

Clcn1 Cas9-KO Strategy

Designer:Xueting Zhang

Reviewer: Yanhua Shen

Date:2019-11-18

Project Overview



Project Name

Clcn1

Project type

Cas9-KO

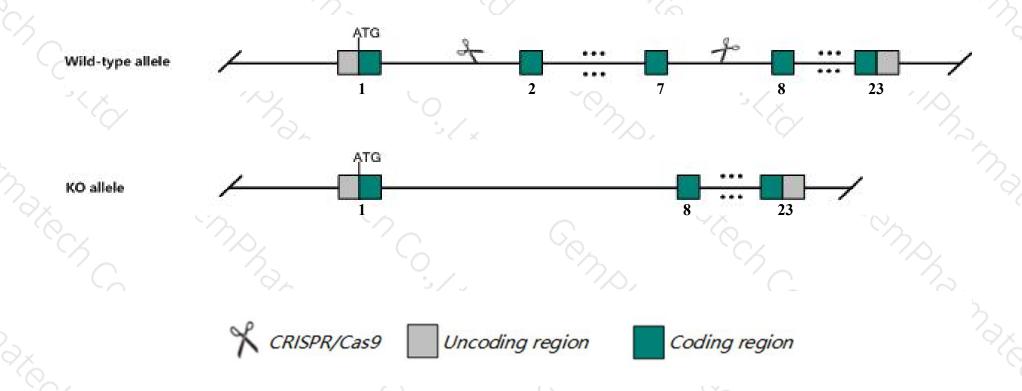
Strain background

C57BL/6JGpt

Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Clcn1* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Clcn1* gene has 10 transcripts. According to the structure of *Clcn1* gene, exon2-exon7 of *Clcn1-201*(ENSMUST00000031894.12) transcript is recommended as the knockout region. The region contains 673bp coding sequence Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Clcn1* gene. The brief process is as follows: CRISPR/Cas9 system

Notice



- ➤ According to the existing MGI data, Mutant mice exhibit mild to severe spasms of the hind limbs and abnormal hind limb reflexes.
- ➤ Transcript *Clcn1*-202&203&209 may not be affected.
- The *Clcn1* 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)



Clcn1 chloride channel, voltage-sensitive 1 [Mus musculus (house mouse)]

Gene ID: 12723, updated on 12-Aug-2019

Summary

☆ ?

Official Symbol Clcn1 provided by MGI

Official Full Name chloride channel, voltage-sensitive 1 provided by MGI

Primary source MGI:MGI:88417

See related Ensembl: ENSMUSG00000029862

Gene type protein coding
RefSeq status VALIDATED
Organism <u>Mus musculus</u>

Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia;

Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus

Also known as adr; mto; Clc1; Clc-1; SMCC1; nmf355; myotonia

Expression Biased expression in mammary gland adult (RPKM 3.1), cerebellum adult (RPKM 2.5) and 12 other tissues See more

Orthologs human all

Genomic context

☆ ?

Location: 6 B2.1; 6 20.57 cM

See Clcn1 in Genome Data Viewer

Exon count: 25

Annotation release Status		Assembly		Location		
108	current	GRCm38.p6 (GCF_000001635.26)	6	NC_000072.6 (4228485142315764)		
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	6	NC_000072.5 (4223668442264655)		

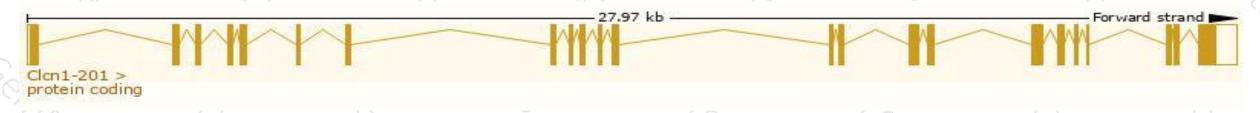
Transcript information (Ensembl)



The gene has 10 transcripts, all transcripts are shown below:

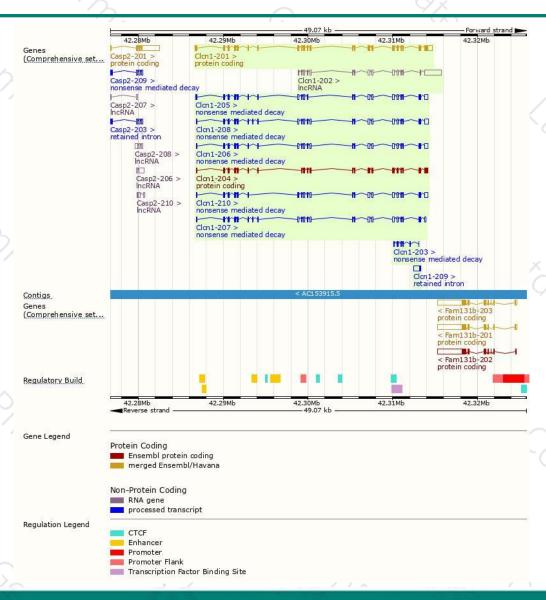
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Clcn1-201	ENSMUST00000031894.12	3545	994aa	Protein coding	CCDS39471	Q64347	TSL:1 GENCODE basic APPRIS P1
Clcn1-204	ENSMUST00000163936.7	2770	<u>923aa</u>	Protein coding	15 0	F6QI82	CDS 5' incomplete TSL:5
Clcn1-208	ENSMUST00000169024.7	3029	<u>262aa</u>	Nonsense mediated decay	84	F7C9S1	CDS 5' incomplete TSL:5
Clcn1-206	ENSMUST00000165780.7	2899	229aa	Nonsense mediated decay	62	F6QL23	CDS 5' incomplete TSL:5
Clcn1-205	ENSMUST00000164091.7	2838	258aa	Nonsense mediated decay	85	E9Q2N3	TSL:5
Clcn1-210	ENSMUST00000170028.7	2817	289aa	Nonsense mediated decay	19 1	F6X177	CDS 5' incomplete TSL:5
Clcn1-207	ENSMUST00000168660.3	2744	<u>257aa</u>	Nonsense mediated decay	84	F7A9J6	CDS 5' incomplete TSL:2
Clcn1-203	ENSMUST00000163235.1	417	<u>104aa</u>	Nonsense mediated decay	62	F6VSA8	CDS 5' incomplete TSL:5
Clcn1-209	ENSMUST00000169902.1	774	No protein	Retained intron			TSL:3
Clcn1-202	ENSMUST00000114684.7	3504	No protein	IncRNA		-	TSL:1

The strategy is based on the design of Clcn1-201 transcript, The transcription is shown below



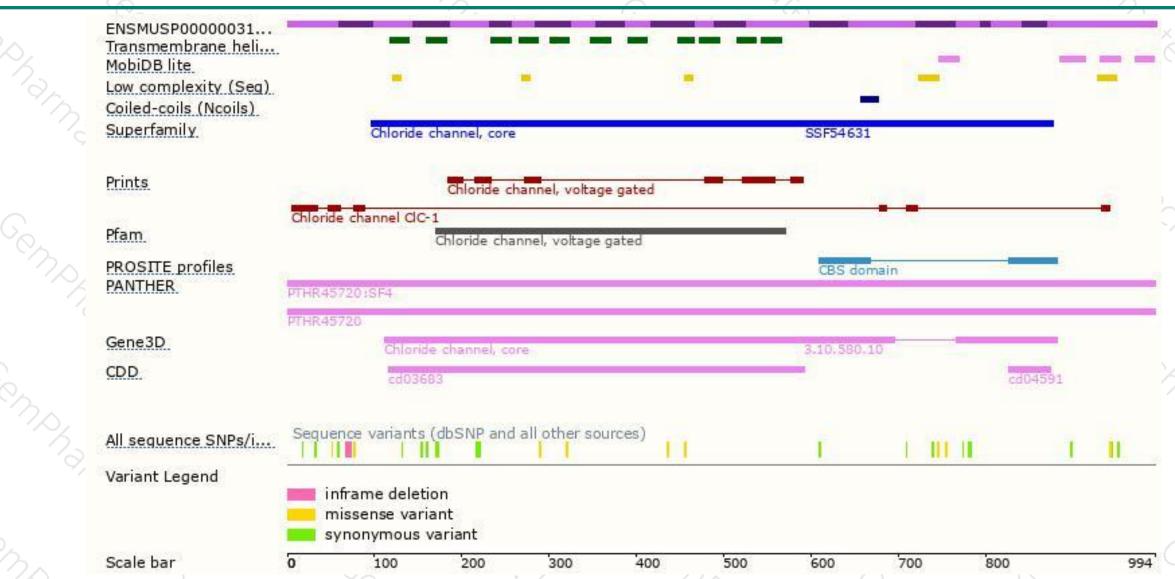
Genomic location distribution





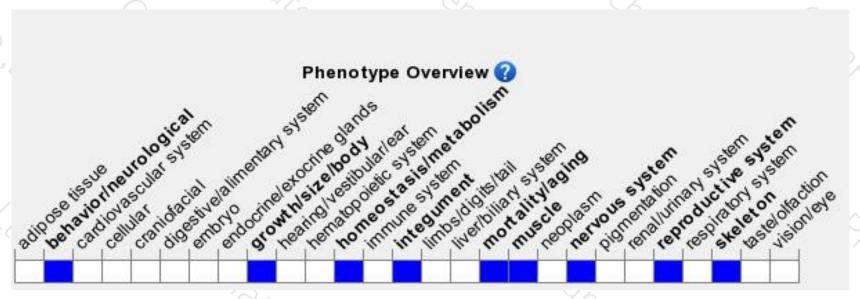
Protein domain





Mouse phenotype description(MGI)





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

According to the existing MGI data, Mutant mice exhibit mild to severe spasms of the hind limbs and abnormal hind limb reflexes.



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





