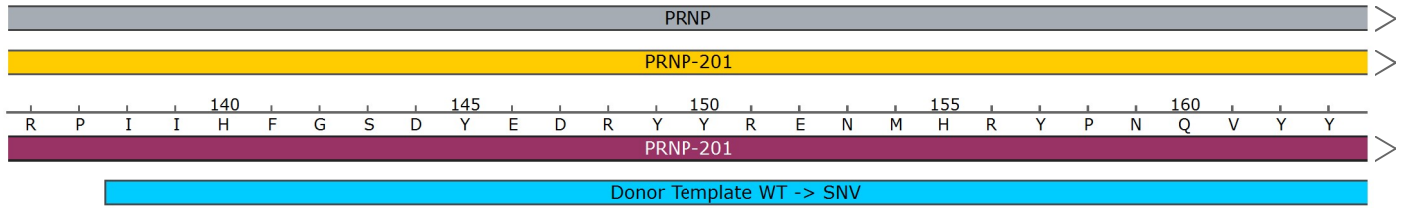


Donor Template WT -> SNV

ATCATAACATTTTCGGCAGTGACTATGAGGACCGTTACTATCGTGAAAACATGCACCGTTACCCCAACCAAGTGTACTACA
AGGCCCATCATAACATTTTCGGCAGTGACTATGAGGACCGTTACTATCGTGAAAACATGCACCGTTACCCCAACCAAGTGTACTACA
TCCGGGTAGTATGTAAGCCGTCACCTGATACTCCTGGCAATGATAGCACTTTTGTACGTGGCAATGGGGTTGGTTTCACATGATGT

680



gRNA Protospacer
PAM

SNV

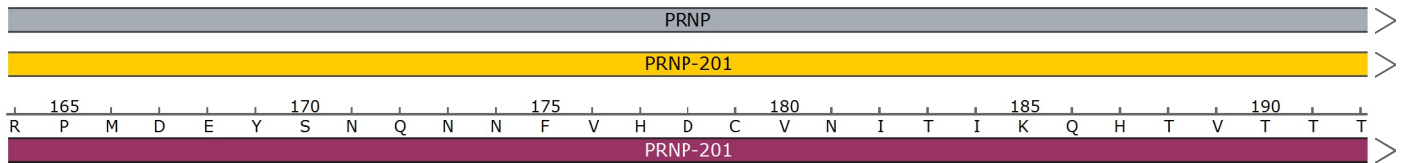
GTTGGTTTCACATGATGT
gRNA Protospacer

Donor Template WT -> SNV

GGCCCATGGATGAGTACAGCA

GGCCCATGGATGAGTACAGCAACCAGAACAACCTTTGTGCACGACTGCGTCAATATCACAATCAAGCAGCACACGGTCAACCACAAC
CCGGGTACCTACTCATGTCGTTGGTCTTGTGAAACACGTGCTGACGCAGTTATAGTGTAGTTTCGTCGTGTGCCAGTGGTGTGG

765



Donor Template WT -> SNV

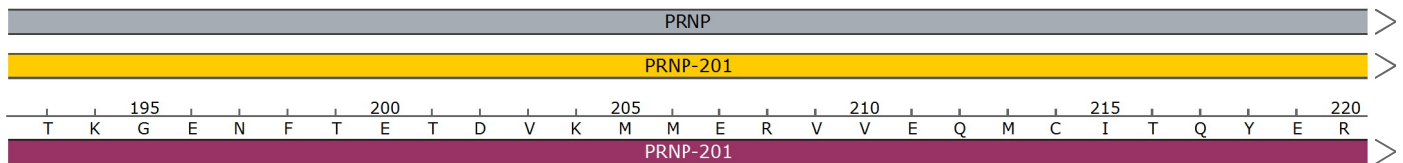
gRNA Protospacer

CCG

gRNA Protospacer

CACCAAGGGGGGAGAAGTTCACCGAGACCGACGTTAAGATGATGGAGCGCGTGGTTGAGCAGATGTGTATCACCCAGTACGAGAGG
GTGGTCCCCCTCTTGAAGTGGCTCTGGCTGCAATTCTACTACCTCGCGCACCAACTCGTCTACACATAGTGGGTTCATGCTCTCC

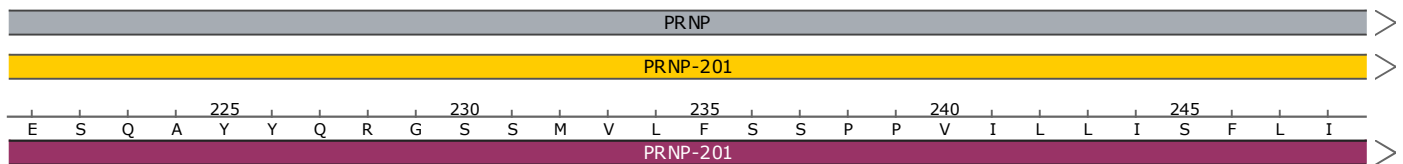
850



CAATTCTACTACCTCGCGCA
Sanger Sequencing Primer

GAATCTCAGGCCTATTACCAGAGAGGATCGAGCATGGTCTCTTCTCTCTCCACCTGTGATCCTCCTGATCTCTTTCCTCATCT
CTTAGAGTCCGGATAATGGTCTCTCTCTAGCTCGTACCAGGAGAAGAGGAGAGGTGGACACTAGGAGGACTAGAGAAAGGAGTAGA

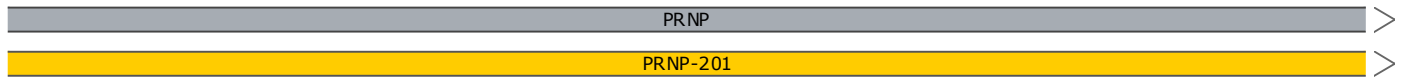
935



E S Q A Y Y Q R G S S M V L F S S P P V I L L I S F L I

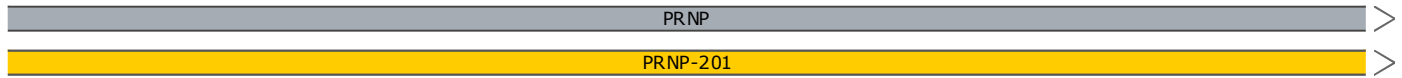
TCCTGATAGTGGGATGAGGAAGGTCTTCCTGTTTTACCACATCTTTCTAATCTTTTTCCAGCTTGAGGGAGGCGGTATCCACCTGC
 AGGACTATCACCTACTCCTTCCAGAAGGACAAAAGTGGTAGAAAAGATTAGAAAAAGGTCGAACTCCCTCCGCCATAGGTGGACG

1020



AGCCCTTTTAGTGGTGGTGTCTCACTCTTTCTTCTCTTTGTCCCGGATAGGCTAATCAATACCCTTGGCACTGATGGGCACTG
 TCGGGAAAATCACCAACACAGAGTGAGAAAGAAGAGAGAAACAGGGCCTATCCGATTAGTTATGGGAACCGTGACTACCCGTGAC

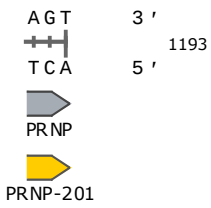
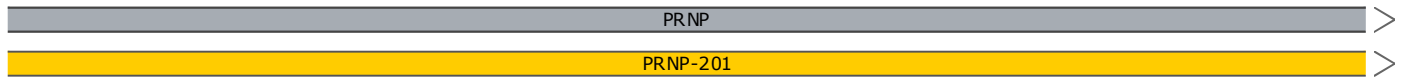
1105



PCR Reverse
 CCCCTTTGATTGAGTTCATCATGAG

GAAAACATAGAGTAGACCTGAGATGCTGGTCAAGCCCCCTTTGATTGAGTTCATCATGAGCCGTTGCTAATGCCAGGCCAGTAAA
 CTTTTGTATCTCATCTGGACTCTACGACCAGTTCGGGGGAAACTAAGTCAAGTAGTACTCGGCAACGATTACGGTCCGGTCATTT

1190



| Feature | Location | Size | Start | End | Type |
|--------------------------------------|---|---------|-------|-----|-----------------|
| ✓ PRNP | 1 .. 1193 | 1193 bp | ■ | ➔ | gene |
| /note | = gene ENSG00000171867 Protein coding | | | | |
| ✓ PRNP-201 | 1 .. 1193 | 1193 bp | ■ | ➔ | prim_transcript |
| /note | = primary transcript ENST00000379440 | | | | |
| PRNP-202 | 1 .. 1193 | 1193 bp | ■ | ➔ | prim_transcript |
| /note | = primary transcript ENST00000424424 | | | | |
| PRNP-203 | 1 .. 1193 | 1193 bp | ■ | ➔ | prim_transcript |
| /note | = primary transcript ENST00000430350 | | | | |
| PRNP-204 | 1 .. 1193 | 1193 bp | ■ | ➔ | prim_transcript |
| /note | = primary transcript ENST00000457586 | | | | |
| ✓ PRNP-201 | 191 .. 952 | 762 bp | ■ | ➔ | CDS |
| /note | = coding sequence ENSP00000368752 | | | | |
| /translation | = MANLGCWMLVLFVATWSDLGLCKKRPKPGGWNTGGSRYPGQGSPGGNRYPPQGGGGWGQPHGGGWGQPHGGGWGQPHGGGWGQPHGGGWGQGGGTHSQWNKPSKPKTNMKHMA GAAAAGAVVGGGLGGYMLGSAMSRPIIHFGSDYEDRYRENMHRYPNQVYYRPMDEYSNQNNFVHDCVNITIKQHTVTTTTKGENFTETDVKMMERVVEQMCITQYERESQAYY QRGSSMVLFSPPVILLISFLIFLIVG* 253 amino acids = 27.7 kDa | | | | |
| PRNP-202 | 191 .. 952 | 762 bp | ■ | ➔ | CDS |
| /note | = coding sequence ENSP00000411599 | | | | |
| /translation | = MANLGCWMLVLFVATWSDLGLCKKRPKPGGWNTGGSRYPGQGSPGGNRYPPQGGGGWGQPHGGGWGQPHGGGWGQPHGGGWGQPHGGGWGQGGGTHSQWNKPSKPKTNMKHMA GAAAAGAVVGGGLGGYMLGSAMSRPIIHFGSDYEDRYRENMHRYPNQVYYRPMDEYSNQNNFVHDCVNITIKQHTVTTTTKGENFTETDVKMMERVVEQMCITQYERESQAYY QRGSSMVLFSPPVILLISFLIFLIVG* 253 amino acids = 27.7 kDa | | | | |
| PRNP-203 | 191 .. 952 | 762 bp | ■ | ➔ | CDS |
| /note | = coding sequence ENSP00000399376 | | | | |
| /translation | = MANLGCWMLVLFVATWSDLGLCKKRPKPGGWNTGGSRYPGQGSPGGNRYPPQGGGGWGQPHGGGWGQPHGGGWGQPHGGGWGQPHGGGWGQGGGTHSQWNKPSKPKTNMKHMA GAAAAGAVVGGGLGGYMLGSAMSRPIIHFGSDYEDRYRENMHRYPNQVYYRPMDEYSNQNNFVHDCVNITIKQHTVTTTTKGENFTETDVKMMERVVEQMCITQYERESQAYY QRGSSMVLFSPPVILLISFLIFLIVG* 253 amino acids = 27.7 kDa | | | | |
| PRNP-204 | 191 .. 952 | 762 bp | ■ | ➔ | CDS |
| /note | = coding sequence ENSP00000415284 | | | | |
| /translation | = MANLGCWMLVLFVATWSDLGLCKKRPKPGGWNTGGSRYPGQGSPGGNRYPPQGGGGWGQPHGGGWGQPHGGGWGQPHGGGWGQPHGGGWGQGGGTHSQWNKPSKPKTNMKHMA GAAAAGAVVGGGLGGYMLGSAMSRPIIHFGSDYEDRYRENMHRYPNQVYYRPMDEYSNQNNFVHDCVNITIKQHTVTTTTKGENFTETDVKMMERVVEQMCITQYERESQAYY QRGSSMVLFSPPVILLISFLIFLIVG* 253 amino acids = 27.7 kDa | | | | |
| ✓ Donor Template WT -> SNV | 602 .. 701 | 100 bp | ■ | | misc_feature |
| ✓ PAM | 661 .. 663 | 3 bp | ■ | | misc_feature |
| ✓ gRNA Protospacer | 664 .. 683 | 20 bp | ■ | | misc_feature |
| ✓ SNV | 668 .. 668 | 1 bp | ■ | | misc_feature |
| /note | = WT = C SNV = T | | | | |

| Primer | Length | Binding Sites | Tm | Date Added |
|---|---------|---------------|------|--------------|
| ✓ PCR Forward | 25-mer | 142 .. 166 | 58°C | Jun 14, 2023 |
| /sequence = TAAATATGGGACTCTGACGTTCTCC 44% GC / 7632.0 Da | | | | |
| ✓ Donor Template WT -> SNV | 100-mer | 602 .. 701 | 75°C | Jun 14, 2023 |
| /sequence = ATCATACATTTTCGGCAGTGACTATGAGGACCGTTACTATCGTGAAAACATGCACCGTTACCCCAACTAAGTGTACTACAGGCCCATGGATGAGTACAGCA 46% GC / 30,787.1 Da | | | | |
| ✓ gRNA Protospacer | 20-mer | 664 .. 683 | 54°C | Jun 14, 2023 |
| /sequence = GCCTGTAGTACACTTGGTTG 50% GC / 6139.1 Da | | | | |
| ✓ Sanger Sequencing Primer | 20-mer | 797 .. 816 | 57°C | Jun 14, 2023 |
| /sequence = ACGCGCTCCATCATCTTAAC 50% GC / 5997.0 Da | | | | |
| ✓ PCR Reverse | 25-mer | 1141 .. 1165 | 58°C | Jun 14, 2023 |
| /sequence = CCCCTTTGATTGAGTTCATCATGAG 44% GC / 7623.0 Da | | | | |