Camk2g Cas9-CKO Strategy

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Project Overview

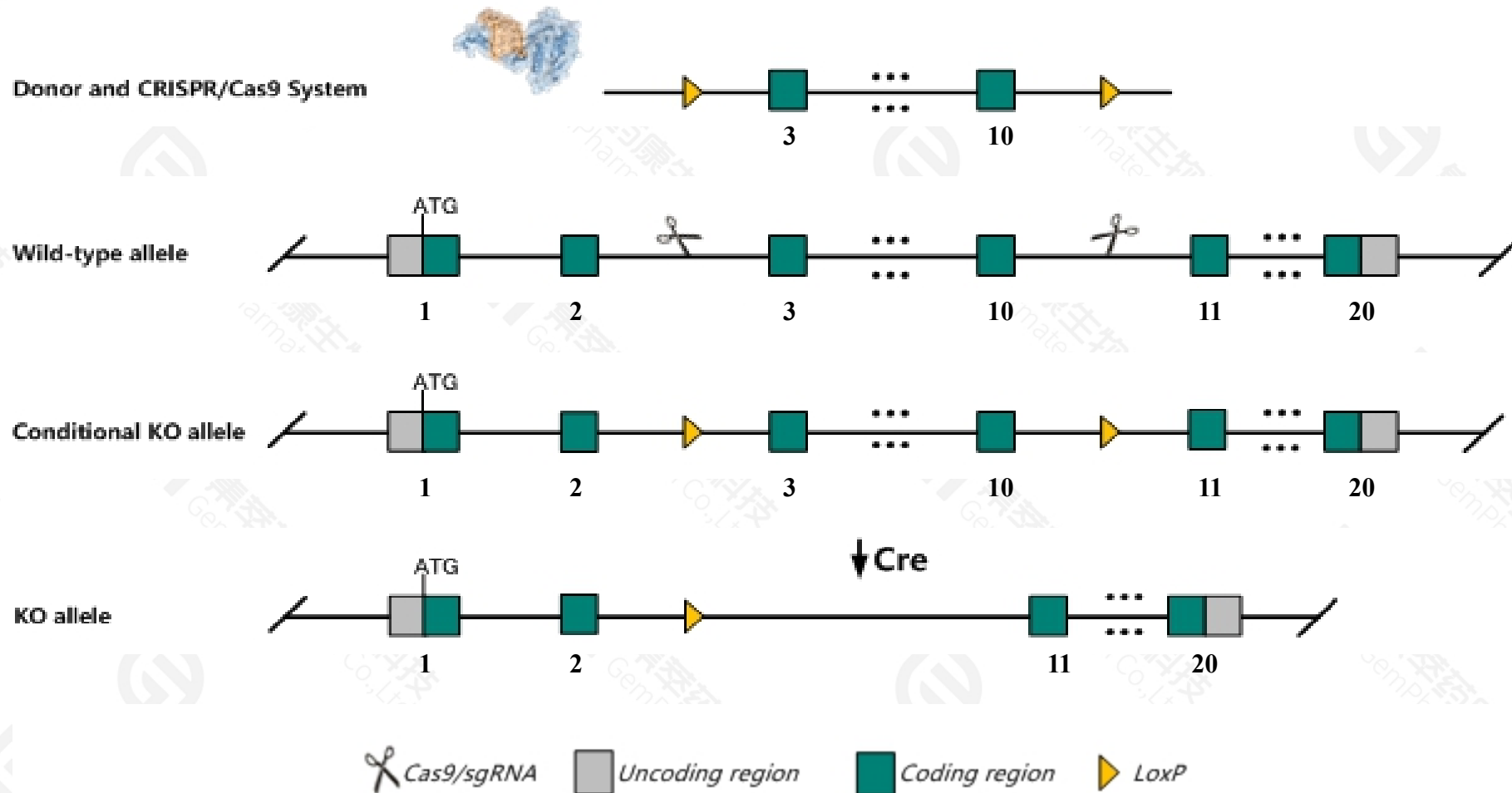|	
Project Name	***Camk2g***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Camk2g* gene. The schematic diagram is as follows:

Technical routes

➢ The *Camk2g* gene has 17 transcripts. According to the structure of *Camk2g* gene, exon3-exon10 of *Camk2g-201*(ENSMUST00000071816.7) transcript is recommended as the knockout region.The region contains 659bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Camk2g* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor 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.

Notice

➢ According to the existing MGI data,mice homozygous for a knock-out allele exhibit female infertility and decreased sensitivity of macrophages to ER stress-induced apoptosis.

➢ The *Camk2g* gene is located on the Chr14. 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）

Camk2g calcium/calmodulin-dependent protein kinase II gamma [Mus musculus (house mouse)]

Gene ID: 12325, updated on 13-Mar-2020

▲ Summary

|	
Official Symbol	Camk2g provided by MGI
Official Full Name	calcium/calmodulin-dependent protein kinase II gamma provided by MGI
Primary source	MGI:MGI:88259
See related	Ensembl:ENSMUSG00000021820
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	Camkg
Expression	Broad expression in cortex adult (RPKM 49.3), cerebellum adult (RPKM 39.7) and 26 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

The gene has 17 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Camk2g-201	ENSMUST00000071816.6	3703	529aa	Protein coding	CCDS26857	Q923T9	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P4
Camk2g-202	ENSMUST00000080440.13	3604	518aa	Protein coding	CCDS26855	Q923T9	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS ALT1
Camk2g-203	ENSMUST00000100837.10	3564	495aa	Protein coding	CCDS26856	Q6ZWS7 Q923T9	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS ALT1
Camk2g-217	ENSMUST00000226630.1	1937	565aa	Protein coding	-	A0A2I3BQP6	GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS ALT1
Camk2g-209	ENSMUST00000224887.1	822	274aa	Protein coding	-	A0A286YCW8	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete
Camk2g-204	ENSMUST00000223712.1	722	241aa	Protein coding	-	A0A286YDL6	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete
Camk2g-212	ENSMUST00000225609.1	673	224aa	Protein coding	-	A0A286YCB8	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete
Camk2g-210	ENSMUST00000225328.1	570	190aa	Protein coding	-	A0A286YCH4	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete
Camk2g-211	ENSMUST00000225463.1	503	168aa	Protein coding	-	A0A286YDK9	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete
Camk2g-205	ENSMUST00000223863.1	663	71aa	Nonsense mediated decay	-	A0A286YCI7	CDS 5' incomplete
Camk2g-207	ENSMUST00000224566.1	551	No protein	Processed transcript	-	-	
Camk2g-215	ENSMUST00000225800.1	535	No protein	Processed transcript	-	-	
Camk2g-208	ENSMUST00000224804.1	452	No protein	Processed transcript	-	-	
Camk2g-214	ENSMUST00000225660.1	2130	No protein	Retained intron	-	-	
Camk2g-216	ENSMUST00000225958.1	2106	No protein	Retained intron	-	-	
Camk2g-213	ENSMUST00000225635.1	456	No protein	Retained intron	-	-	
Camk2g-206	ENSMUST00000224245.1	343	No protein	Retained intron	-	-	

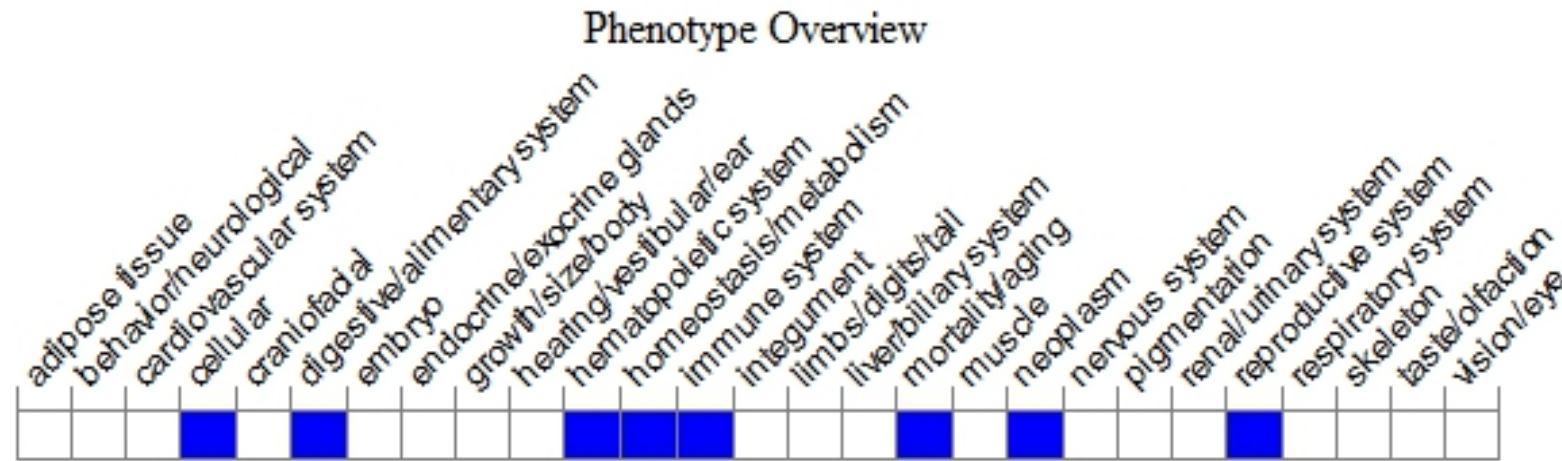The strategy is based on the design of *Camk2g-201* transcript,the transcription is shown below:

< Camk2g-201
protein coding

Reverse strand

59.21 kb
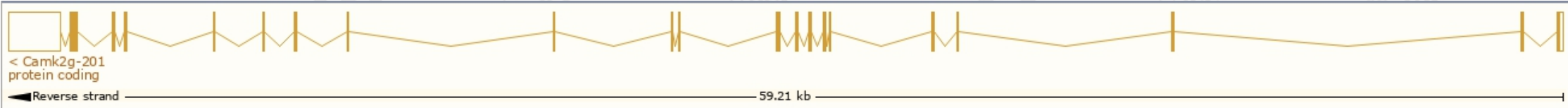
Genomic location distribution
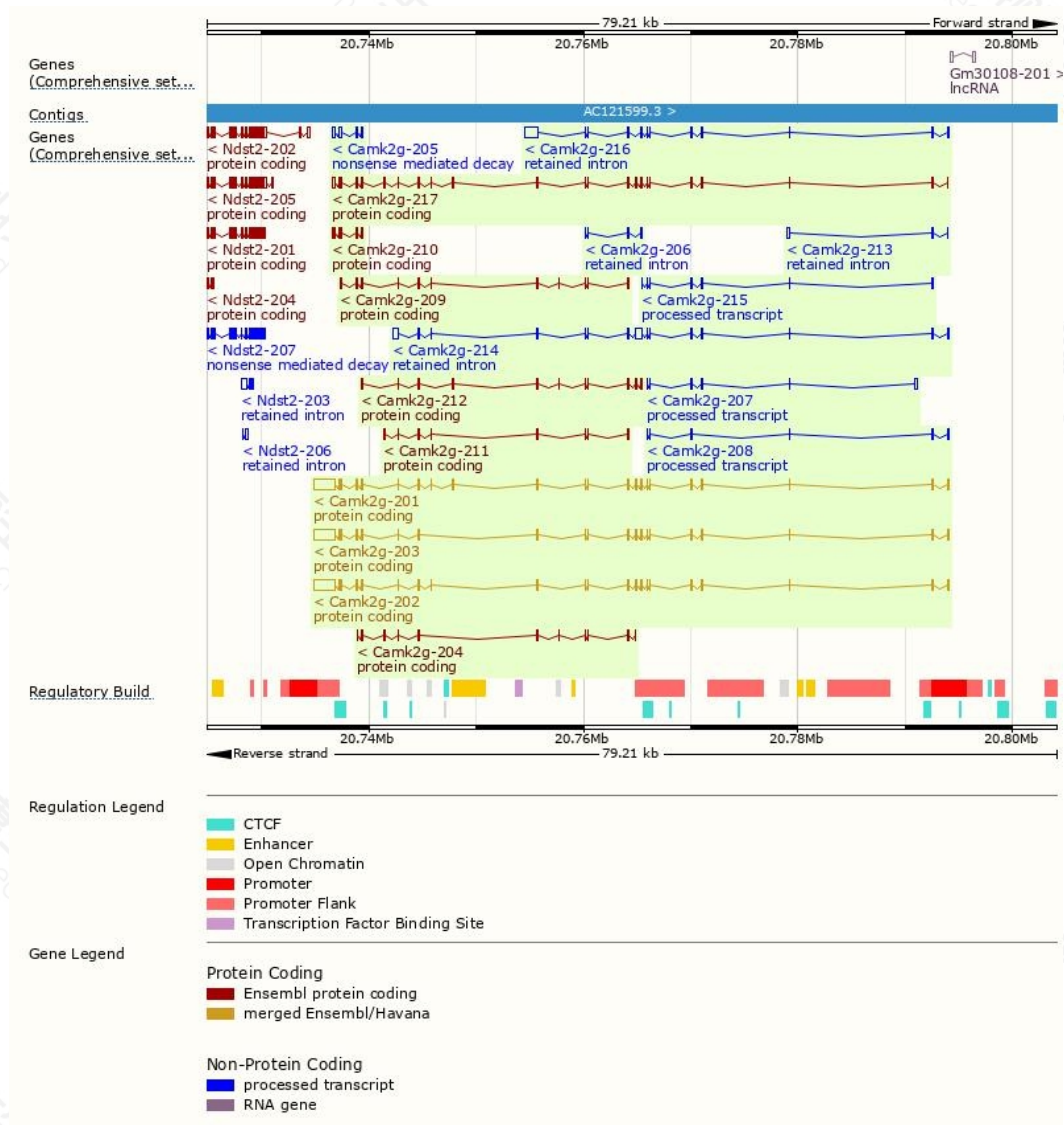
Protein domain

Mouse phenotype description(MGI)

Phenotype Overview

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 knock-out allele exhibit female infertility and decreased sensitivity of macrophages to ER stress-induced apoptosis.

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