Clcn6 Cas9-KO Strategy

Designer:

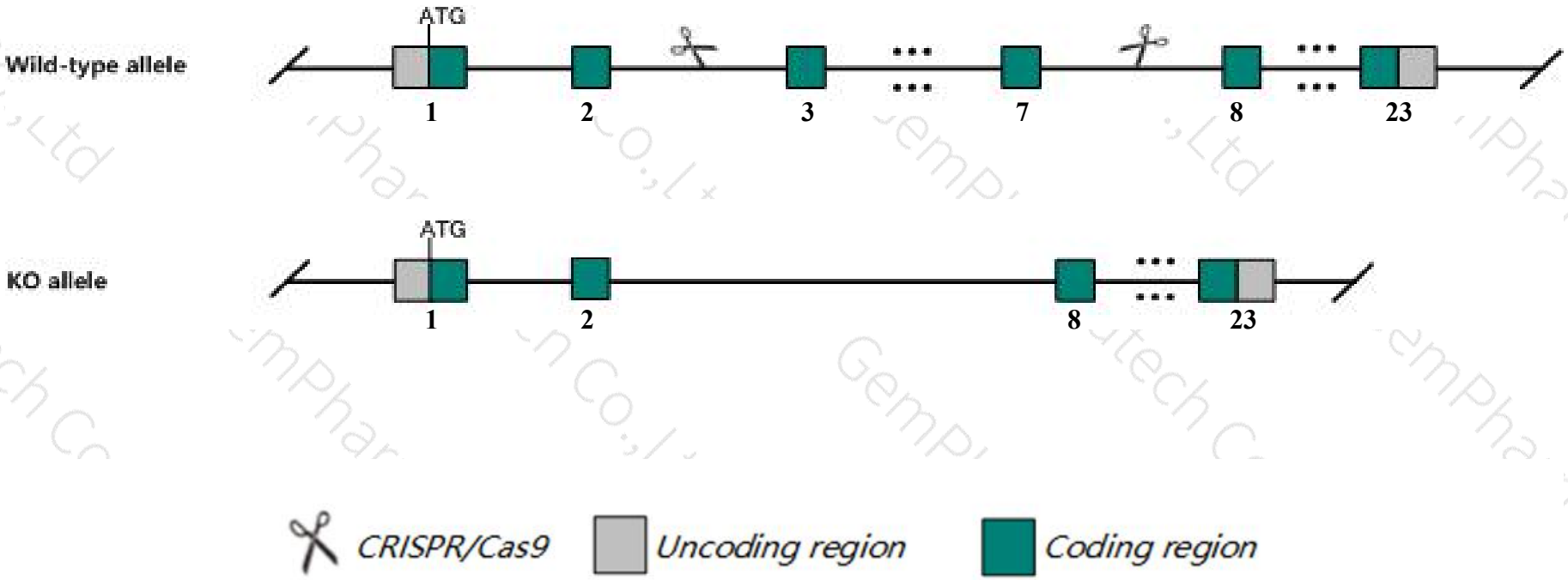
GemPharmatech Co.,Ltd

Project Overview

Project Name	***Clcn6***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Clcn6* gene. The schematic diagram is as follows:



Technical routes

➢ The *Clcn6* gene has 5 transcripts. According to the structure of *Clcn6* gene, exon3-exon7 of *Clcn6-201* (ENSMUST00000030879.11) transcript is recommended as the knockout region.The region contains 433bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Clcn6* gene. The brief process is as follows: CRISPR/Cas9 system

Notice

➢ According to the existing MGI data,Mice homozygous for a knock-out allele exhibit impaired nociception, mild behavioral abnormalities, and a progressive neuropathy of the central and peripheral nervous systems with features of neuronal ceroid lipofuscinosis (a lysosomal storage disease).

➢ The *Clcn6* gene is located on the Chr4. 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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Clcn6 chloride channel, voltage-sensitive 6 [Mus musculus (house mouse)]

Gene ID: 26372, updated on 31-Jan-2019

Summary

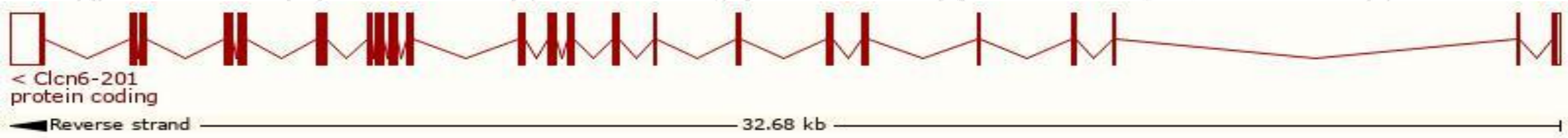
Official Symbol	Clcn6 provided by MGI
Official Full Name	chloride channel, voltage-sensitive 6 provided by MGI
Primary source	MGI:MGI:1347049
See related	Ensembl:ENSMUSG00000029016
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AI850629, Clc6
Summary	This gene encodes a member of the ClC chloride channel and transporter family of proteins. The encoded protein may function as a vesicular Cl-/H+ antiporter. Homozygous knockout mice exhibit decreased pain sensitivity, behavioral abnormalities and features of lysosomal storage disease. [provided by RefSeq, Aug 2015]
Expression	Ubiquitous expression in whole brain E14.5 (RPKM 17.3), CNS E14 (RPKM 16.1) and 28 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

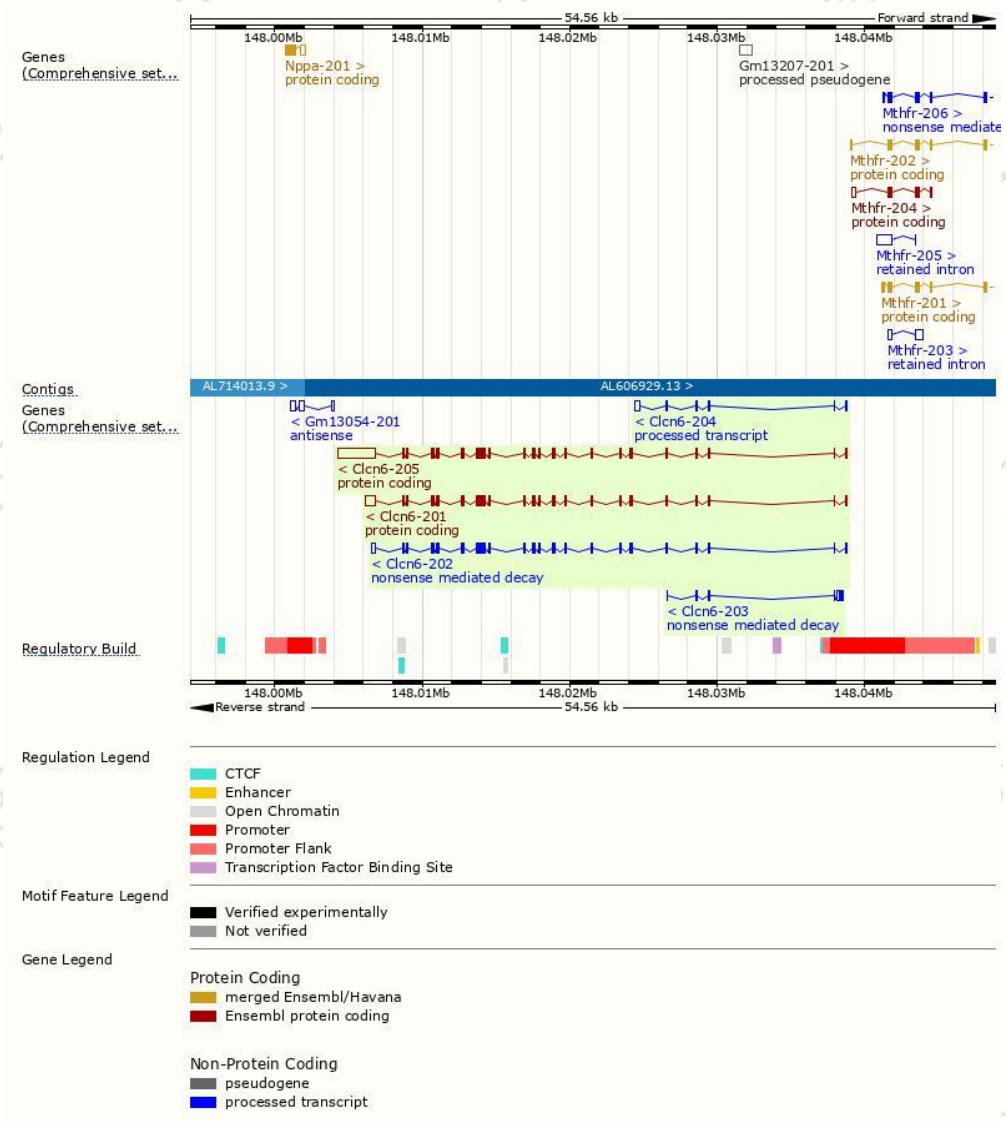
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Clcn6-201	ENSMUST00000030879.11	3306	870aa	Protein coding	CCDS18928	O35454 Q3UM91	TSL:1 GENCODE basic APPRIS P2
Clcn6-205	ENSMUST00000137724.7	5198	873aa	Protein coding	-	A2A7F6	TSL:5 GENCODE basic APPRIS ALT1
Clcn6-202	ENSMUST00000105711.2	2783	731aa	Nonsense mediated decay	-	E9Q741	TSL:5
Clcn6-203	ENSMUST00000131232.1	627	60aa	Nonsense mediated decay	-	D6RHT0	TSL:5
Clcn6-204	ENSMUST00000134377.7	660	No protein	Processed transcript	-	-	TSL:3

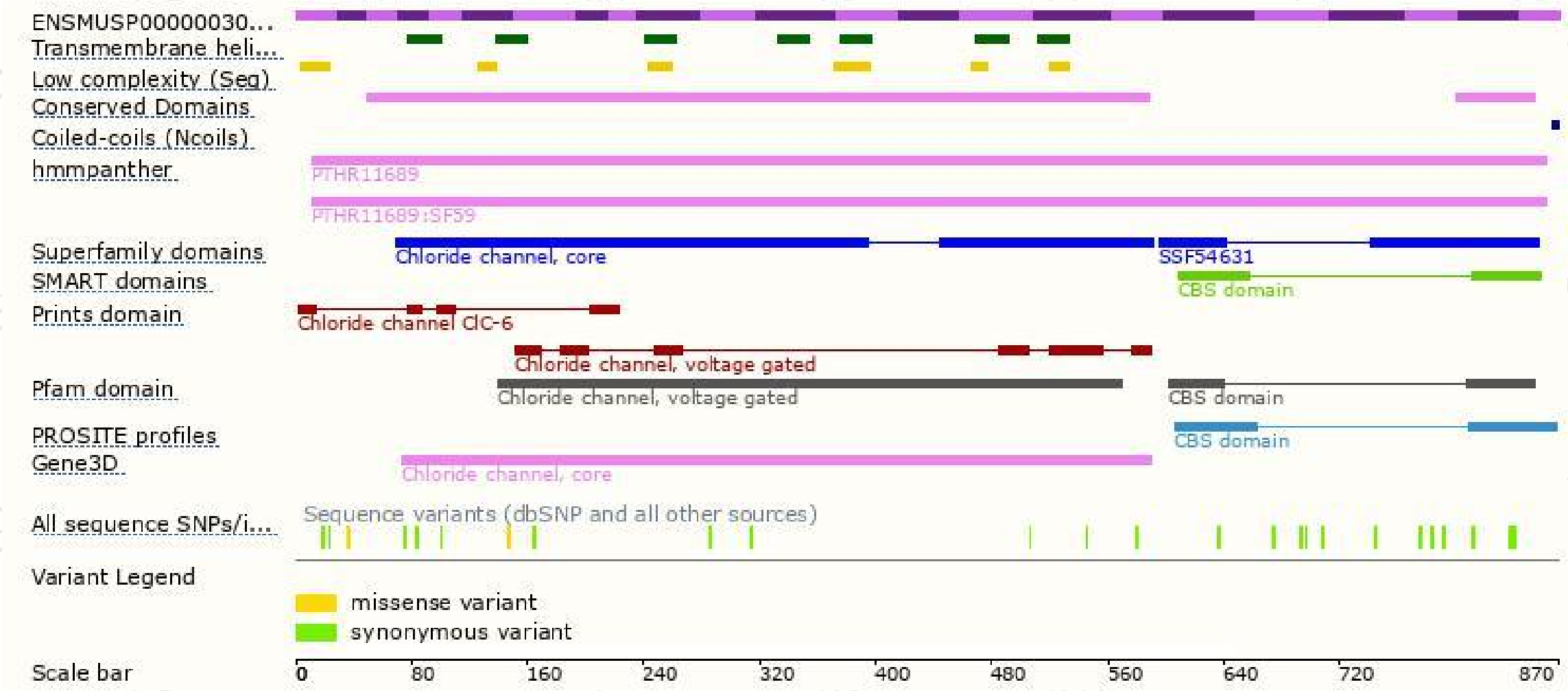
The strategy is based on the design of *Clcn6-201* transcript,The transcription is shown below



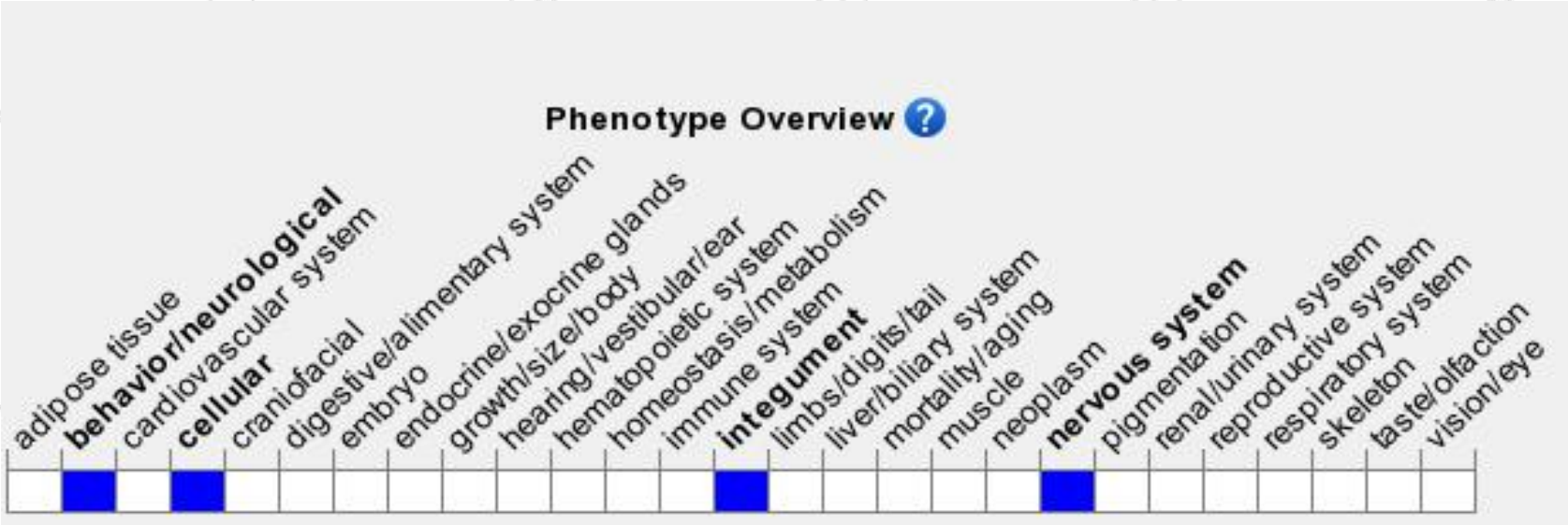
Genomic location distribution



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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