene ENSG00000173715 Protein coding			

Primer	Length	Binding Sites	Tm	Date Added
✓ PCR Forward /sequence = CTAGGATGCTTAGGCTGGTCAGAAC 52% GC / 7722.1 Da	25-mer	396 .. 420 →	60°C	Aug 18, 2023
✓ Sanger Sequencing Primer /sequence = CAGGACAACCTTTGGGCTGG 58% GC / 5868.9 Da	19-mer	699 .. 717 →	57°C	Aug 18, 2023
✓ Donor Template WT -> SNV /sequence = CTTGATGTCGTGGTAGCGCTCGGCGGCCAGCTCTGCAGCCACGGCTCCACTGCCTGCACCCAGCCGCTGTAGGCCACGATGCCGTCTCAATGGCTTCG 66% GC / 30,684.8 Da	100-mer	752 .. 851 ←	84°C	Aug 18, 2023
✓ gRNA Protospacer /sequence = CATCGTGGCCTACAGCGGCC 70% GC / 6079.0 Da	20-mer	770 .. 789 →	64°C	Aug 18, 2023
✓ PCR Reverse /sequence = GTCTCGGCTTAAATGGTATTTTCTC 40% GC / 7629.0 Da	25-mer	1302 .. 1326 ←	56°C	Aug 18, 2023