

# Hapln1 Cas9-CKO Strategy

**Designer: Yupeng Yang** 

Reviewer: Wenjing Li

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# **Project Overview**



**Project Name** 

Hapln1

**Project type** 

Cas9-CKO

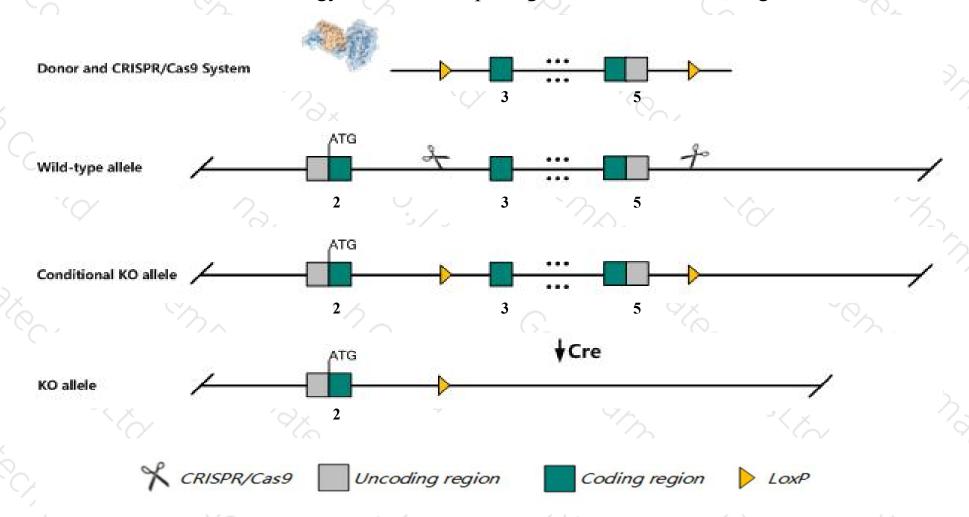
Strain background

C57BL/6JGpt

### Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Hapln1* gene. The schematic diagram is as follows:



### **Technical routes**



- ➤ The *Hapln1* gene has 2 transcripts. According to the structure of *Hapln1* gene, exon3-exon5 of *Hapln1*201(ENSMUST00000022108.8) transcript is recommended as the knockout region. The region contains 965bp coding sequence.

  Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Hapln1* 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.

### **Notice**



- > According to the existing MGI data, homozygotes for a targeted null mutation exhibit defects in cartilage development and delayed bone formation with short limbs and craniofacial anomalies. Mutants usually die as neonates due to respiratory failure, but some survive and develop dwarfism.
- > The *Hapln1* gene is located on the Chr13. 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)



#### Hapln1 hyaluronan and proteoglycan link protein 1 [Mus musculus (house mouse)]

Gene ID: 12950, updated on 13-Mar-2020

#### Summary

☆ ?

Official Symbol Hapln1 provided by MGI

Official Full Name hyaluronan and proteoglycan link protein 1 provided by MGI

Primary source MGI:MGI:1337006

See related Ensembl: ENSMUSG00000021613

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 BB099155, CLP, Crtl1, Crtl1l, LP, LP-1

Expression Biased expression in limb E14.5 (RPKM 38.8), CNS E14 (RPKM 5.4) and 4 other tissuesSee more

Orthologs <u>human</u> all

# Transcript information (Ensembl)



The gene has 2 transcripts, all transcripts are shown below:

all the							
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Hapln1-201	ENSMUST00000022108.8	5715	356aa	Protein coding	CCDS26671	Q9QUP5	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Hapln1-202	ENSMUST00000225678.1	464	No protein	Retained intron	-		

The strategy is based on the design of *Hapln1-201* transcript, the transcription is shown below:

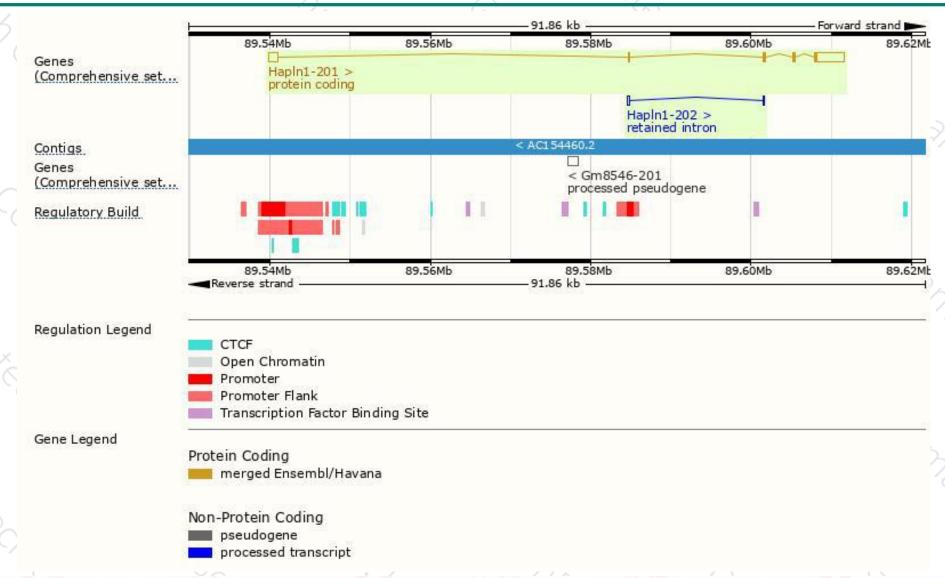
T1.86 kb

Forward strand

HapIn1-201 > protein coding

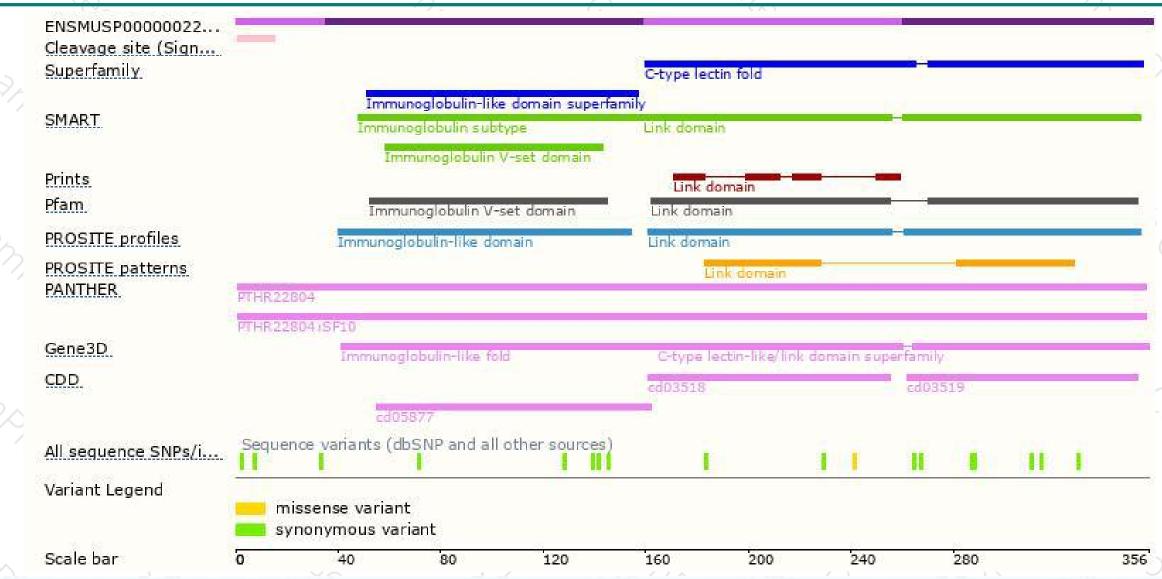
### Genomic location distribution





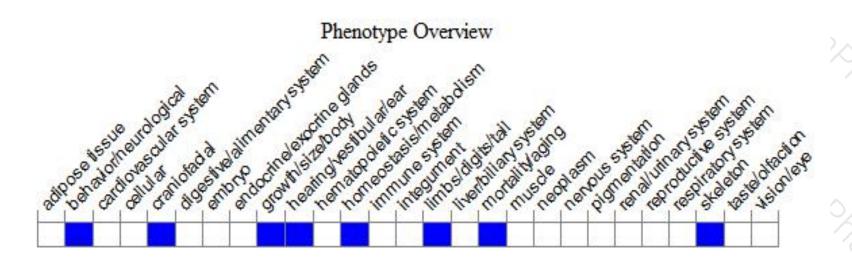
### Protein domain





## 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,homozygotes for a targeted null mutation exhibit defects in cartilage development and delayed bone formation with short limbs and craniofacial anomalies. Mutants usually die as neonates due to respiratory failure, but some survive and develop dwarfism.



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





