Lhx5 Cas9-CKO Strategy

Designer: Xueting Zhang

Reviewer: Daohua Xu

Design Date: 2020-7-16

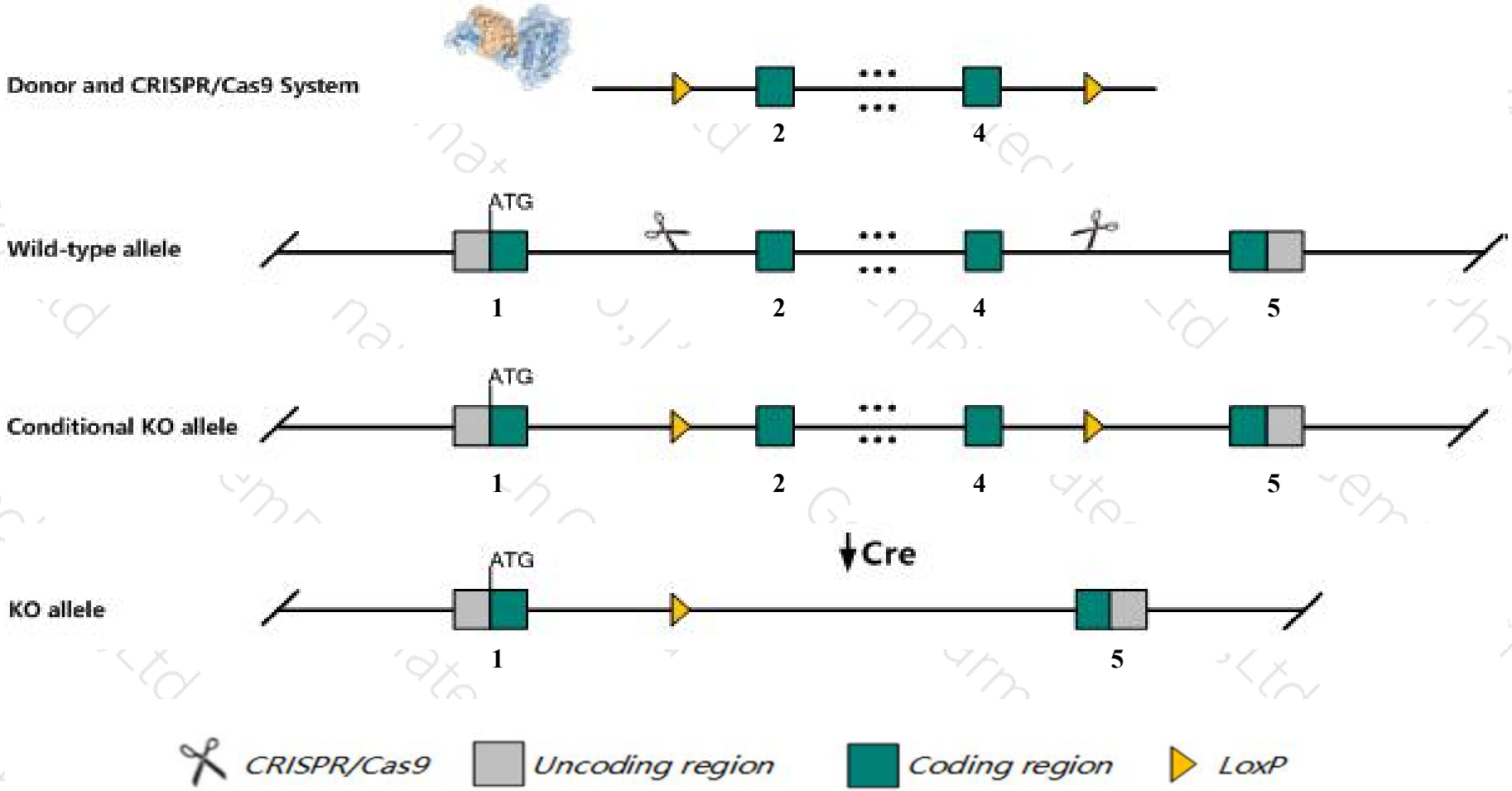
集萃药康 GemPharmatech
GemPharmatech Co.,Ltd

Project Overview

Project Name	***Lhx5***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Lhx5* gene. The schematic diagram is as follows:



Technical routes

➢ The *Lhx5* gene has 1 transcript. According to the structure of *Lhx5* gene, exon2-exon4 of *Lhx5-201*(ENSMUST00000031591.9) transcript is recommended as the knockout region.The region contains 668bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Lhx5* 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,most mice homozygous for a null mutation display defective hippocampal development and die within a few days after birth. Postmitotic hippocampal cells are unable to differentiate properly and migrate to correct positions, resulting in structural anomalies of the Ammon's horn and the dentate gyrus.

➢ The floxed region is near to the N-terminal of *Gm27199* gene,this strategy may influence the regulatory function of the N-terminal of *Gm27199* gene.

➢ The *Lhx5* gene is located on the Chr5. 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）

Lhx5 LIM homeobox protein 5 [Mus musculus (house mouse)]

Gene ID: 16873, updated on 13-Mar-2020

Summary

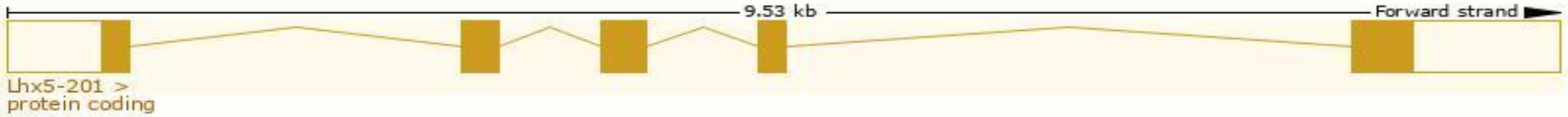
Official Symbol	Lhx5 provided by MGI
Official Full Name	LIM homeobox protein 5 provided byMGI
Primary source	MGI:MGI:107792
See related	Ensembl:ENSMUSG00000029595
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Lim2
Expression	Biased expression in CNS E11.5 (RPKM 6.4), whole brain E14.5 (RPKM 6.0) and 5 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

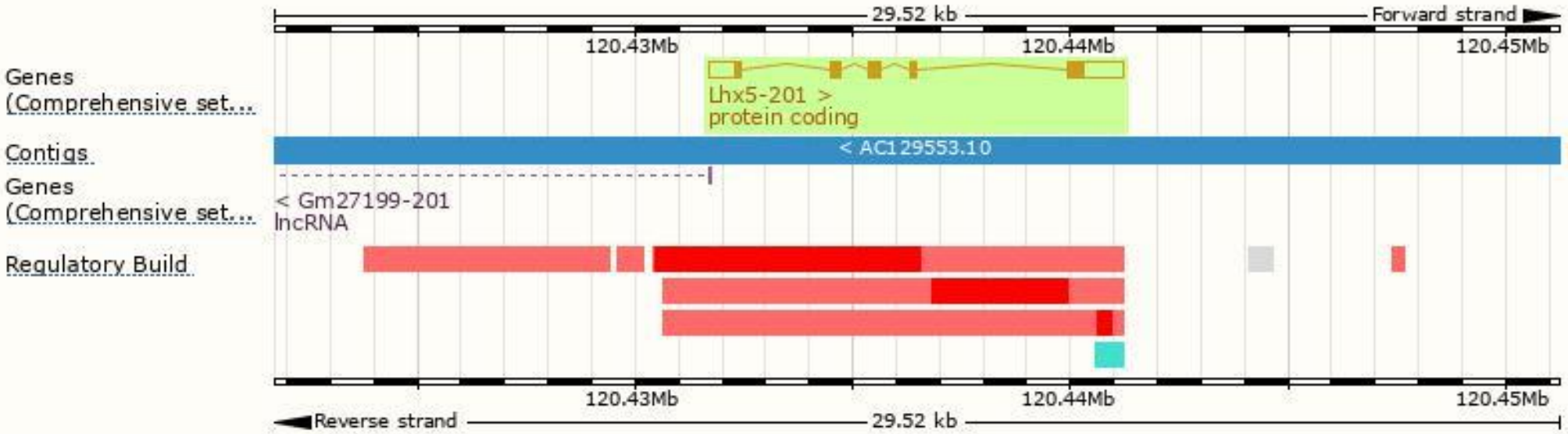
The gene has 1 transcript,and the transcript is shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Lhx5-201	ENSMUST00000031591.9	2689	402aa	Protein coding	CCDS19617	P61375 Q543P4	TSL:1 GENCODE basic APPRIS P1

The strategy is based on the design of *Lhx5-201* transcript,the transcription is shown below:



Genomic location distribution



Regulation Legend

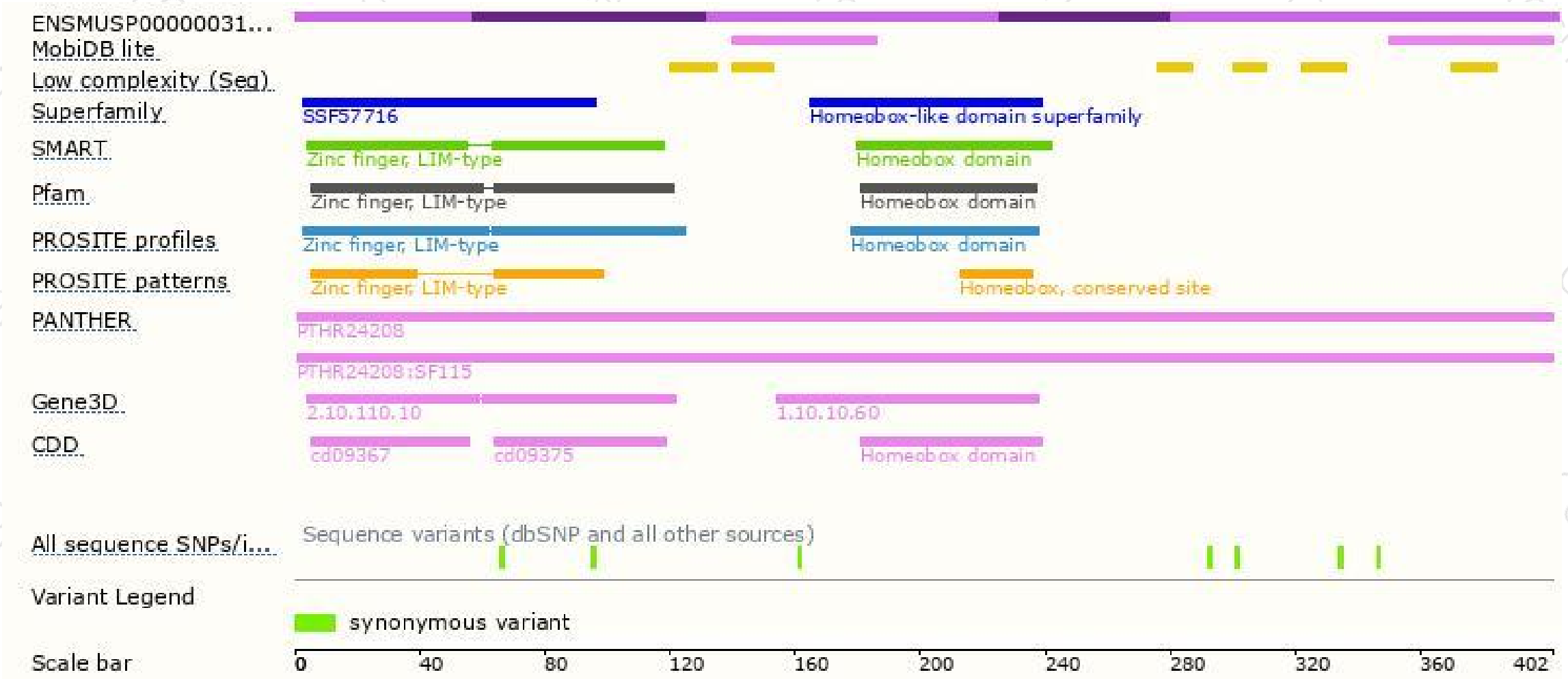
- CTCF
- Open Chromatin
- Promoter
- Promoter Flank

Gene Legend

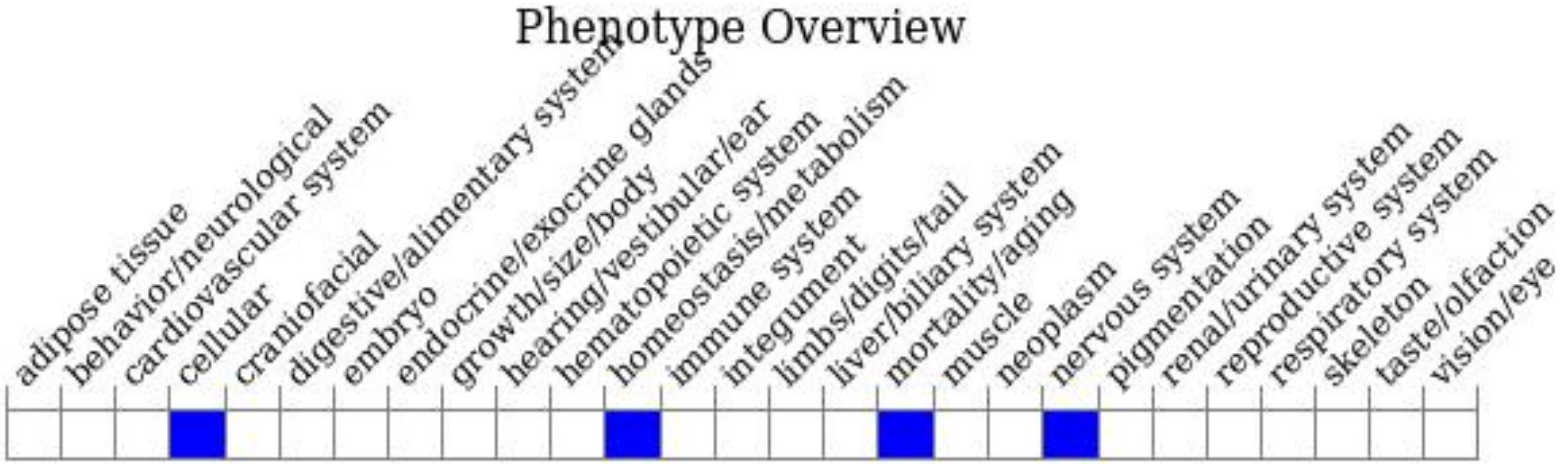
Protein Coding
- merged Ensembl/Havana

Non-Protein Coding
- RNA gene

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,most mice homozygous for a null mutation display defective hippocampal development and die within a few days after birth. Postmitotic hippocampal cells are unable to differentiate properly and migrate to correct positions, resulting in structural anomalies of the Ammon's horn and the dentate gyrus.

集萃药康
GemPharmatech

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

Tel: 400-9660890

集萃药康生物科技
GemPharmatech Co.,Ltd

GemPharmatech Co.,Ltd