Cacna1i Cas9-CKO Strategy

Designer: JiaYu

Reviewer: Xiaojing Li

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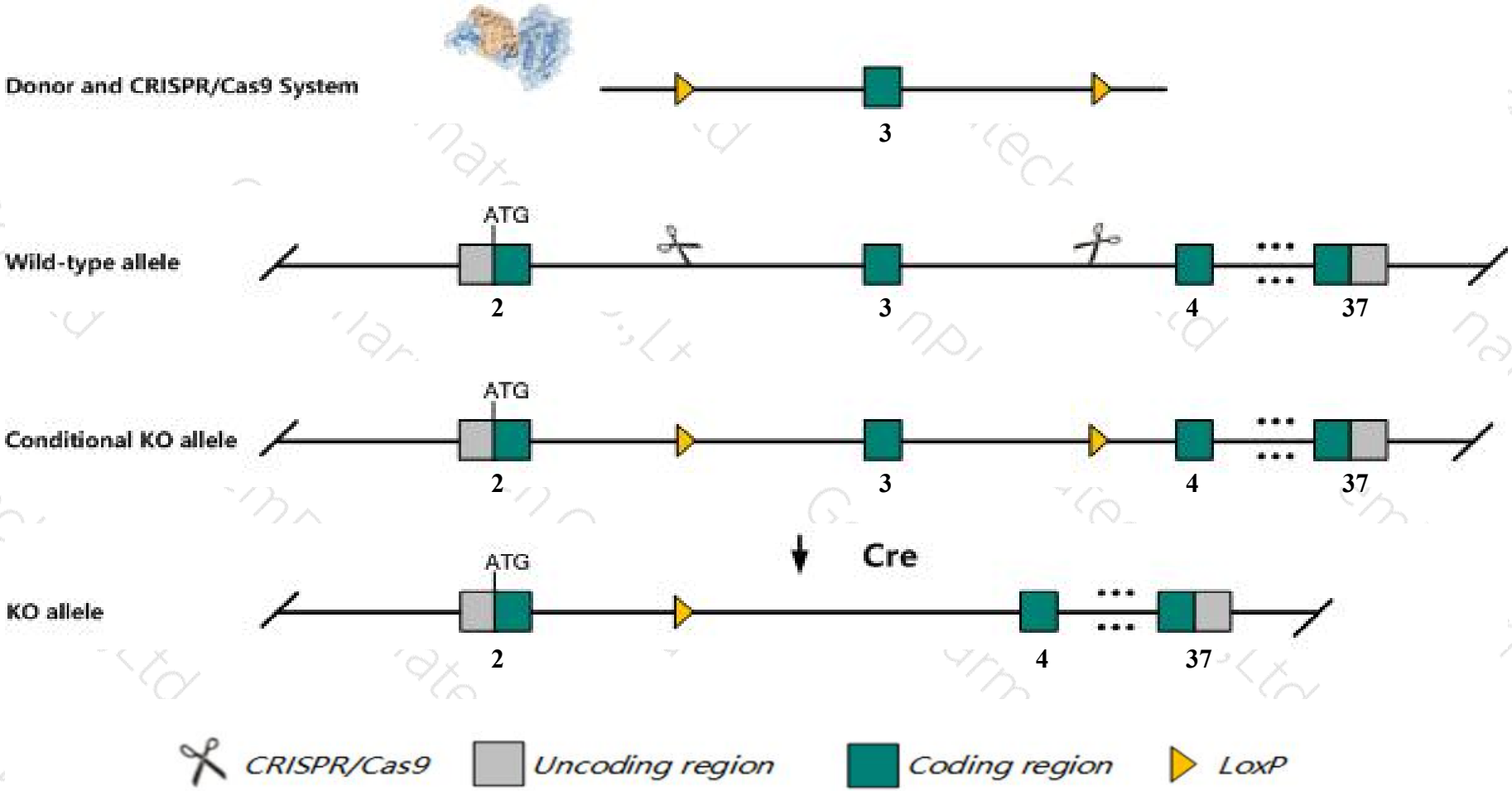
Project Overview

Project Name	***Cacna1i***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Cacna1i* gene. The schematic diagram is as follows:



Technical routes

- The *Cacna1i* gene has 6 transcripts. According to the structure of *Cacna1i* gene, exon3 of *Cacna1i-202* (ENSMUST00000160424.7) transcript is recommended as the knockout region.The region contains 112bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Cacna1i* 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.

Notice

➢ The *Cacna1i* 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 201,203,204 CDS 5' and 3' incomplete the influences is unknown.

➢ 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）

集萃药康 GemPharmatech

Cacna1i calcium channel, voltage-dependent, alpha 1I subunit [Mus musculus (house mouse)]

Gene ID: 239556, updated on 31-Jan-2019

Summary

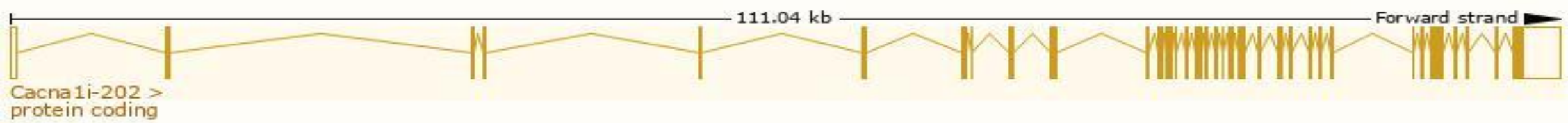
Official Symbol	Cacna1i provided by MGI
Official Full Name	calcium channel, voltage-dependent, alpha 1I subunit provided by MGI
Primary source	MGI:MGI:2178051
See related	Ensembl:ENSMUSG00000022416
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Ca(V)3.3
Expression	Biased expression in spleen adult (RPKM 8.3), frontal lobe adult (RPKM 8.1) and 8 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

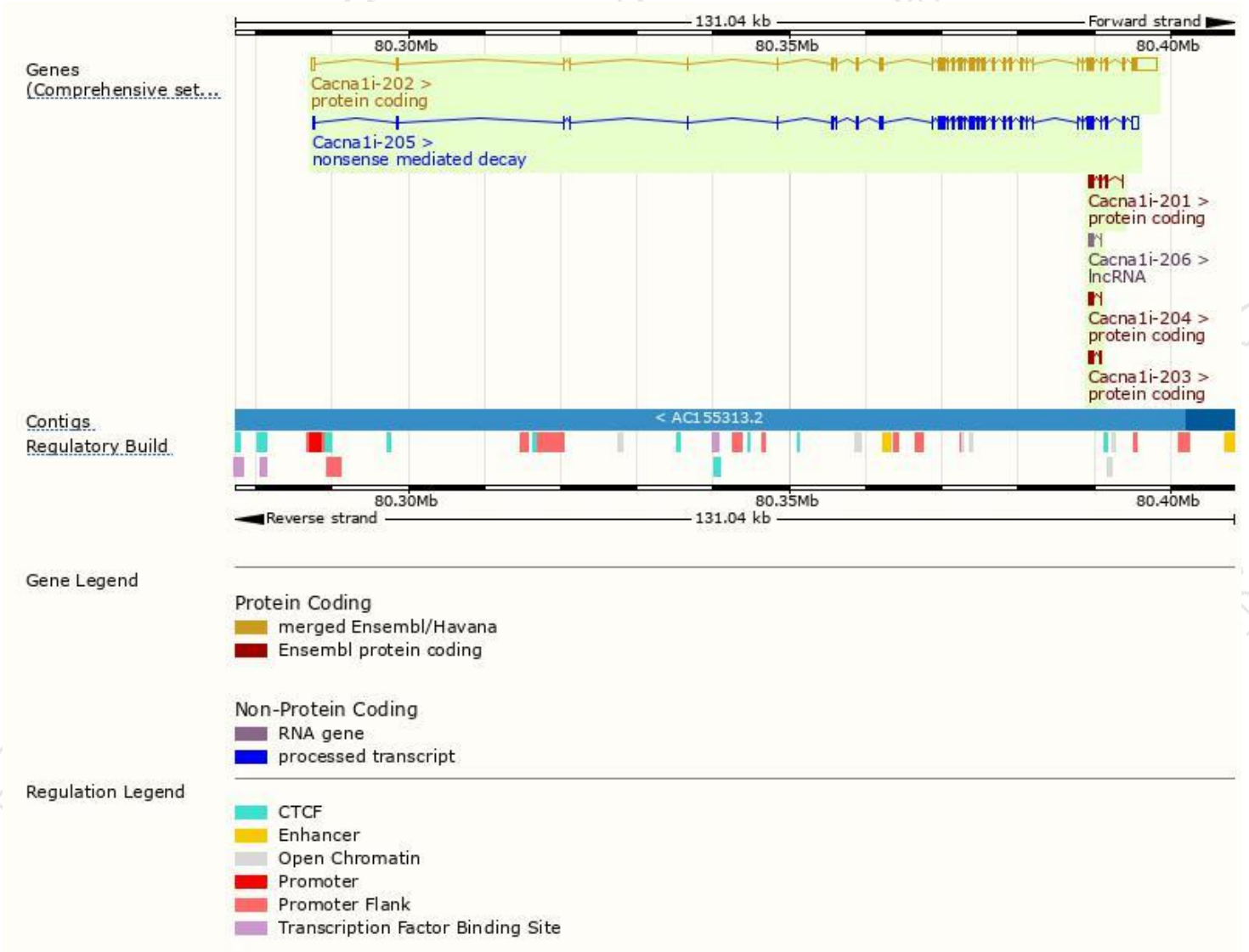
The gene has 6 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Cacna1i-202	ENSMUST00000160424.7	9781	2199aa	Protein coding	CCDS49670	E9Q7P2	TSL:1 GENCODE basic APPRIS P1
Cacna1i-201	ENSMUST00000160175.1	779	260aa	Protein coding	-	F6V0N5	CDS 5' and 3' incomplete TSL:5
Cacna1i-203	ENSMUST00000161863.7	394	132aa	Protein coding	-	F6TAX9	CDS 5' and 3' incomplete TSL:5
Cacna1i-204	ENSMUST00000162025.7	313	105aa	Protein coding	-	F7BQ85	CDS 5' and 3' incomplete TSL:5
Cacna1i-205	ENSMUST00000162155.7	6913	1835aa	Nonsense mediated decay	-	E0CXK2	TSL:5
Cacna1i-206	ENSMUST00000162913.7	273	No protein	Processed transcript	-	-	TSL:5

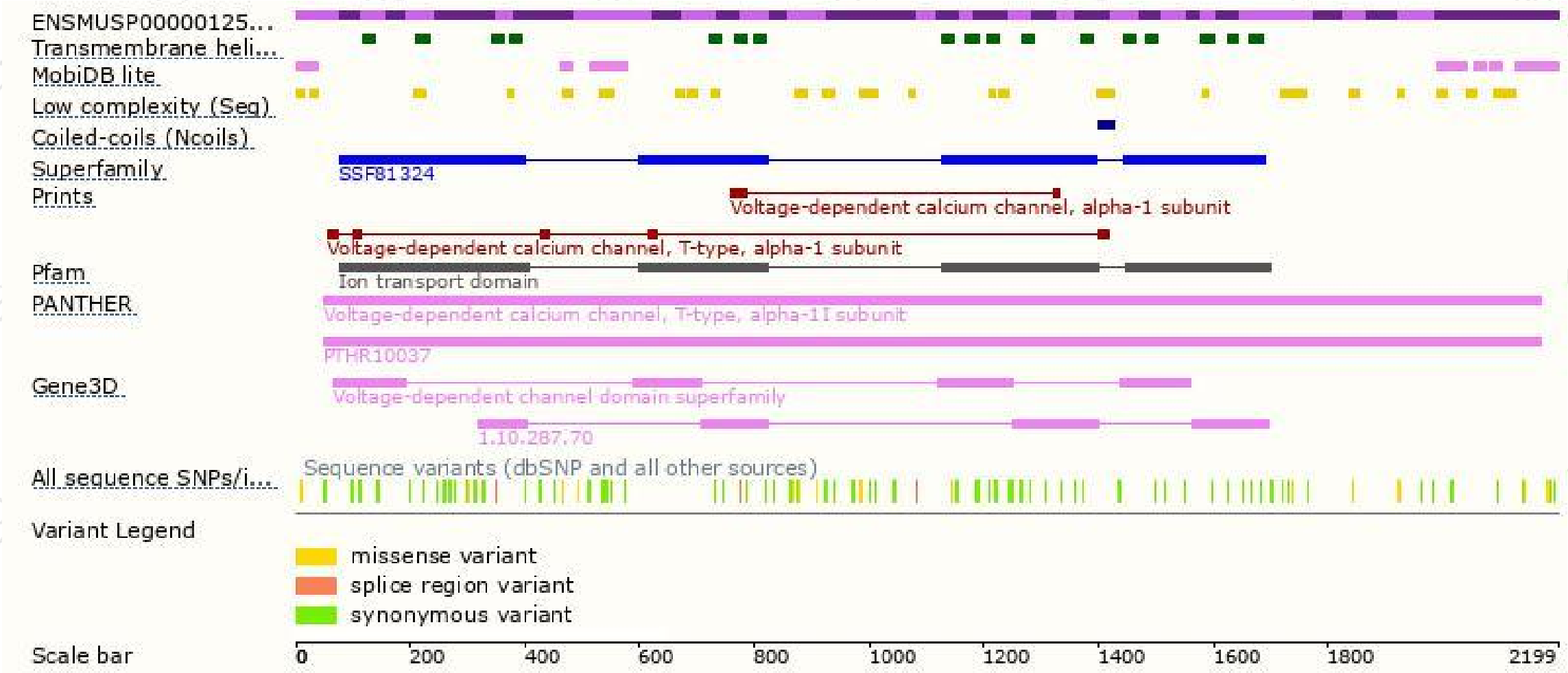
The strategy is based on the design of *Cacna1i-202* transcript,The transcription is shown below



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/).

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

Tel: 400-9660890

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GemPharmatech Co.,Ltd