

Clcn5 Cas9-CKO Strategy

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Design Date:

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Project Overview

Project Name

Clcn5

Project type

Cas9-CKO

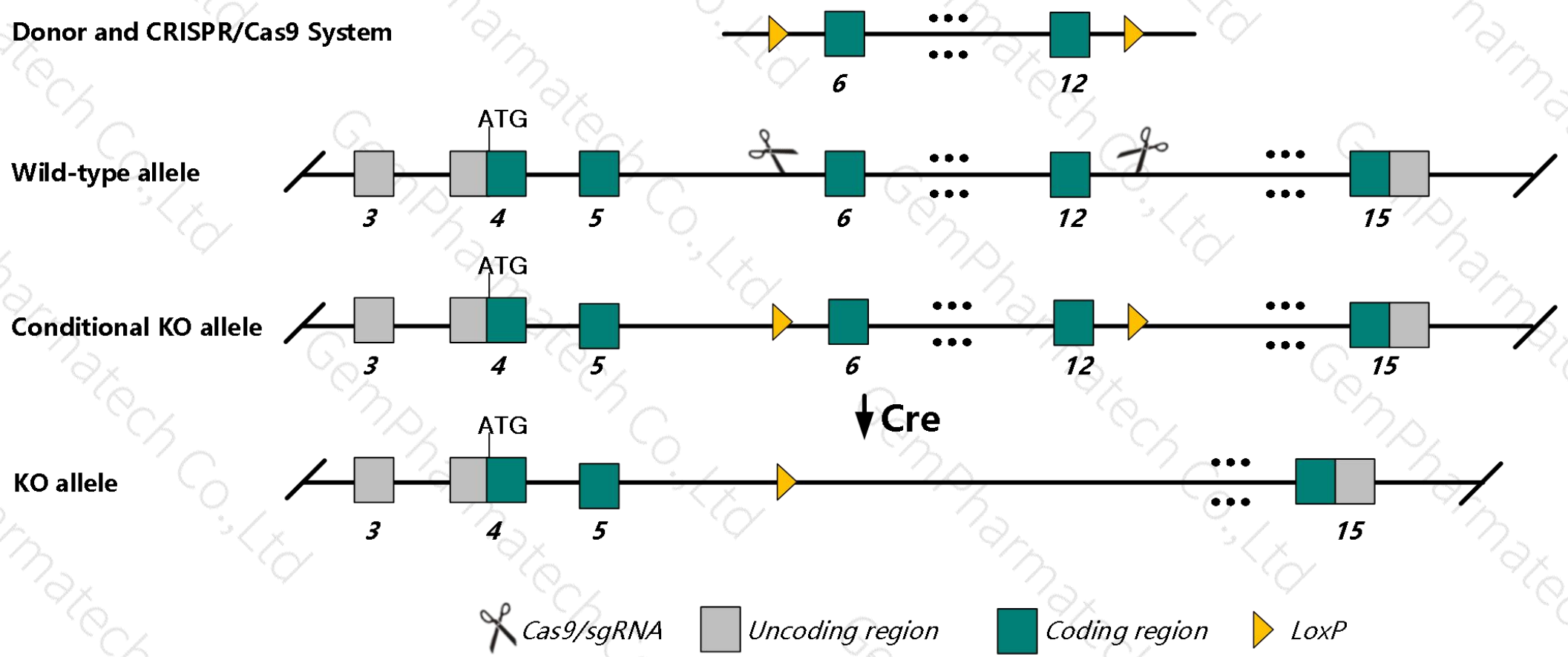
Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Cln5* gene. The schematic diagram is as follows:

Donor and CRISPR/Cas9 System



- The *Clcn5* gene has 5 transcripts. According to the structure of *Clcn5* gene, exon6-exon12 of *Clcn5*-202 (ENSMUST00000115746.7) transcript is recommended as the knockout region. The region contains 1429bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Clcn5* 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.

- According to the existing MGI data, Mice homozygous for targeted mutations that inactivate this gene display impaired endocytosis of filtered proteins by kidney proximal tubular cells and provide a model of Dents disease.
- The *Clcn5* gene is located on the ChrX. 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)

Clcn5 chloride channel, voltage-sensitive 5 [*Mus musculus* (house mouse)]

Gene ID: 12728, updated on 10-Oct-2019

Summary

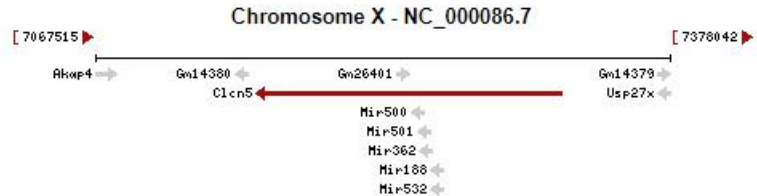
- Official Symbol** Clcn5 provided by [MGI](#)
- Official Full Name** chloride channel, voltage-sensitive 5 provided by [MGI](#)
- Primary source** [MGI:MGI:99486](#)
- See related** [Ensembl:ENSMUSG00000004317](#)
- Gene type** protein coding
- RefSeq status** VALIDATED
- Organism** [Mus musculus](#)
- Lineage** Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
- Also known as** Clc5; Clc-5; Sfc13; Clc4-1; T25545; Clcn4-1; DXImx42e; 5430408K11Rik; D930009B12Rik
- Expression** Broad expression in limb E14.5 (RPKM 8.4), placenta adult (RPKM 7.7) and 26 other tissues [See more](#)
- Orthologs** [human](#) [all](#)

Genomic context

Location: X A1.1; X 3.21 cM See Clcn5 in [Genome Data Viewer](#)

Exon count: 19

Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 (GCF_000001635.26)	X	NC_000086.7 (7153810..7319358, complement)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	X	NC_000086.6 (6735945..6765840, complement)

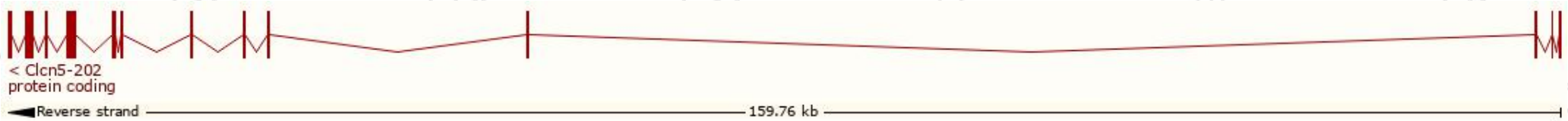


Transcript information (Ensembl)

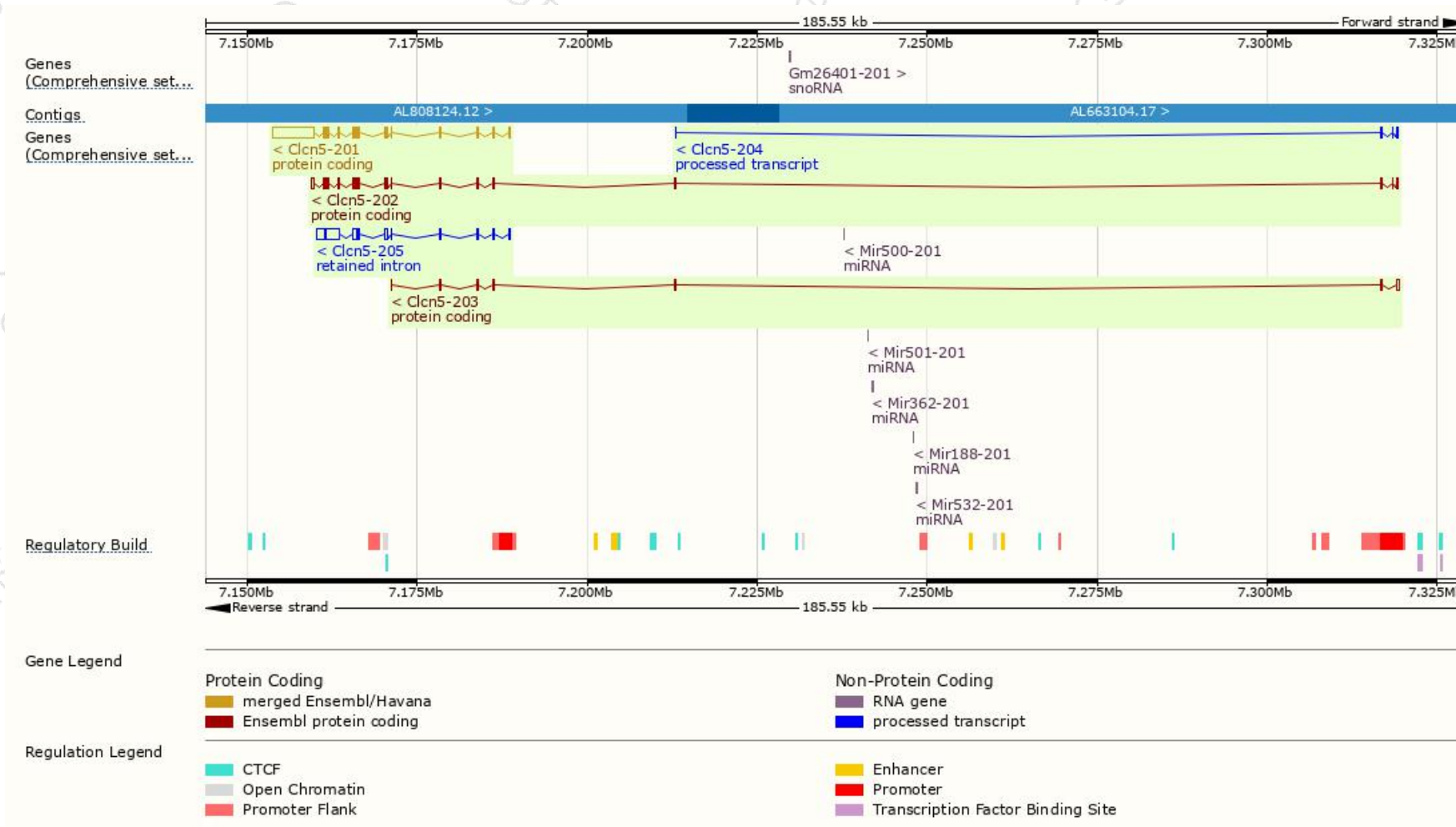
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Clcn5-201	ENSMUST00000004428.13	8390	746aa	Protein coding	CCDS29963	Q9WVD4	TSL:1 GENCODE basic APPRIS P3
Clcn5-202	ENSMUST00000115746.7	3052	816aa	Protein coding	CCDS57727	B1ATV0	TSL:5 APPRIS ALT1
Clcn5-203	ENSMUST00000128319.1	1073	223aa	Protein coding	-	B1ATV1	CDS 3' incomplete TSL:5
Clcn5-204	ENSMUST00000132126.1	355	No protein	Processed transcript	-	-	TSL:5
Clcn5-205	ENSMUST00000154382.1	4731	No protein	Retained intron	-	-	TSL:1

The strategy is based on the design of *Clcn5-202* transcript,The transcription is shown below



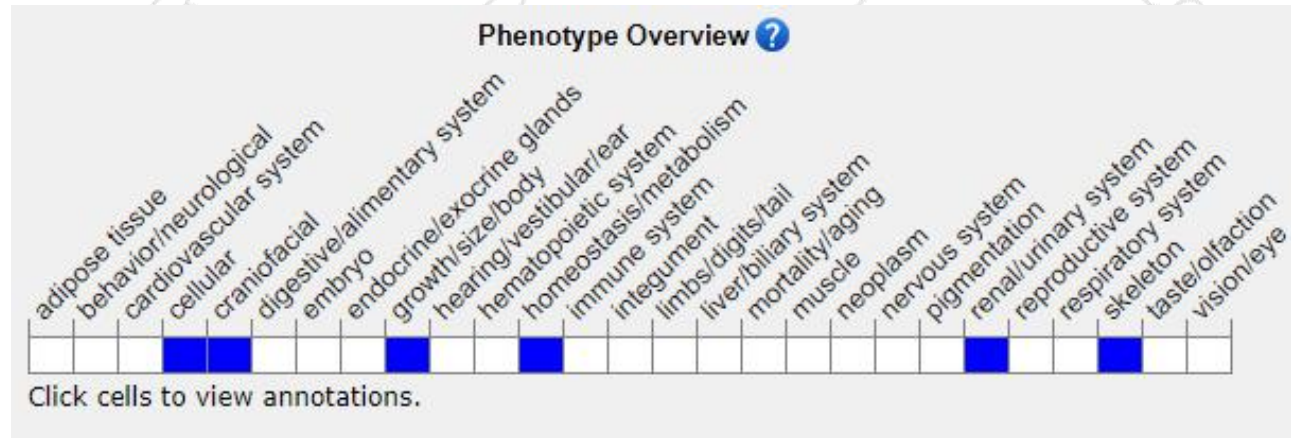
Genomic location distribution



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, Mice homozygous for targeted mutations that inactivate this gene display impaired endocytosis of filtered proteins by kidney proximal tubular cells and provide a model of Dents disease.

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

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