

# Kcna6 Cas9-CKO Strategy

Designer: Xueting Zhang

Reviewer: Yanhua Shen

Date:2020-02-19

## **Project Overview**



**Project Name** 

Kcna6

**Project type** 

Cas9-CKO

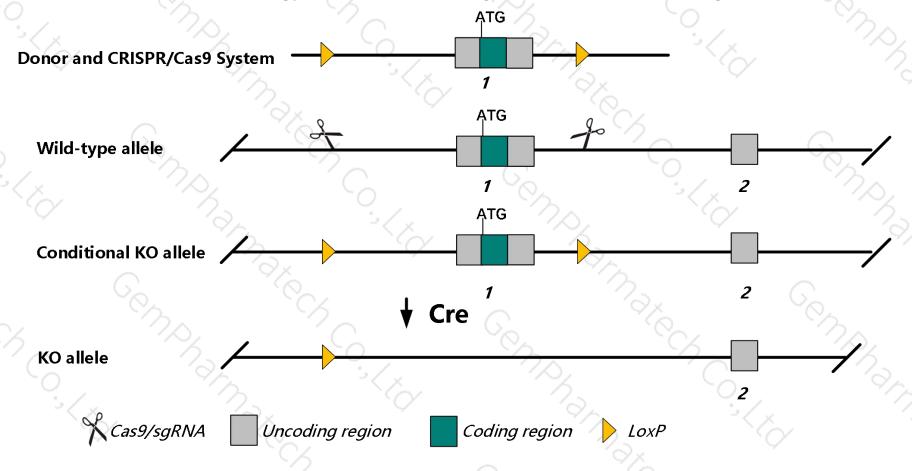
Strain background

C57BL/6JGpt

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Kcna6 gene. The schematic diagram is as follows:



### Technical routes



- The *Kcna6* gene has 4 transcripts. According to the structure of *Kcna6* gene, exon1 of *Kcna6-204* (ENSMUST00000185333.1) transcript is recommended as the knockout region. The region contains all 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 *Kcna6* 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, Homozygous mutation of this gene results in an increased thermal nociceptive threshold and in females an increase in circulating triglyceride levels.
- > *Gm26673* gene will be deleted.
- > The *Kcna6* gene is located on the Chr6. 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)



Kcna6 potassium voltage-gated channel, shaker-related, subfamily, member 6 [ Mus musculus (house mouse) ]

Gene ID: 16494, updated on 12-Aug-2019

#### Summary

☆ ?

Official Symbol Kcna6 provided by MGI

Official Full Name potassium voltage-gated channel, shaker-related, subfamily, member 6 provided by MGI

Primary source MGI:MGI:96663

See related Ensembl: ENSMUSG00000038077

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 Kv1.6; MK1.6

Expression Biased expression in cerebellum adult (RPKM 9.9), cortex adult (RPKM 8.5) and 7 other tissues See more

Orthologs <u>human</u> all

#### Genomic context

☆ ?

**Location:** 6 F3; 6 61.72 cM

See Kcna6 in Genome Data Viewer

Exon count: 4

Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 (GCF_000001635.26)	6	NC_000072.6 (126708328126740834, complement)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	6	NC_000072.5 (126658347126690692, complement)

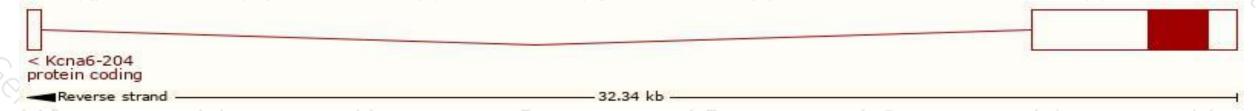
## Transcript information (Ensembl)



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

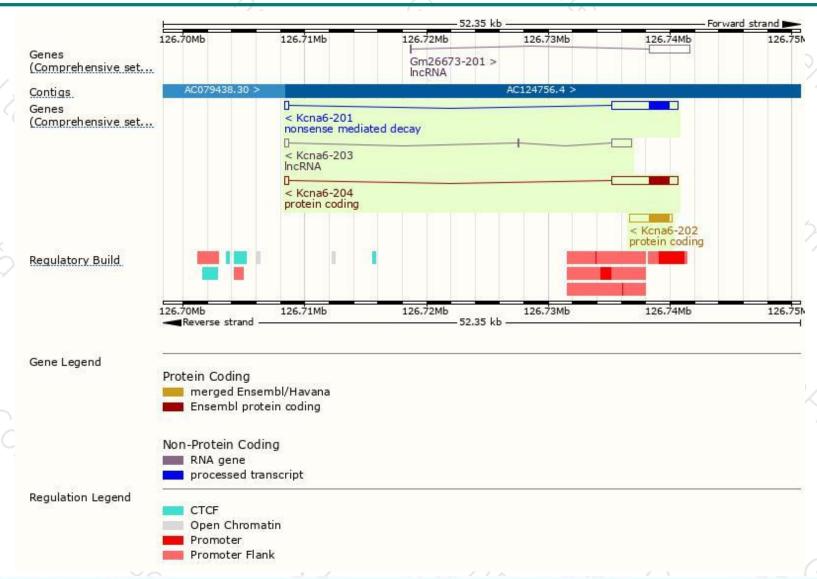
Name	Transcript ID	bp	Protein	Biotype	ccps	UniProt	Flags
		555		1000000000			Value Co. Data Company C. Maria Co. Data Co.
Kcna6-204	ENSMUST00000185333.1	5836	<u>529aa</u>	Protein coding	CCDS20556	Q61923	TSL:1 GENCODE basic APPRIS P1
Kcna6-202	ENSMUST00000112242.1	3464	<u>529aa</u>	Protein coding	CCDS20556	Q61923	TSL:NA GENCODE basic APPRIS P1
Kcna6-201	ENSMUST00000040751.5	5827	<u>529aa</u>	Nonsense mediated decay	CCDS20556	Q61923	TSL:1
Kcna6-203	ENSMUST00000128137.1	2080	No protein	IncRNA	62	-	TSL:1

The strategy is based on the design of *Kcna6-204* transcript, The transcription is shown below



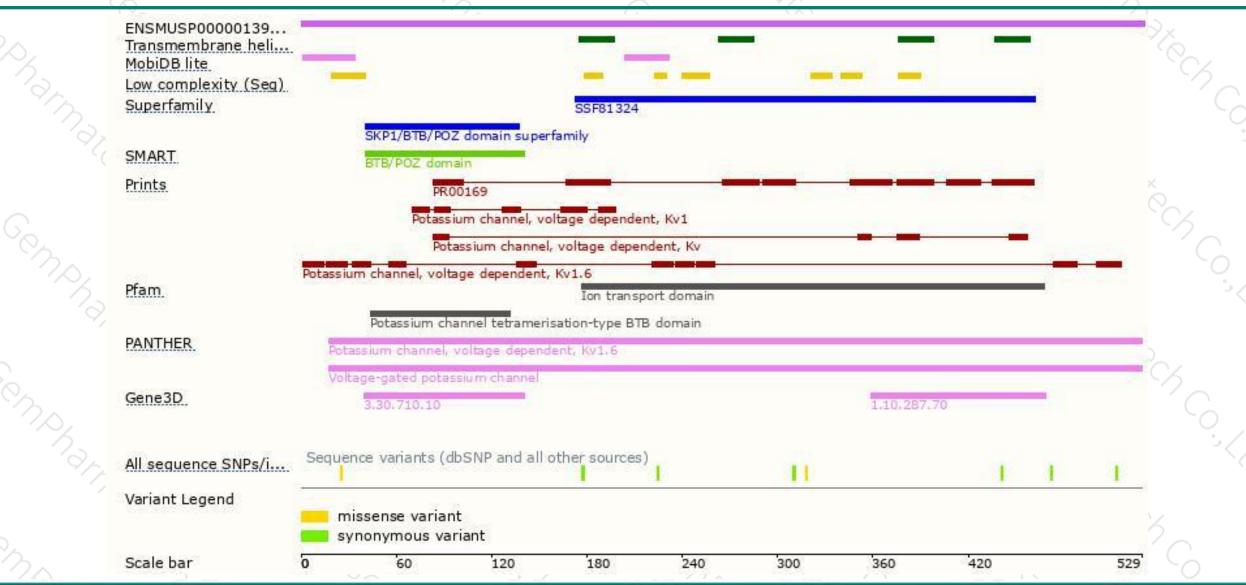
### Genomic location distribution





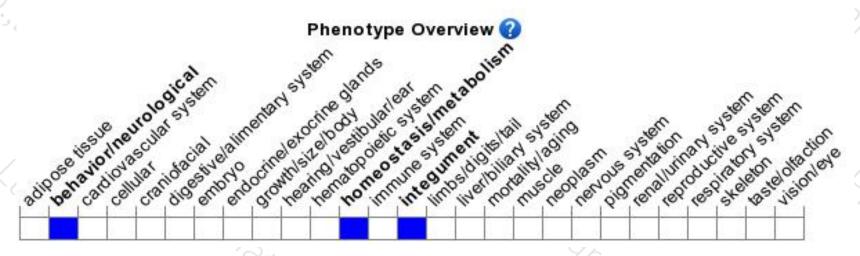
### Protein domain





## 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, Homozygous mutation of this gene results in an increased thermal nociceptive threshold and in females an increase in circulating triglyceride levels.



If you have any questions, you are welcome to inquire. Tel: 400-9660890





