# *Camk2d* Cas9-CKO Strategy

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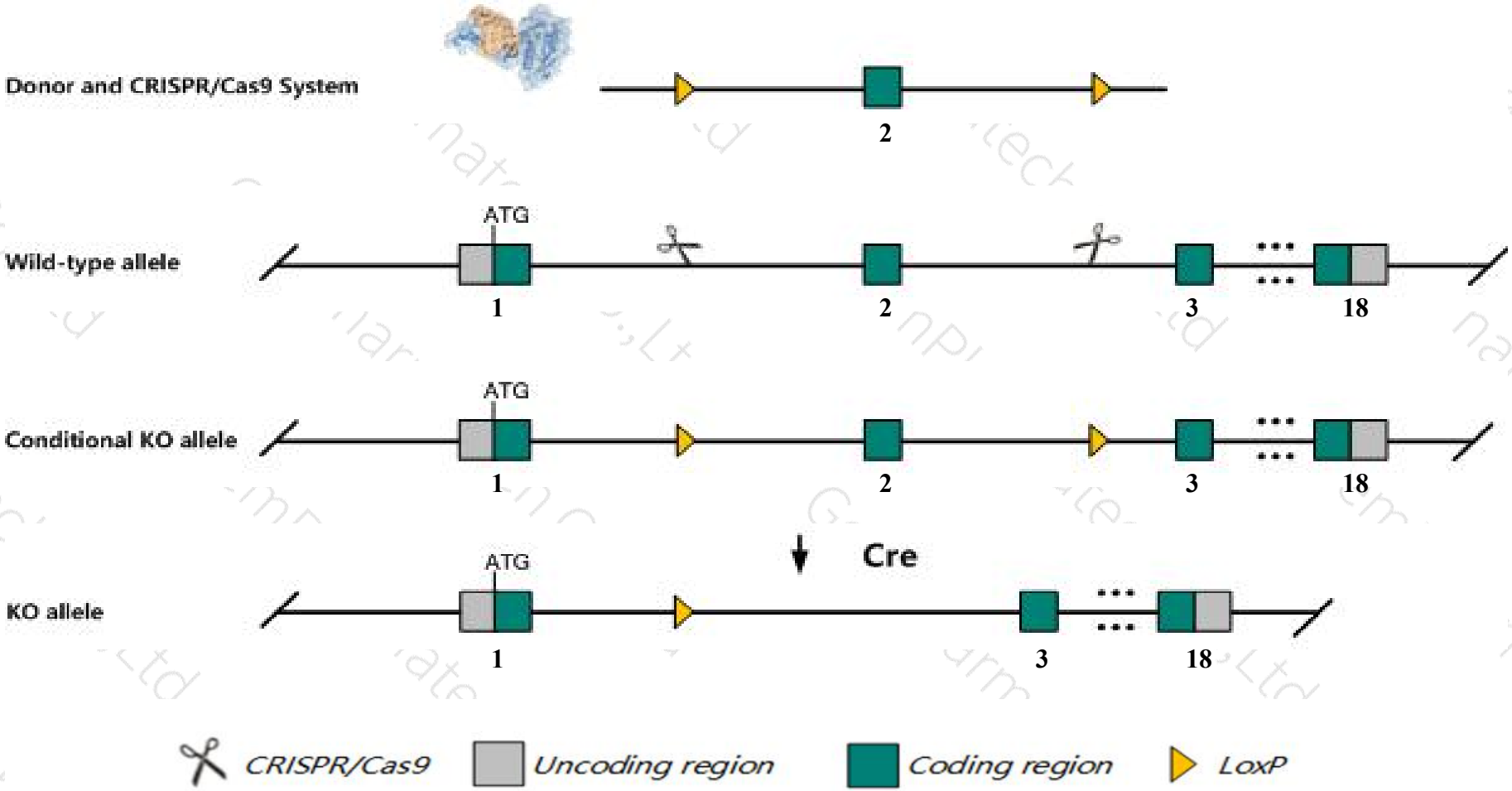
# Project Overview

| | |
|---|---|
| **Project Name** | ***Camk2d*** |
| **Project type** | **Cas9-CKO** |
| **Strain background** | **C57BL/6JGpt** |

# Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Camk2d* gene. The schematic diagram is as follows:



# Technical routes

- The *Camk2d* gene has 28 transcripts. According to the structure of *Camk2d* gene, exon2 of *Camk2d-226* (ENSMUST00000199300.4) transcript is recommended as the knockout region.The region contains 95bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Camk2d* 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

- According to the existing MGI data,Mice homozygous for a knock-out allele exhibit reduced response to heart induced stress. Mice homozygous for an allele that produces an oxidant-resistant product exhibit reduced response to myocardial infarction in a diabetic model.
- *Gm43011* gene will be destroyed in this strategy.
- Transcript *Camk2d*-201&211&214&217&218&220&222&223&224&225&227 may not be affected.
- The *Camk2d* gene is located on the Chr3. 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）

集萃药康 GemPharmatech

**Camk2d calcium/calmodulin-dependent protein kinase II, delta [ *Mus musculus* (house mouse) ]**

Gene ID: 108058, updated on 10-Oct-2019

## Summary

| | |
|---|---|
| **Official Symbol** | Camk2d provided by MGI |
| **Official Full Name** | calcium/calmodulin-dependent protein kinase II, delta provided by MGI |
| **Primary source** | MGI:MGI:1341265 |
| **See related** | Ensembl:ENSMUSG00000053819 |
| **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** | CaMK II; [d]-CaMKII; 2810011D23Rik; 8030469K03Rik |
| **Expression** | Ubiquitous expression in bladder adult (RPKM 17.7), adrenal adult (RPKM 15.0) and 24 other tissues See more |
| **Orthologs** | human all |

## Genomic context

**Location:** 3; 3 G1

See Camk2d in Genome Data Viewer

**Exon count:** 26

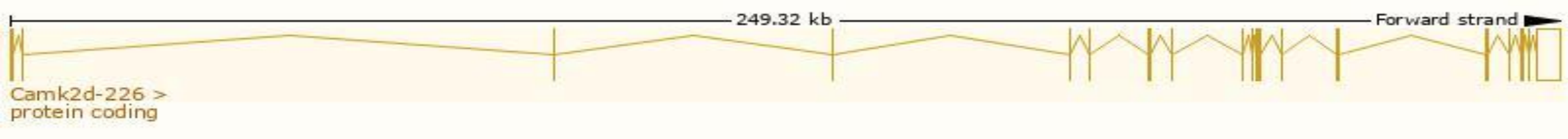
| Annotation release | Status | Assembly | Chr | Location |
|---|---|---|---|---|
| 108 | current | GRCm38.p6 (GCF_000001635.26) | 3 | NC_000069.6 (126595487..126846326) |
| Build 37.2 | previous assembly | MGSCv37 (GCF_000001635.18) | 3 | NC_000069.5 (126299891..126547972) |

# Transcript information（Ensembl）

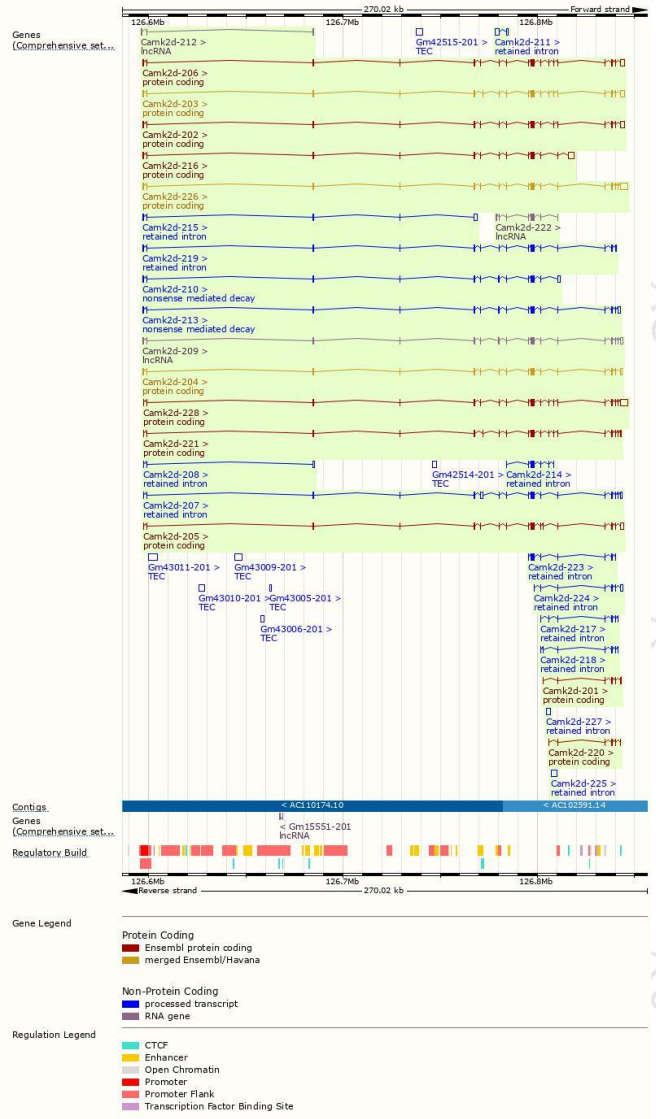
The gene has 28 transcripts,all transcripts are shown below:

| Name | Transcript ID | bp | Protein | Biotype | CCDS | UniProt | Flags |
|---|---|---|---|---|---|---|---|
| Camk2d-226 | ENSMUST00000199300.4 | 5524 | 499aa | Protein coding | CCDS17823 | Q6PHZ2 | TSL:1 GENCODE basic APPRIS P3 |
| Camk2d-228 | ENSMUST00000200171.4 | 5396 | 533aa | Protein coding | CCDS84670 | A0A0G2JGS4 | TSL:5 GENCODE basic APPRIS ALT1 |
| Camk2d-216 | ENSMUST00000163226.7 | 4204 | 361aa | Protein coding | CCDS80017 | Q8CCM0 | TSL:1 GENCODE basic |
| Camk2d-206 | ENSMUST00000106402.7 | 4180 | 512aa | Protein coding | CCDS80015 | Q6PHZ2 | TSL:1 GENCODE basic |
| Camk2d-203 | ENSMUST00000106399.7 | 4169 | 512aa | Protein coding | CCDS38624 | E9Q1W0 | TSL:1 GENCODE basic |
| Camk2d-202 | ENSMUST00000066466.12 | 4075 | 492aa | Protein coding | CCDS80016 | Q6PHZ2 | TSL:1 GENCODE basic |
| Camk2d-204 | ENSMUST00000106400.8 | 2722 | 478aa | Protein coding | CCDS51065 | Q6PHZ2 | TSL:1 GENCODE basic |
| Camk2d-221 | ENSMUST00000171289.7 | 2217 | 533aa | Protein coding | CCDS84671 | E9Q1T1 | TSL:5 GENCODE basic APPRIS ALT1 |
| Camk2d-205 | ENSMUST00000106401.7 | 3059 | 488aa | Protein coding | - | E9Q1V9 | TSL:5 GENCODE basic |
| Camk2d-201 | ENSMUST00000066452.13 | 896 | 160aa | Protein coding | - | E9PXV3 | CDS 5' incomplete TSL:5 |
| Camk2d-220 | ENSMUST00000170149.1 | 640 | 169aa | Protein coding | - | F6RWZ9 | CDS 5' incomplete TSL:3 |
| Camk2d-213 | ENSMUST00000145454.7 | 3178 | 142aa | Nonsense mediated decay | - | E9QAJ4 | TSL:2 |
| Camk2d-210 | ENSMUST00000134466.7 | 2708 | 147aa | Nonsense mediated decay | - | D6RDQ8 | TSL:2 |
| Camk2d-207 | ENSMUST00000129293.7 | 3479 | No protein | Retained intron | - | - | TSL:1 |
| Camk2d-211 | ENSMUST00000134987.2 | 3114 | No protein | Retained intron | - | - | TSL:1 |
| Camk2d-225 | ENSMUST00000198637.1 | 3088 | No protein | Retained intron | - | - | TSL:NA |
| Camk2d-227 | ENSMUST00000200010.1 | 2369 | No protein | Retained intron | - | - | TSL:NA |
| Camk2d-215 | ENSMUST00000149311.7 | 2328 | No protein | Retained intron | - | - | TSL:2 |
| Camk2d-219 | ENSMUST00000169051.7 | 1960 | No protein | Retained intron | - | - | TSL:2 |
| Camk2d-224 | ENSMUST00000198487.4 | 1844 | No protein | Retained intron | - | - | TSL:5 |
| Camk2d-208 | ENSMUST00000131156.7 | 992 | No protein | Retained intron | - | - | TSL:2 |
| Camk2d-214 | ENSMUST00000147043.5 | 746 | No protein | Retained intron | - | - | TSL:2 |
| Camk2d-223 | ENSMUST00000197660.4 | 744 | No protein | Retained intron | - | - | TSL:2 |
| Camk2d-217 | ENSMUST00000167417.4 | 618 | No protein | Retained intron | - | - | TSL:5 |
| Camk2d-218 | ENSMUST00000167985.7 | 591 | No protein | Retained intron | - | - | TSL:5 |
| Camk2d-209 | ENSMUST00000131869.7 | 2993 | No protein | lncRNA | - | - | TSL:1 |
| Camk2d-222 | ENSMUST00000171950.7 | 734 | No protein | lncRNA | - | - | TSL:5 |
| Camk2d-212 | ENSMUST00000143383.7 | 206 | No protein | lncRNA | - | - | TSL:5 |

The strategy is based on the design of *Camk2d-226* transcript,The transcription is shown below

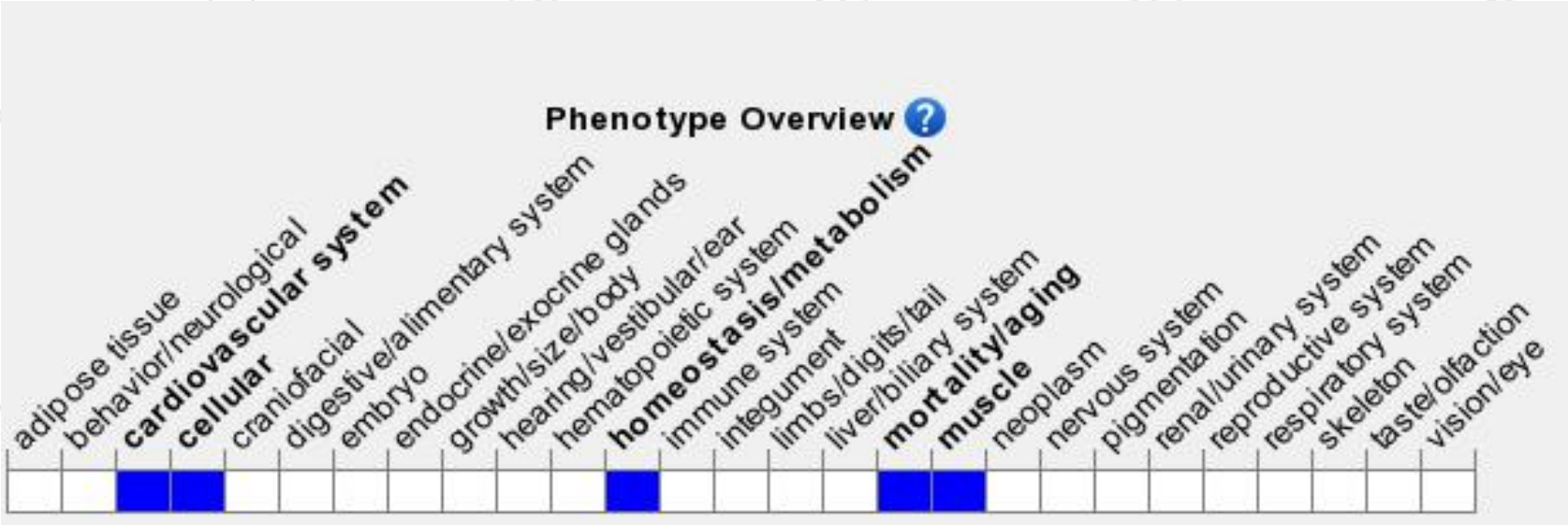


# Genomic location distribution



# 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 knock-out allele exhibit reduced response to heart induced stress. Mice homozygous for an allele that produces an oxidant-resistant product exhibit reduced response to myocardial infarction in a diabetic model.

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

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