

Gprc5b Cas9-CKO Strategy

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Design Date: 2019-8-5

Project Overview

Project Name

Gprc5b

Project type

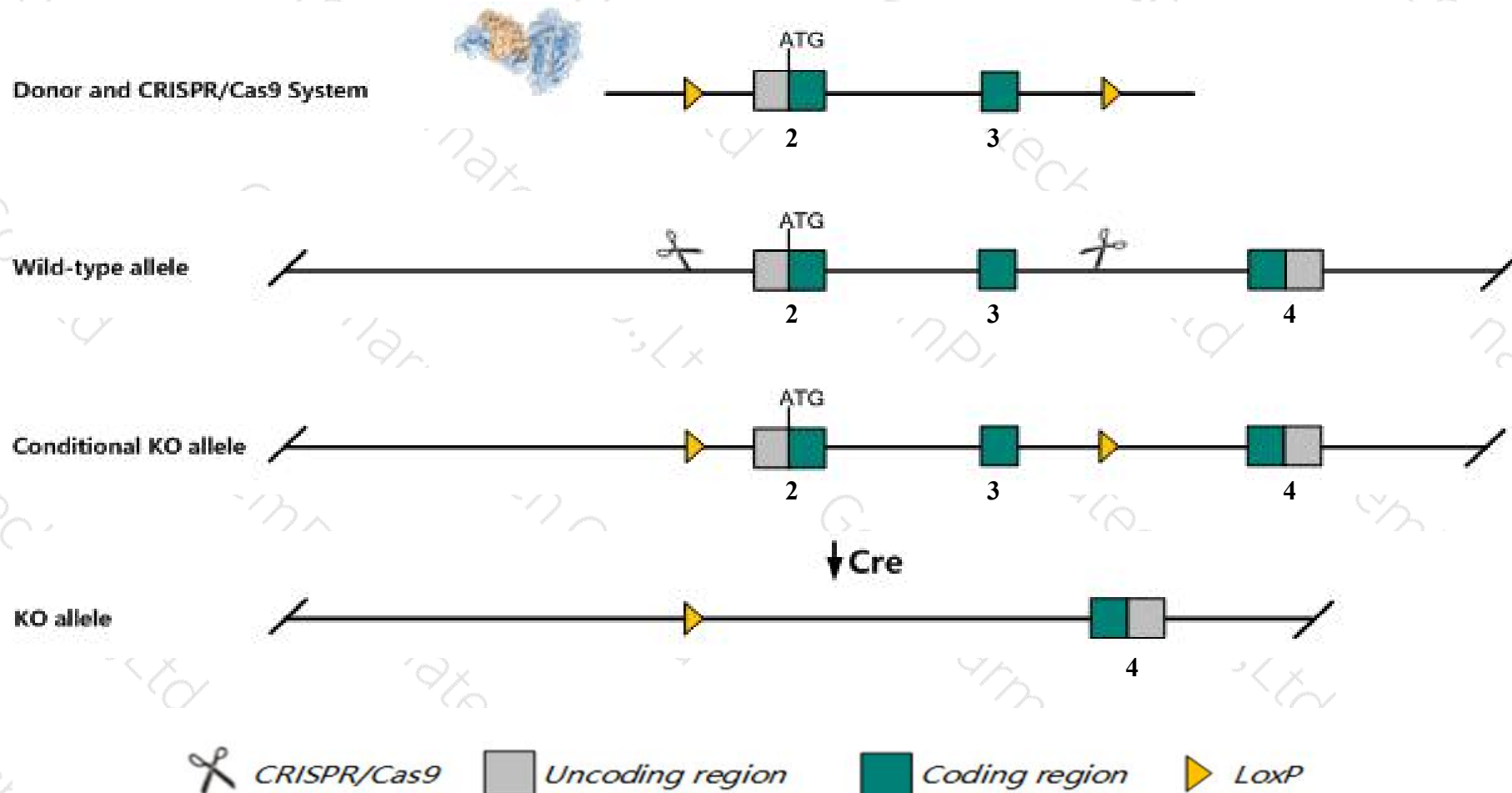
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Gprc5b* gene. The schematic diagram is as follows:



Technical routes

- The *Gprc5b* gene has 4 transcripts. According to the structure of *Gprc5b* gene, exon2-exon3 of *Gprc5b-201* (ENSMUST00000008878.9) transcript is recommended as the knockout region. The region contains start codon ATG. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Gprc5b* gene. The brief process is as follows: CRISPR/Cas9 system 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 will be 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.

- According to the existing MGI data, Mice homozygous for a reporter allele exhibit partial neonatal and postnatal lethality, altered spontaneous activity pattern, and decreased response to a new environment.
- Transcript *Gprc5b*-202 may not be affected.
- The floxed region is near to the N-terminal of *Gm44763* gene, this strategy may influence the regulatory function of the N-terminal of *Gm44763* gene.
- The *Gprc5b* gene is located on the Chr7. 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)

Gprc5b G protein-coupled receptor, family C, group 5, member B [Mus musculus (house mouse)]

Gene ID: 64297, updated on 31-Jan-2019

Summary



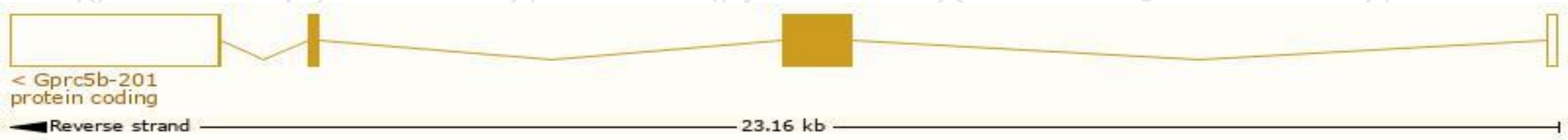
Official Symbol	Gprc5b provided by MGI
Official Full Name	G protein-coupled receptor, family C, group 5, member B provided by MGI
Primary source	MGI:MGI:1927596
See related	Ensembl:ENSMUSG000000008734
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AW125761, Raig2
Expression	Broad expression in cerebellum adult (RPKM 63.2), frontal lobe adult (RPKM 44.6) and 17 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

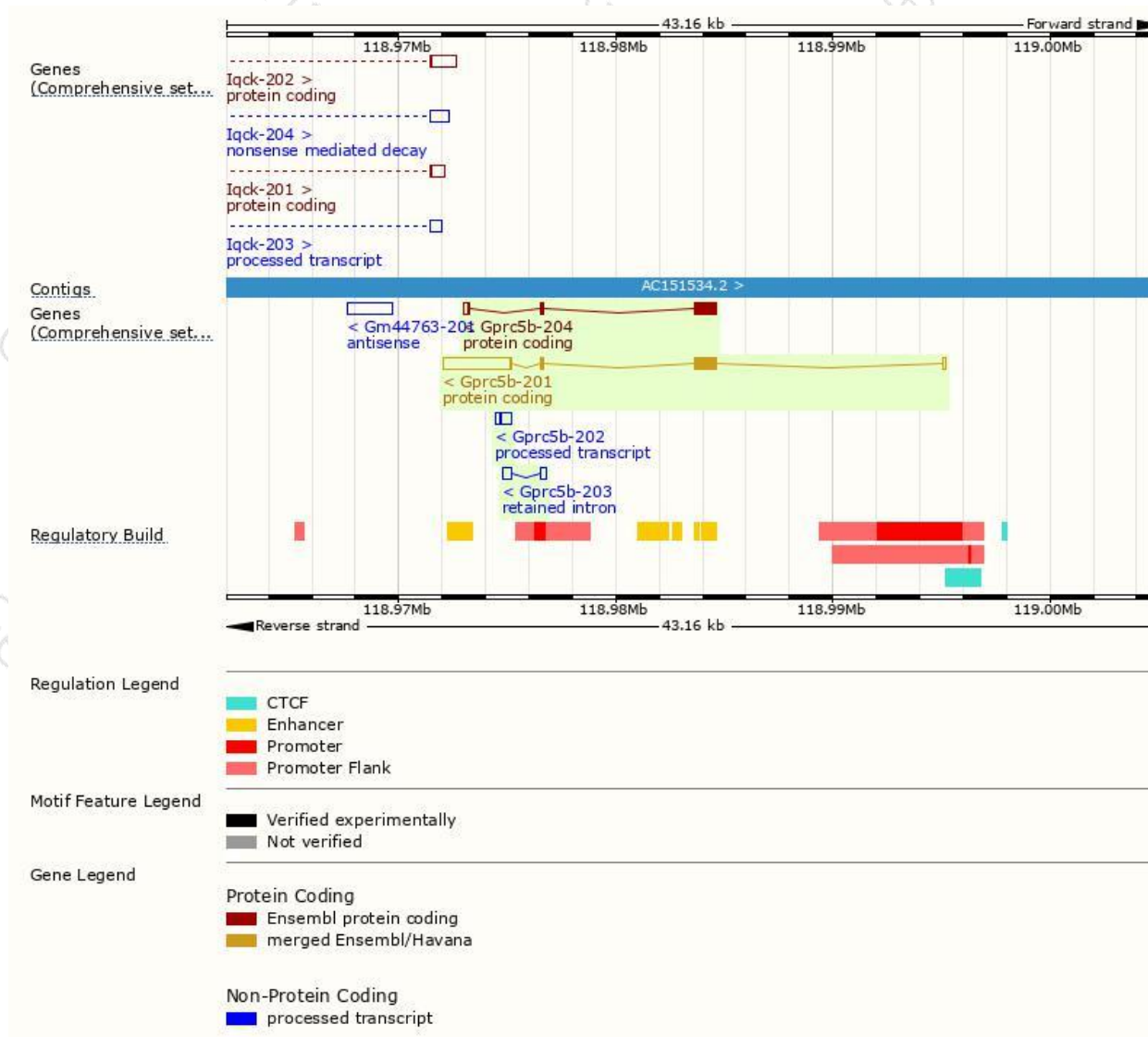
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Gprc5b-201	ENSMUST00000008878.9	4518	410aa	Protein coding	CCDS21777	Q923Z0	TSL:1 GENCODE basic APPRIS P3
Gprc5b-204	ENSMUST00000208394.1	1416	411aa	Protein coding	CCDS85400	D2DFA9	TSL:1 GENCODE basic APPRIS ALT2
Gprc5b-202	ENSMUST00000207582.1	692	No protein	Processed transcript	-	-	TSL:5
Gprc5b-203	ENSMUST00000207614.1	686	No protein	Retained intron	-	-	TSL:2

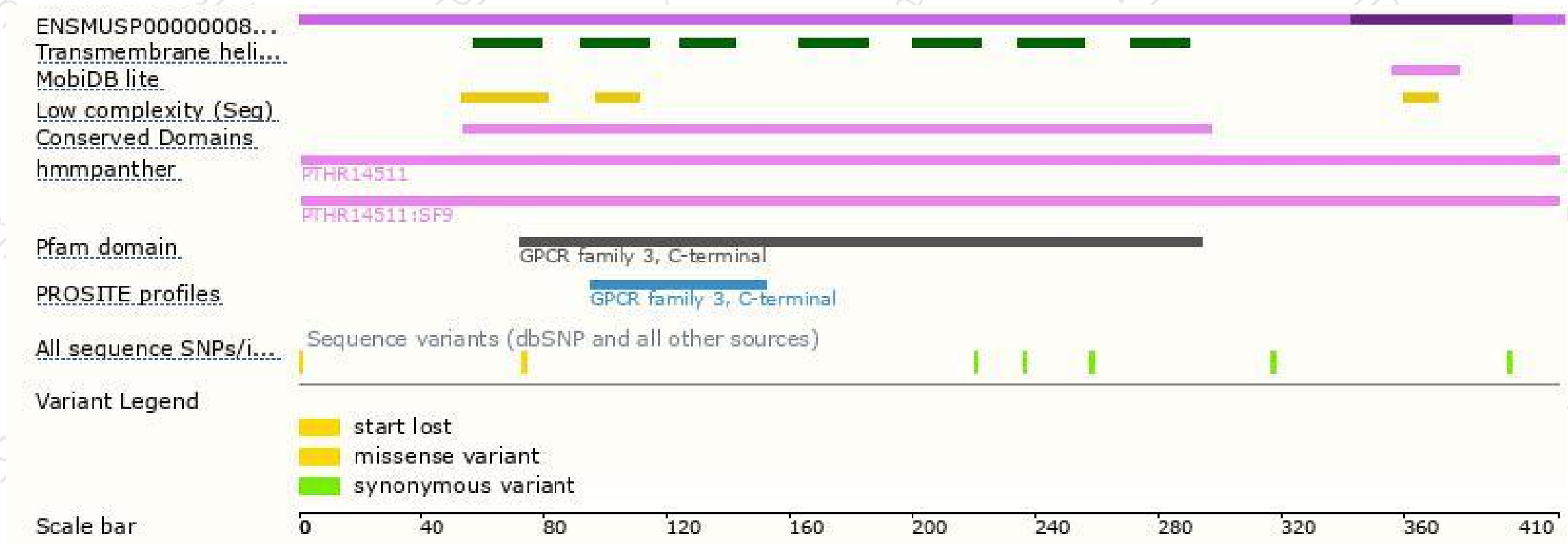
The strategy is based on the design of *Gprc5b-201* transcript,The transcription is shown below



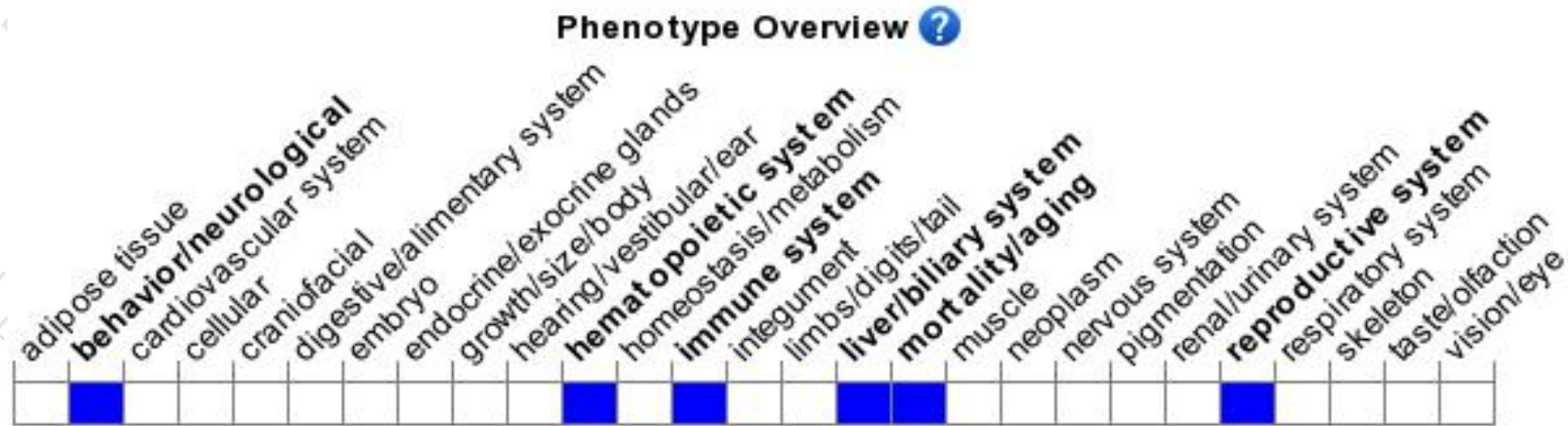
Genomic location distribution



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/>).

According to the existing MGI data, Mice homozygous for a reporter allele exhibit partial neonatal and postnatal lethality, altered spontaneous activity pattern, and decreased response to a new environment.

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

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