

***Gramd1c* Cas9-CKO Strategy**

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Project Overview

Project Name

Gramd1c

Project type

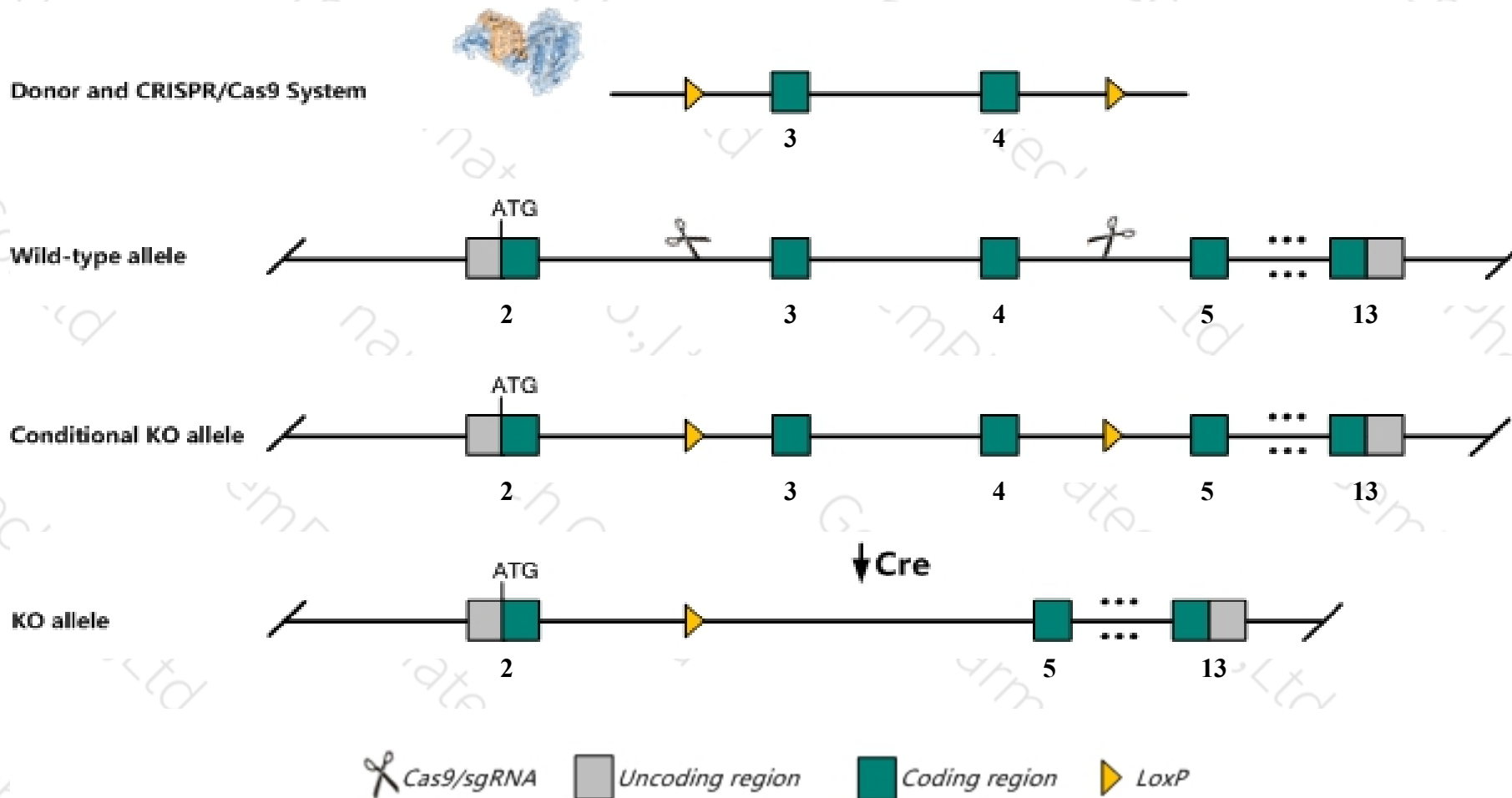
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Gramd1c* gene. The schematic diagram is as follows:



- The *Gramd1c* gene has 6 transcripts. According to the structure of *Gramd1c* gene, exon3-exon4 of *Gramd1c-201*(ENSMUST00000036174.9) transcript is recommended as the knockout region. The region contains 296bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Gramd1c* gene. The brief process is as follows: sgRNA was transcribed in vitro, donor vector was constructed. Cas9, sgRNA and Donor 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 flox mice was knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues and cell types.

- The *Gramd1c* gene is located on the Chr16. 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.
- This strategy is designed based on genetic information in existing databases. Due to the complexity of biological processes, all risk of loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information (NCBI)

Gramd1c GRAM domain containing 1C [Mus musculus (house mouse)]

Gene ID: 207798, updated on 13-Mar-2020

Summary



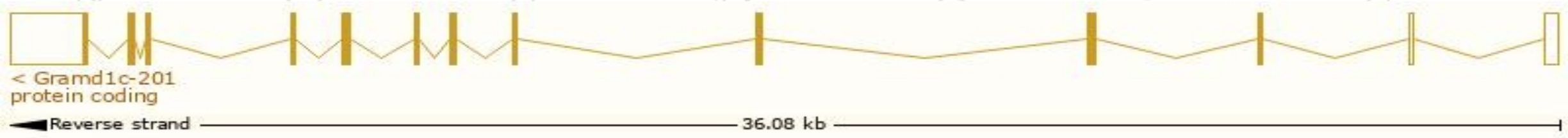
Official Symbol	Gramd1c provided by MGI
Official Full Name	GRAM domain containing 1C provided by MGI
Primary source	MGI:MGI:2443024
See related	Ensembl:ENSMUSG00000036292
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	4921521N14Rik
Expression	Broad expression in testis adult (RPKM 5.0), liver E18 (RPKM 1.8) and 18 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

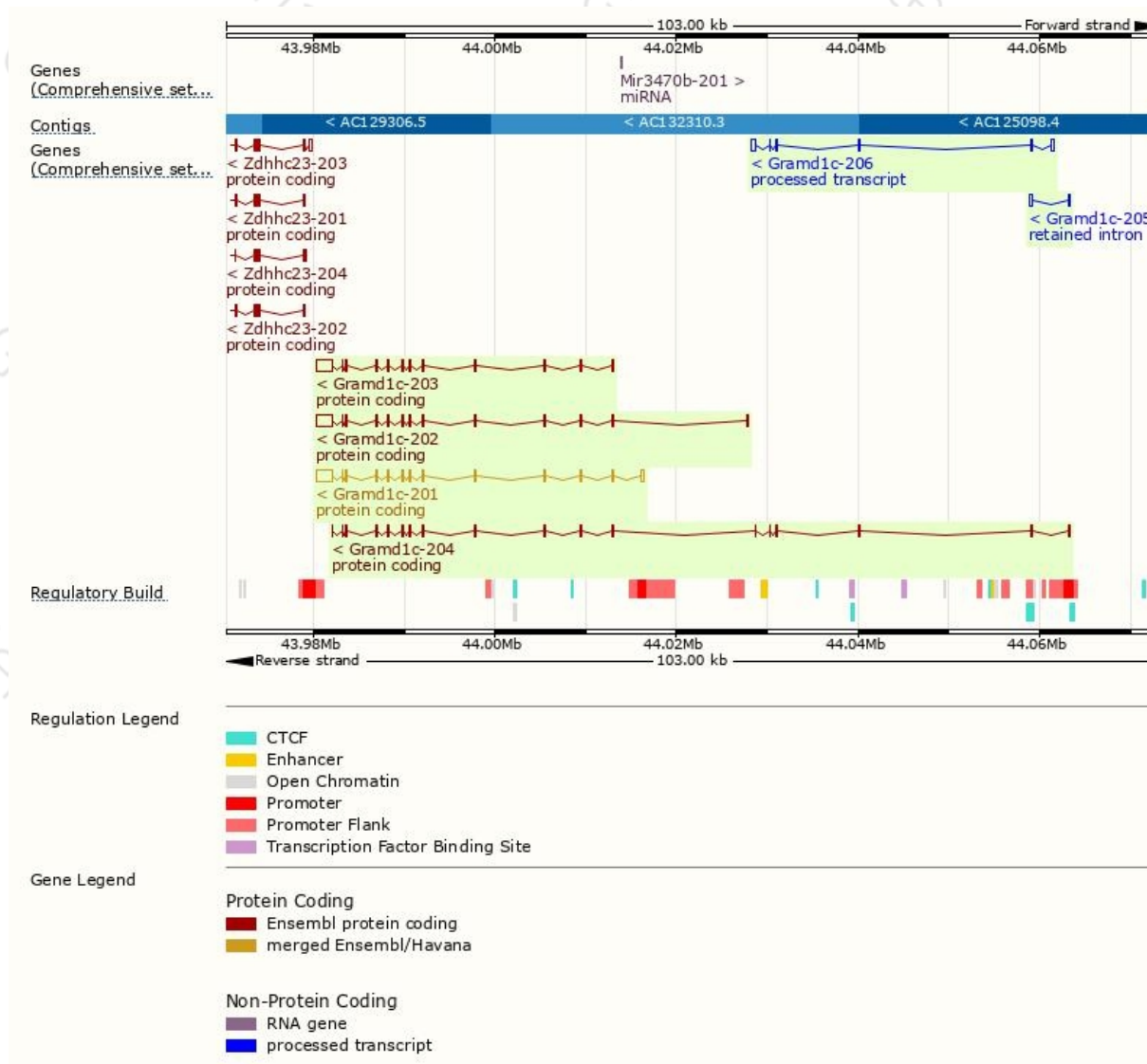
The gene has 6 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Gramd1c-201	ENSMUST00000036174.9	3501	457aa	Protein coding	CCDS28181	A0A3P9AQ15	TSL:1 GENCODE basic
Gramd1c-202	ENSMUST00000114677.1	3271	457aa	Protein coding	CCDS28181	A0A3P9AQ15	TSL:1 GENCODE basic
Gramd1c-203	ENSMUST00000179565.7	3159	457aa	Protein coding	CCDS28181	A0A3P9AQ15	TSL:5 GENCODE basic
Gramd1c-204	ENSMUST00000214098.1	2046	662aa	Protein coding	-	Q8CI52	TSL:5 GENCODE basic APPRIS P1
Gramd1c-206	ENSMUST00000216149.1	1144	No protein	Processed transcript	-	-	TSL:5
Gramd1c-205	ENSMUST00000214764.1	374	No protein	Retained intron	-	-	TSL:3

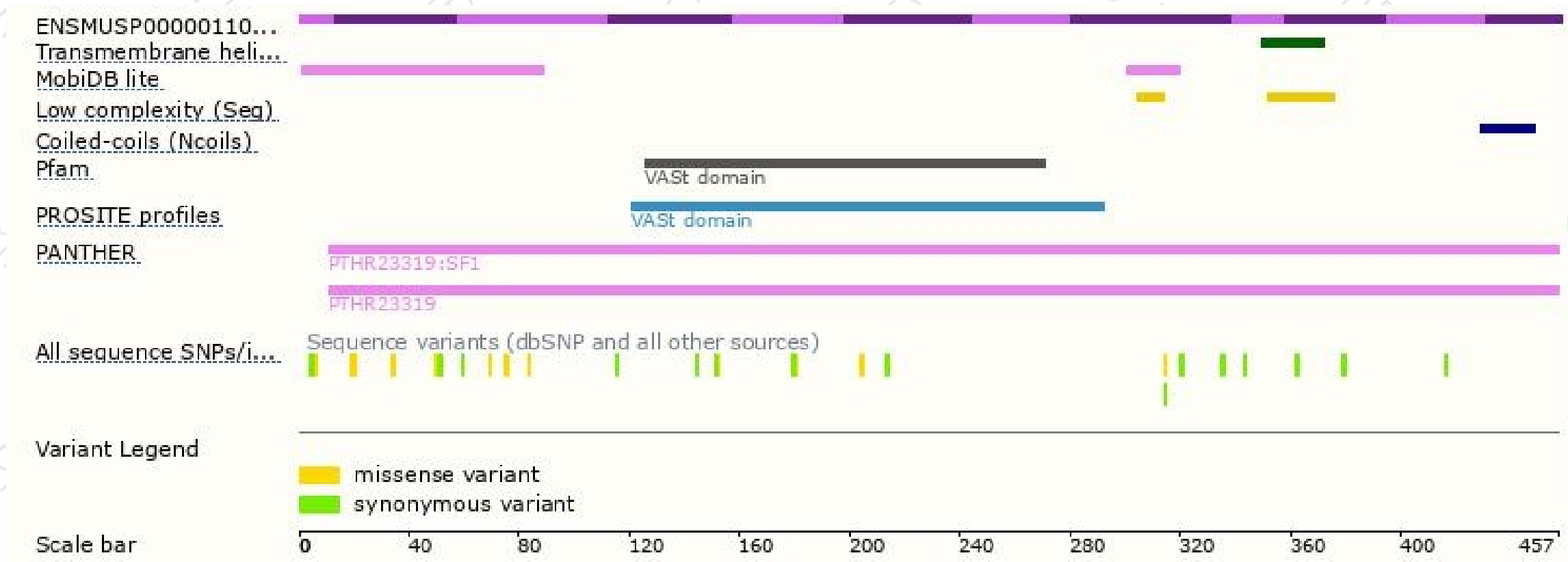
The strategy is based on the design of *Gramd1c-201* transcript,the transcription is shown below:



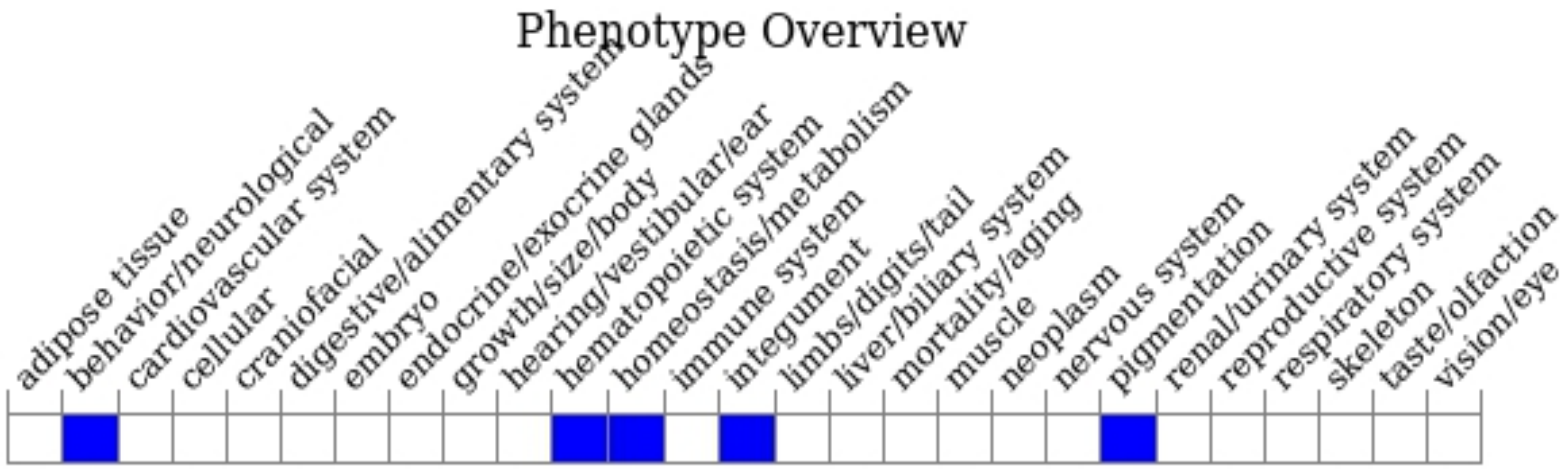
Genomic location distribution



Protein domain



Mouse phenotype description(MGI)



Phenotypes affected by the gene are marked in blue. Data quoted from MGI database(<http://www.informatics.jax.org/>).

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

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