Kcnn4 Cas9-KO Strategy

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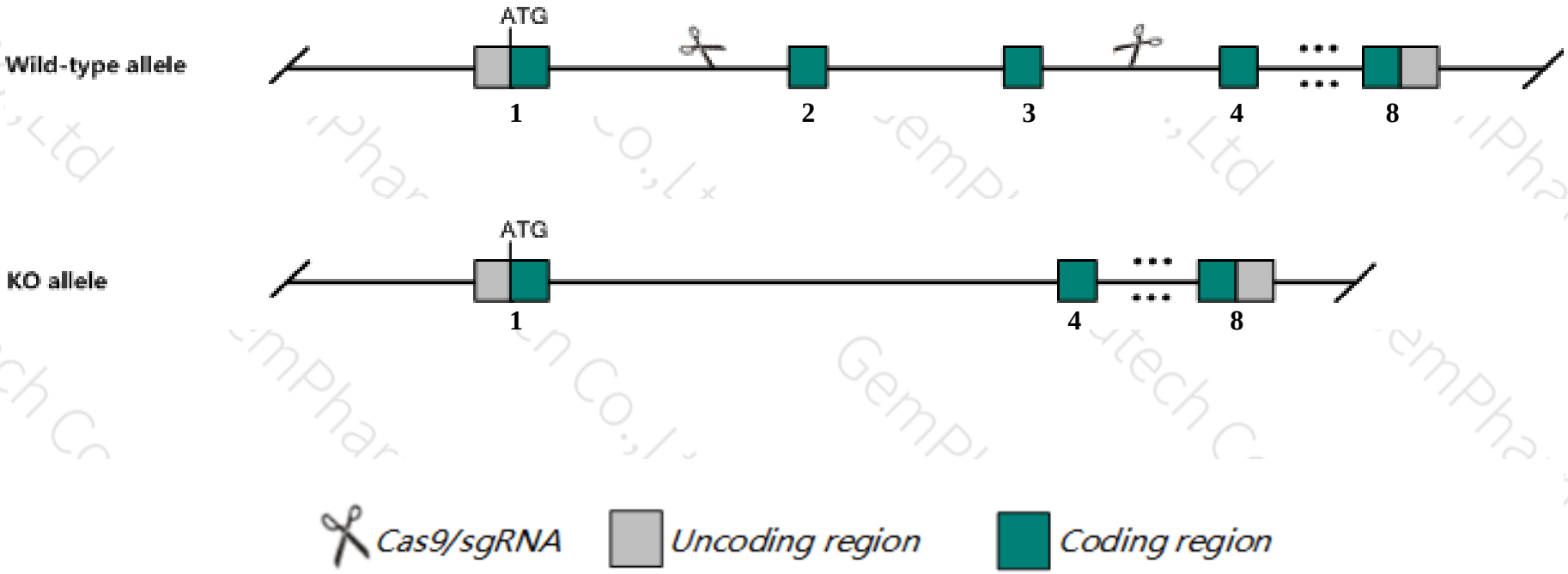
Project Overview

Project Name	***Kcnn4***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Kcnn4* gene. The schematic diagram is as follows:



Technical routes

➢ The *Kcnn4* gene has 5 transcripts. According to the structure of *Kcnn4* gene, exon2-exon3 of *Kcnn4-201* (ENSMUST00000171904.2) transcript is recommended as the knockout region.The region contains 518bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Kcnn4* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA 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.

Notice

- According to the existing MGI data,Homozygous null male mice have increased parotid gland weight and both sexes have impaired volume regulation in erythrocytes and T lymphocytes.

- The *Kcnn4* gene is located on the Chr7. 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information（NCBI）

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Kcnn4 potassium intermediate/small conductance calcium-activated channel, subfamily N, member 4 [Mus musculus (house mouse)]

Gene ID: 16534, updated on 2-Apr-2019

Summary

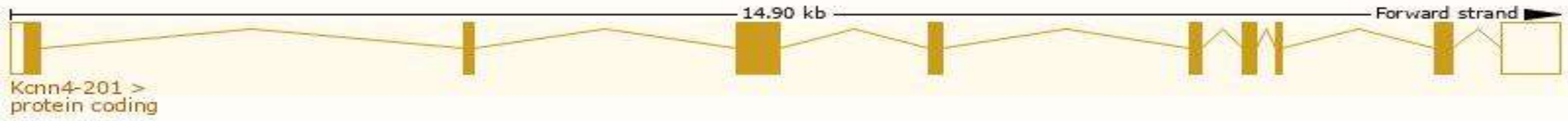
Official Symbol	Kcnn4 provided by MGI
Official Full Name	potassium intermediate/small conductance calcium-activated channel, subfamily N, member 4 provided byMGI
Primary source	MGI:MGI:1277957
See related	Ensembl:ENSMUSG00000054342
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	IK1, IKCA1, KCA4, KCa3.1, SK4, SKCas, mIKCa1
Expression	Biased expression in thymus adult (RPKM 85.0), liver E14.5 (RPKM 56.8) and 11 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

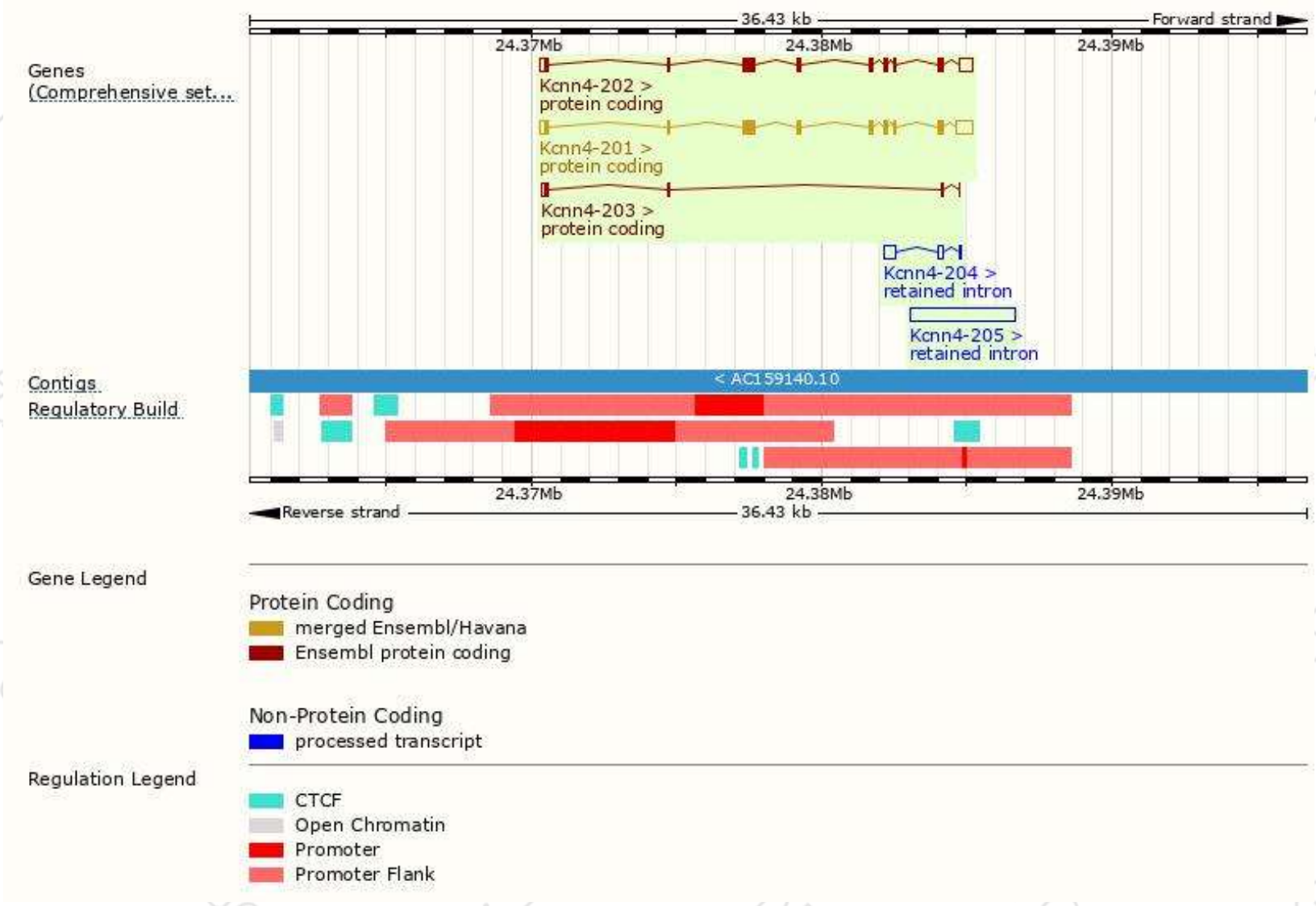
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Kcnn4-201	ENSMUST00000171904.2	1990	425aa	Protein coding	CCDS20948	O89109	TSL:1 GENCODE basic APPRIS P1
Kcnn4-202	ENSMUST00000205428.1	1945	425aa	Protein coding	CCDS20948	O89109	TSL:1 GENCODE basic APPRIS P1
Kcnn4-203	ENSMUST00000205626.1	461	104aa	Protein coding	-	A0A0U1RP82	TSL:5 GENCODE basic
Kcnn4-205	ENSMUST00000206910.1	3637	No protein	Retained intron	-	-	TSL:NA
Kcnn4-204	ENSMUST00000205881.1	614	No protein	Retained intron	-	-	TSL:2

The strategy is based on the design of *Kcnn4-201* transcript,The transcription is shown below

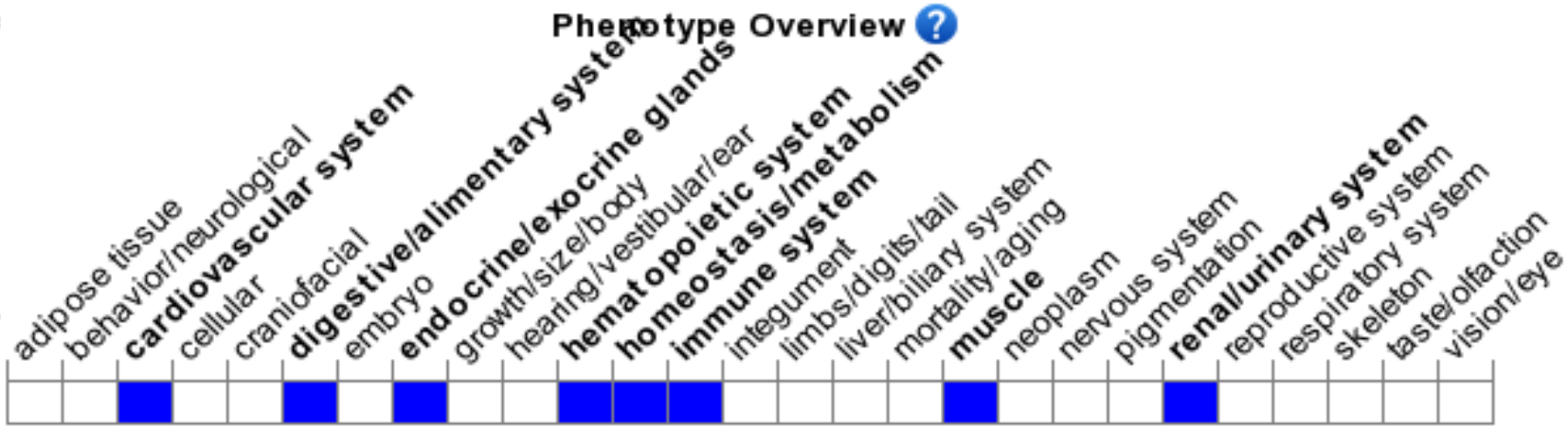


Genomic location distribution



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.

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