

Chp1 Cas9-CKO Strategy

Designer: Yun Li

Reviewer:Longyun Hu

Design Date: 2024-3-12

Overview

Target Gene Name

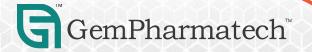
• Chp1

Project Type

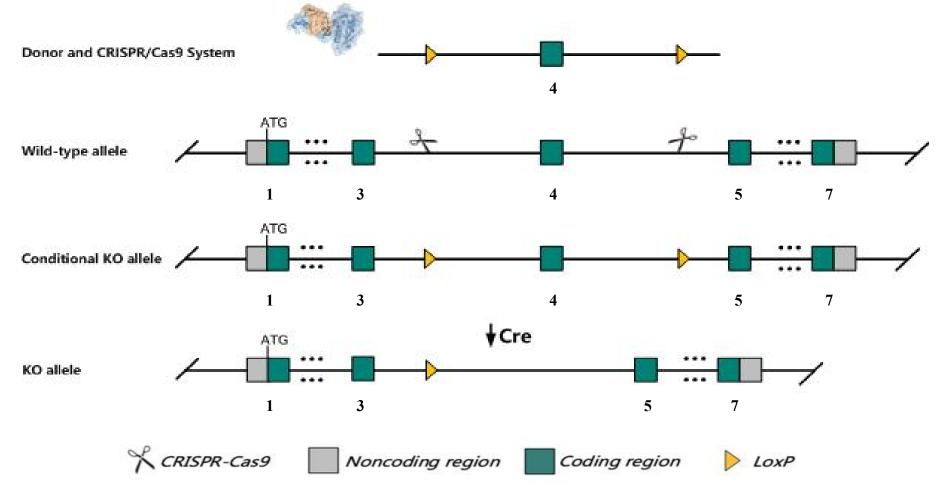
• Cas9-CKO

Genetic Background

• C57BL/6JGpt



Strain Strategy



Schematic representation of CRISPR-Cas9 engineering used to edit the Chp1 gene.



Technical Information

- The *Chp1* gene has 5 transcripts. According to the structure of *Chp1* gene, exon4 of *Chp1*-201 (ENSMUST00000014221.13) transcript is recommended as the knockout region. The region contains 128bp coding sequence. Knocking out the region will result in disruption of protein function.
- In this project we use CRISPR-Cas9 technology to modify *Chp1* 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 on-target amplicon sequencing. A stable F1-generation mouse strain was obtained by mating positive F0-generation mice with C57BL/6JGpt mice and confirmation of the desired mutant allele was carried out by PCR and on-target amplicon sequencing.
- 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.



Gene Information

Chp1 calcineurin-like EF hand protein 1 [Mus musculus (house mouse)]

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Gene ID: 56398, updated on 5-Mar-2024



Official Symbol Chp1 provided by MGI

Official Full Name calcineurin-like EF hand protein 1 provided by MGI

Primary source MGI:MGI:1927185

See related Ensembl: ENSMUSG00000014077 Alliance Genome: MGI: 1927185

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 Chp; p24; vac; Cahp; Sid470p; 1500003O03Rik

Summary Predicted to enable several functions, including calcium ion binding activity; calcium-dependent protein binding activity; and microtubule binding activity. Predicted to

be involved in several processes, including microtubule cytoskeleton organization; regulation of protein modification process; and regulation of protein transport. Predicted to act upstream of or within protein transport. Predicted to be located in several cellular components, including Golgi membrane; microtubule cytoskeleton; and transport vesicle. Is expressed in genitourinary system. Human ortholog(s) of this gene implicated in spastic ataxia. Orthologous to human CHP1 (calcineurin like

EF-hand protein 1). [provided by Alliance of Genome Resources, Apr 2022]

Expression Ubiquitous expression in subcutaneous fat pad adult (RPKM 137.3), mammary gland adult (RPKM 107.2) and 28 other tissues See more

Orthologs human all

Source: https://www.ncbi.nlm.nih.gov/

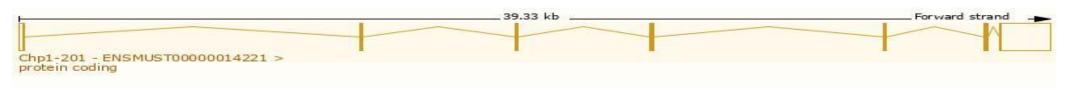


Transcript Information

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

Transcript ID A	Name 🌲	bp 🌲	Protein 🌲	Biotype	CCDS 🍦	UniProt Match	Flags
ENSMUST00000014221.13	Chp1-201	2609	<u>195aa</u>	Protein coding	CCDS16604&	P61022₽	Ensembl Canonical GENCODE basic APPRIS P1 TSL:1
ENSMUST00000119172.2	Chp1-202	3027	<u>191aa</u>	Protein coding		B0R091 ₽	GENCODE basic TSL:2
ENSMUST00000132448.8	Chp1-203	518	<u>33aa</u>	Protein coding		B0R092₽	TSL:5 CDS 3' incomplete
ENSMUST00000137585.3	Chp1-204	738	No protein	Retained intron		-	TSL:2
ENSMUST00000154406.8	Chp1-205	543	No protein	Retained intron		-	TSL:2

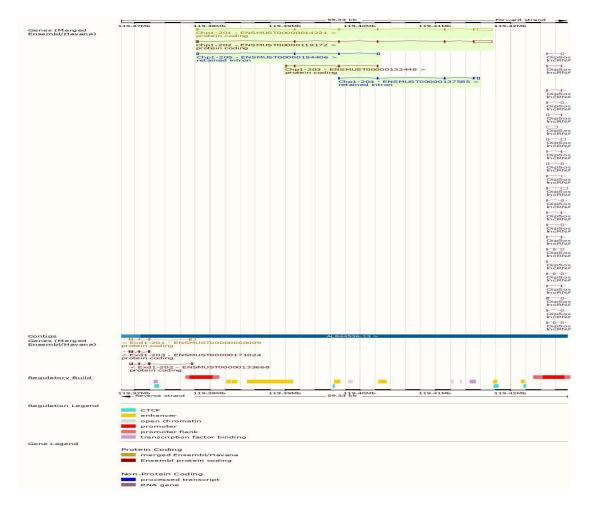
The strategy is based on the design of *Chp1*-201 transcript, the transcription is shown below:

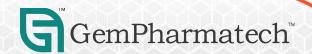


Source: https://www.ensembl.org



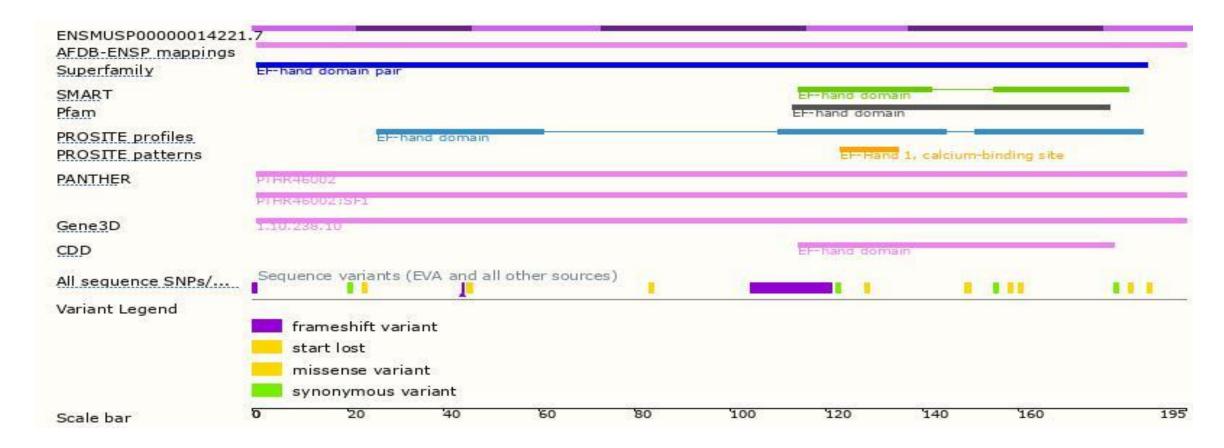
Genomic Information





Source: : https://www.ensembl.org

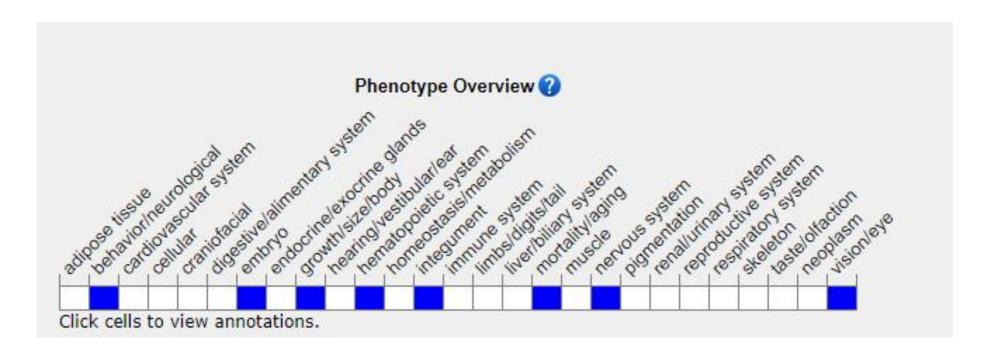
Protein Information





Source: : https://www.ensembl.org

Mouse Phenotype Information (MGI)



• Mice homozygous for an ENU mutation display ataxia and progressive Purkinje cell axonal dystrophy.



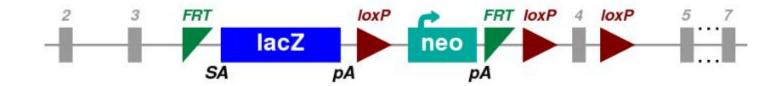
Source: https://www.informatics.jax.org

Important Information

- *Chp1* is located on Chr2. If the knockout mice are crossed with other mouse strains to obtain double homozygous mutant offspring, please avoid the situation that the second gene is 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 the existing technology level.



References



https://www.mousephenotype.org/data/genes/MGI:1927185

Mutation details: The L1L2_Bact_P cassette was inserted at position 119401802 of Chromosome 2 upstream of the critical exon(s) (Build GRCm39). The cassette is composed of an FRT site followed by lacZ sequence and a loxP site. This first loxP site is followed by a neomycin resistance gene under the control of the human beta-actin promoter, SV40 polyA, a second FRT site and a second loxP site. A third loxP site is inserted downstream of the targeted exon(s) at position 119402773 The critical exon(s) is/are thus flanked by loxP sites. A "conditional ready" (floxed) allele can be created by flp recombinase expression in mice carrying this allele. Subsequent cre expression results in a knockout mouse. If cre expression occurs without flp expression, a reporter knockout mouse will be created. Further information on targeting strategies used for this and other IKMC alleles can be found at http://www.informatics.jax.org/mgihome/nomen/IKMC_schematics.shtml (*J:157065*)

https://www.informatics.jax.org/allele/MGI:4842153

