

# Cdk2 Cas9-CKO Strategy

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# **Project Overview**

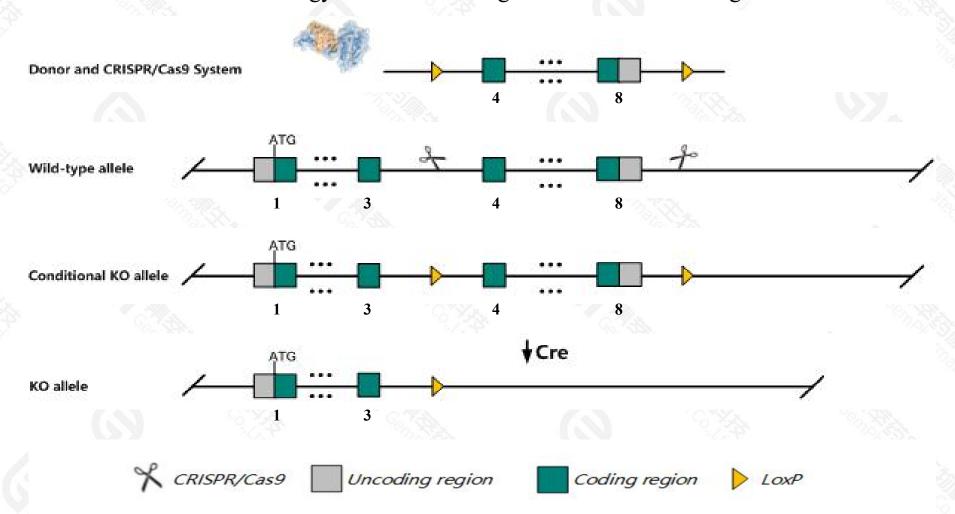


Project Name	Cdk2
Project type	Cas9-CKO
Strain background	C57BL/6JGpt

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Cdk2* gene. The schematic diagram is as follows:



### **Technical routes**



- The *Cdk2* gene has 7 transcripts. According to the structure of *Cdk2* gene, exon4-exon8 of *Cdk2-202*(ENSMUST00000026416.15) transcript is recommended as the knockout region. The region contains most of the coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Cdk2* 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 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, reproductive system abnormalities are observed in mice homozygous for disruptions in this gene. Gametogenesis fails in both males and females, leading to atrophy of the testes and ovaries. Both sexes are sterile.
- The floxed region is about 1.1kb and 1kb from *Rab5b* and *Pmel* gene. Knockout the region may affect the function of *Rab5b* and *Pmel* gene.
- > Some amino acids remain at the N terminal, and part of the protein's function may be preserved.
- > The Cdk2 gene is located on the Chr10. 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)



#### Cdk2 cyclin-dependent kinase 2 [Mus musculus (house mouse)]

Gene ID: 12566, updated on 13-Mar-2020

#### Summary

☆ ?

Official Symbol Cdk2 provided by MGI

Official Full Name cyclin-dependent kinase 2 provided by MGI

Primary source MGI:MGI:104772

See related Ensembl: ENSMUSG00000025358

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 A630093N05Rik

Expression Broad expression in limb E14.5 (RPKM 35.9), thymus adult (RPKM 24.5) and 25 other tissuesSee more

Orthologs human all

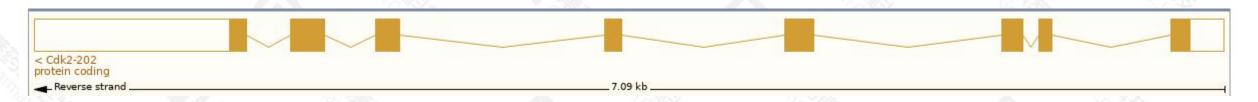
## Transcript information (Ensembl)



The gene has 7 transcripts, all transcripts are shown below:

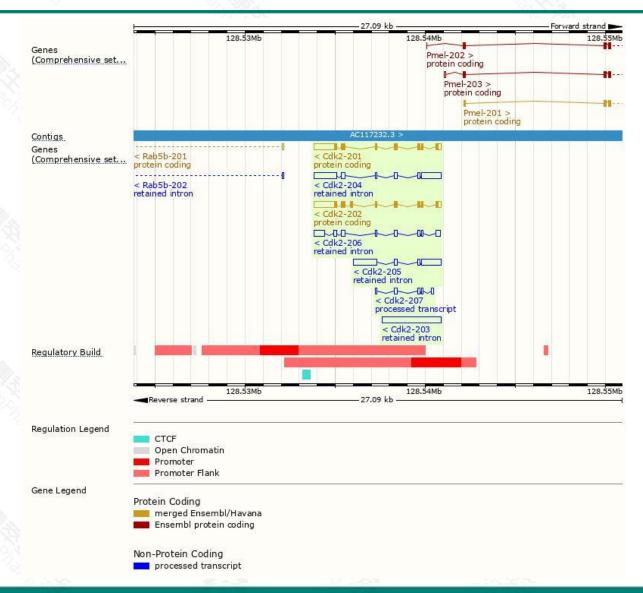
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Cdk2-202	ENSMUST00000026416.14	2406	346aa	Protein coding	CCDS24289	P97377 Q3UGB9	TSL:1 GENCODE basic
Cdk2-201	ENSMUST00000026415.8	2240	298aa	Protein coding	CCDS24288	P97377 Q3U6X7	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P
Cdk2-207	ENSMUST00000220407.1	549	No protein	Processed transcript	1.0	1.0	TSL:2
Cdk2-203	ENSMUST00000219047.1	3264	No protein	Retained intron	-	12	TSL:NA
Cdk2-204	ENSMUST00000219099.1	2971	No protein	Retained intron	-	-	TSL:1
Cdk2-205	ENSMUST00000219601.1	2673	No protein	Retained intron			TSL:1
Cdk2-206	ENSMUST00000219983.1	1708	No protein	Retained intron		100	TSL:1

The strategy is based on the design of Cdk2-202 transcript, the transcription is shown below:



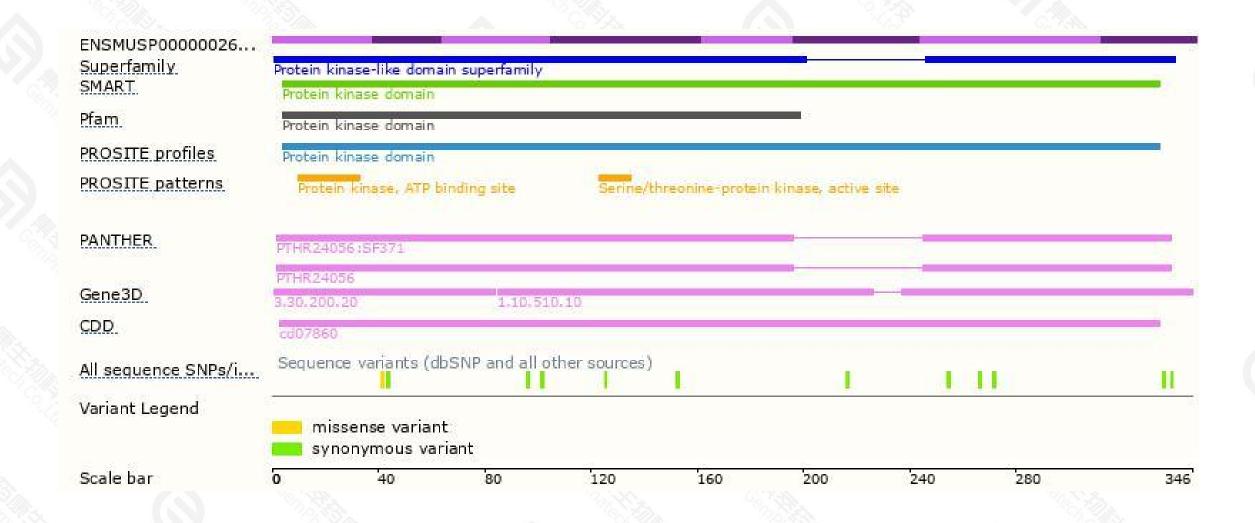
### Genomic location distribution





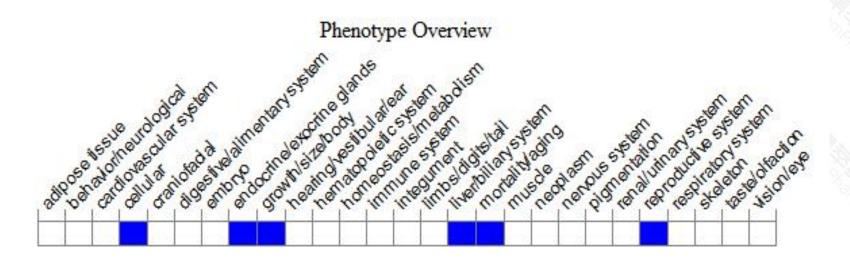
### Protein domain





## Mouse phenotype description(MGI)





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

According to the existing MGI data,reproductive system abnormalities are observed in mice homozygous for disruptions in this gene. Gametogenesis fails in both males and females, leading to atrophy of the testes and ovaries. Both sexes are sterile.



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

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