

***Gramd4* Cas9-KO Strategy**

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Project Overview

Project Name

Gramd4

Project type

Cas9-KO

Strain background

C57BL/6JGpt

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Gramd4* gene. The schematic diagram is as follows:



- The *Gramd4* gene has 5 transcripts. According to the structure of *Gramd4* gene, exon3 of *Gramd4*-201(ENSMUST00000088931.9) transcript is recommended as the knockout region. The region contains 121bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Gramd4* gene. The brief process is as follows: CRISPR/Cas9 system were microinjected into the fertilized eggs of C57BL/6JGpt mice. Fertilized eggs were transplanted to obtain positive F0 mice which were confirmed by PCR and sequencing. A stable F1 generation mouse model was obtained by mating positive F0 generation mice with C57BL/6JGpt mice.

- The *Gramd4* gene is located on the Chr15. If the knockout mice are crossed with other mice strains to obtain double gene positive homozygous mouse offspring, please avoid the two genes on the same chromosome.
- Transcript *Gramd4*-205 may not be affected.
- This strategy is designed based on genetic information in existing databases. Due to the complexity of biological processes, all risk of the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)

Gramd4 GRAM domain containing 4 [Mus musculus (house mouse)]

Gene ID: 223752, updated on 13-Mar-2020

Summary



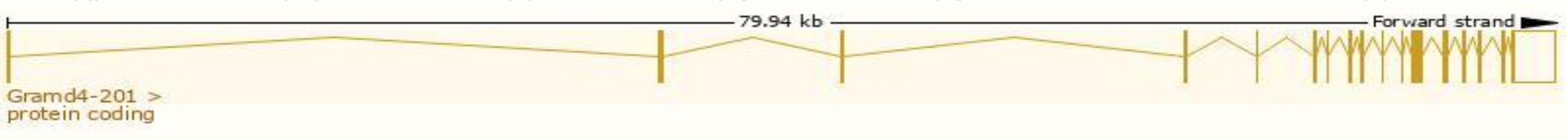
Official Symbol	Gramd4 provided by MGI
Official Full Name	GRAM domain containing 4 provided by MGI
Primary source	MGI:MGI:2676308
See related	Ensembl:ENSMUSG00000035900
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	9930016O13
Expression	Ubiquitous expression in whole brain E14.5 (RPKM 10.5), CNS E18 (RPKM 10.3) and 27 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

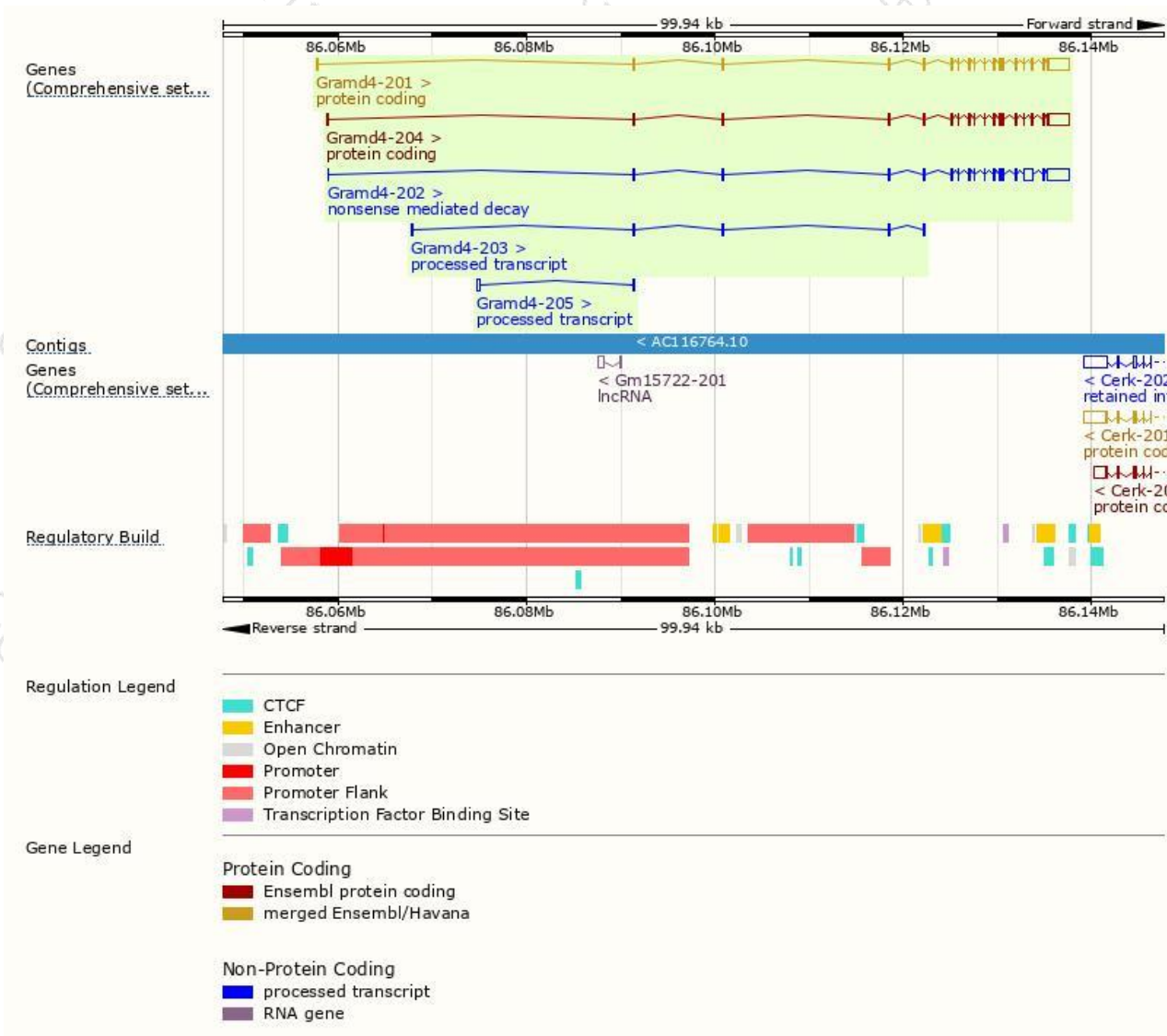
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Gramd4-204	ENSMUST00000138134.7	4292	627aa	Protein coding	CCDS56998	D3YUE7	TSL:1 GENCODE basic
Gramd4-201	ENSMUST00000088931.9	4115	633aa	Protein coding	CCDS27727	Q8CB44	TSL:1 GENCODE basic APPRIS P1
Gramd4-202	ENSMUST00000123349.1	4861	323aa	Nonsense mediated decay	-	D6RET7	TSL:2
Gramd4-203	ENSMUST00000123474.1	597	No protein	Processed transcript	-	-	TSL:5
Gramd4-205	ENSMUST00000147286.1	374	No protein	Processed transcript	-	-	TSL:2

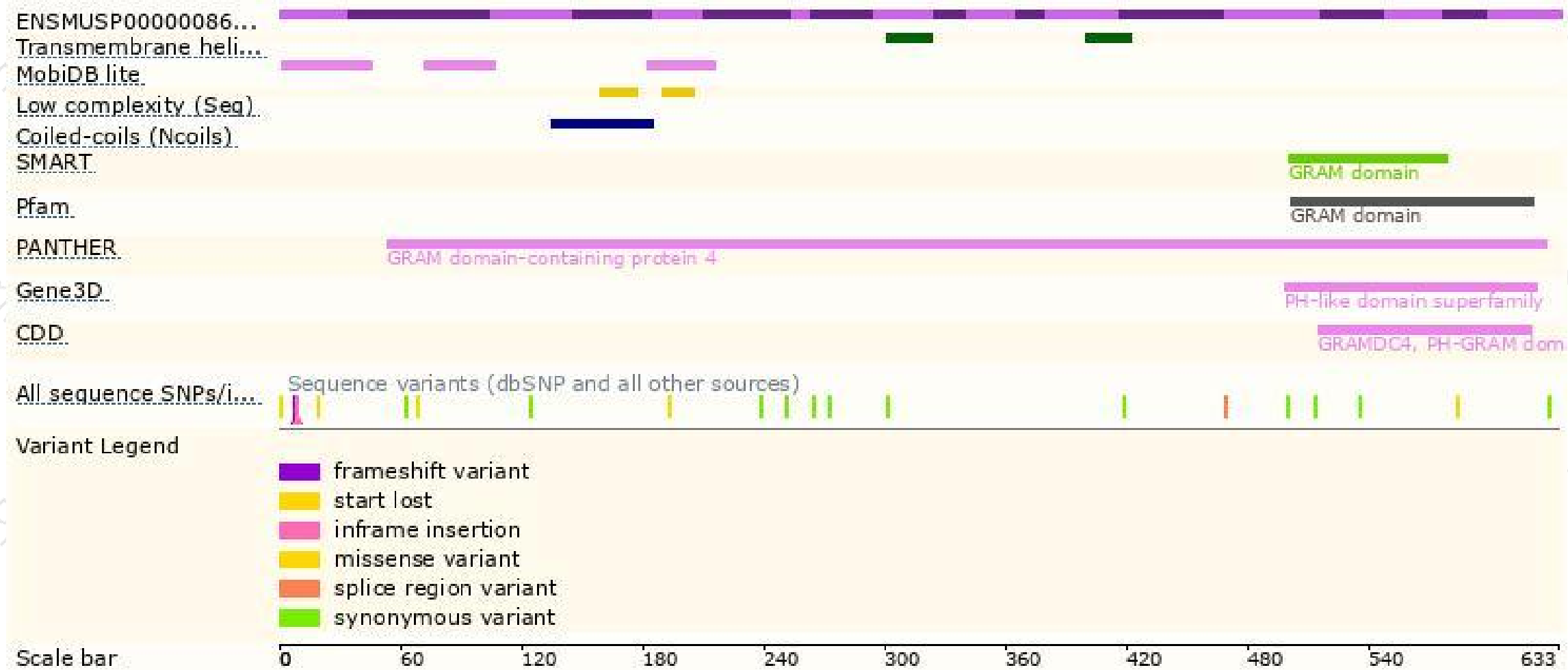
The strategy is based on the design of *Gramd4-201* transcript,the transcription is shown below:



Genomic location distribution



Protein domain



If you have any questions, you are welcome to inquire.

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