Sp8 Cas9-CKO Strategy

Designer: Huimin Su

Reviewer: Ruirui Zhang

Design Date: 2020/2/11

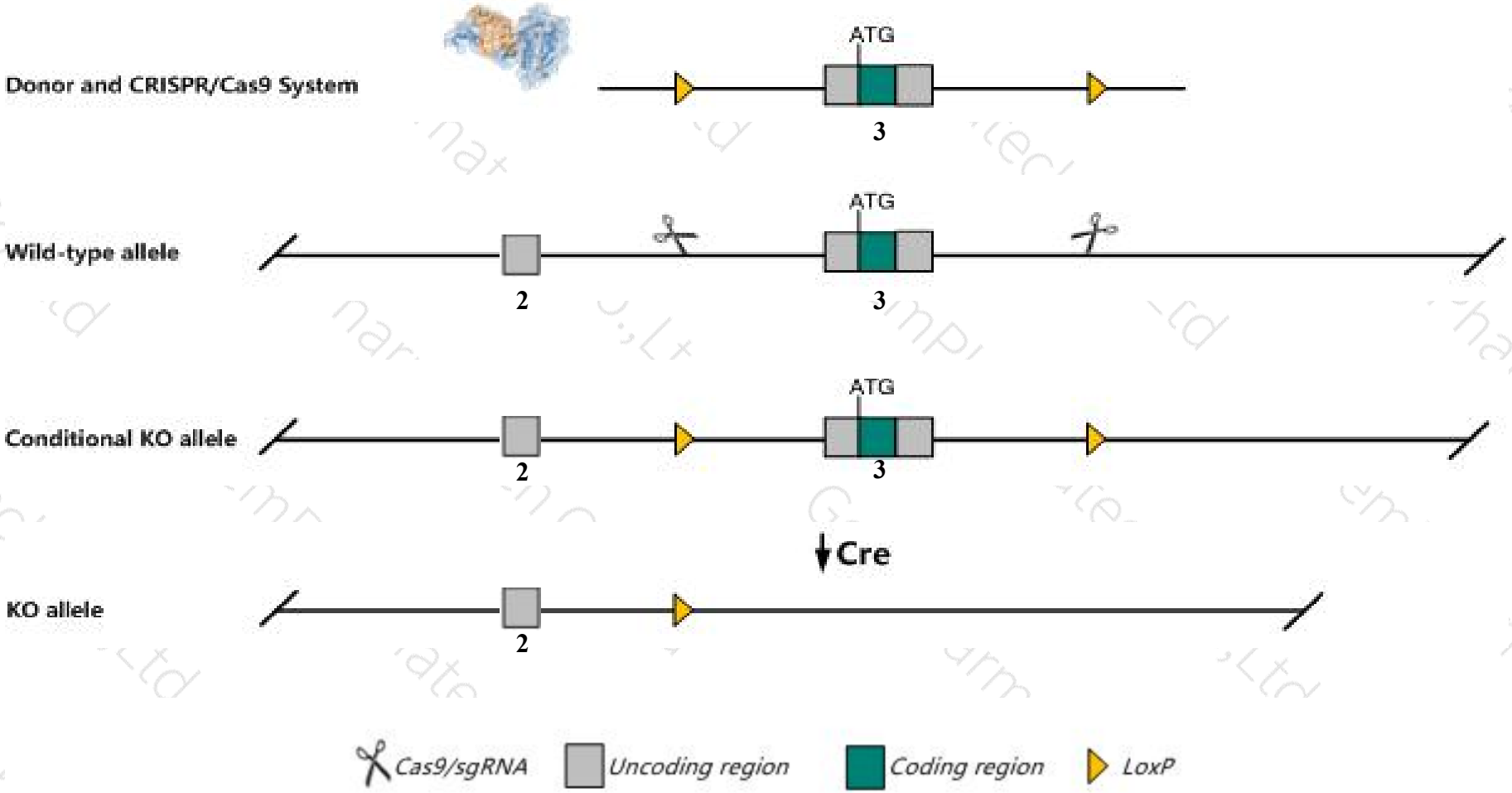
Project Overview

Project Name	***Sp8***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

集萃药康 GemPharmatech

This model will use CRISPR/Cas9 technology to edit the *Sp8* gene. The schematic diagram is as follows:



Technical routes

- The *Sp8* gene has 2 transcripts. According to the structure of *Sp8* gene, exon3 of *Sp8-201* (ENSMUST00000063918.3) transcript is recommended as the knockout region.The region contains all of the coding sequence.Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Sp8* 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,Homozygous mutant fetuses are characterized by truncated limbs, the lack of a tail, and neural tube defects.

➢ The *Sp8* gene is located on the Chr12. 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）

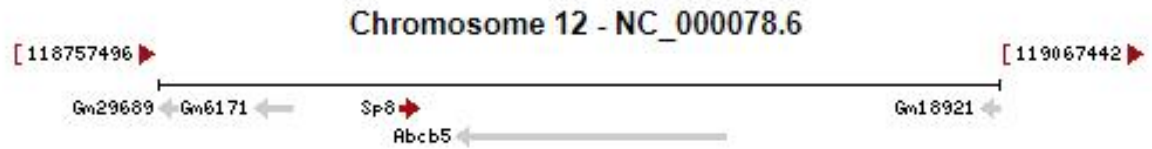
集萃药康 GemPharmatech

Sp8 trans-acting transcription factor 8 [*Mus musculus* (house mouse)]

Gene ID: 320145, updated on 28-Jan-2020

Summary

Official Symbol	Sp8 provided by MGI
Official Full Name	trans-acting transcription factor 8 provided by MGI
Primary source	MGI:MGI:2443471
See related	Ensembl:ENSMUSG00000048562
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	mBtd; D930049B17Rik
Expression	Biased expression in frontal lobe adult (RPKM 5.4), whole brain E14.5 (RPKM 1.9) and 4 other tissues See more
Orthologs	human all

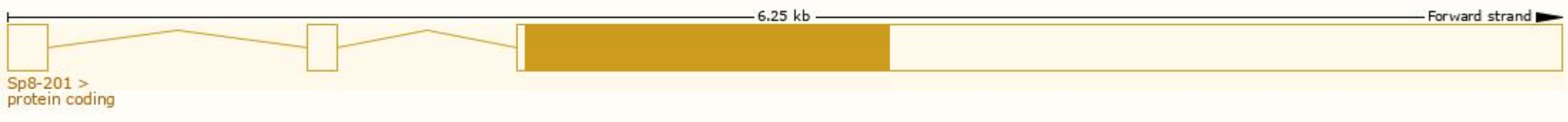


Transcript information（Ensembl）

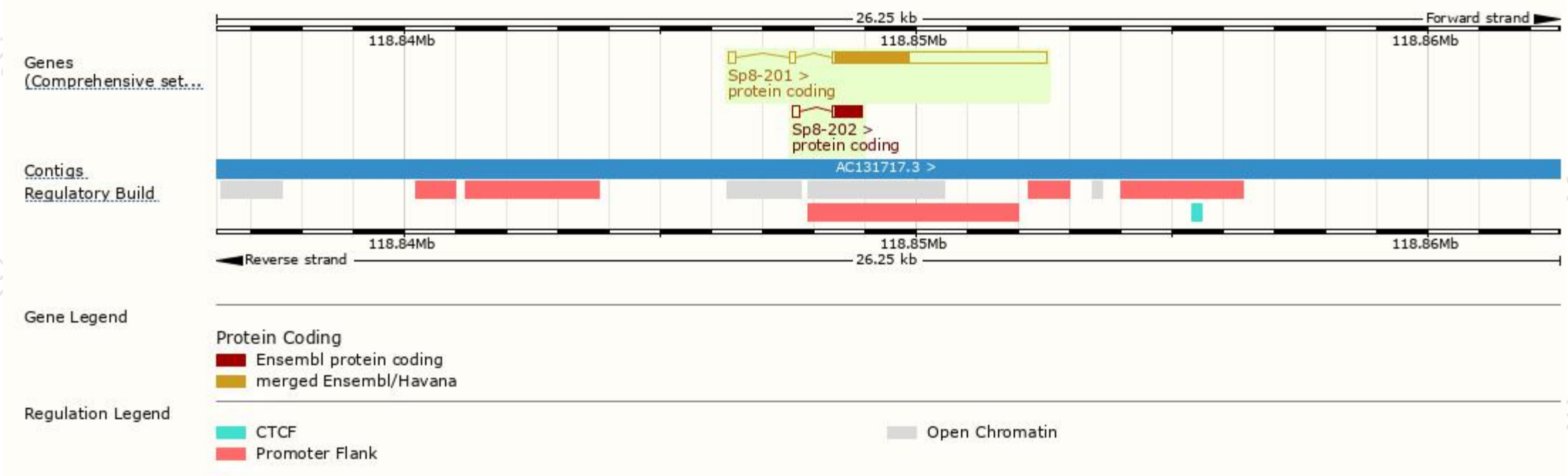
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Sp8-201	ENSMUST00000063918.3	4478	486aa	Protein coding	CCDS26213	Q5QR90 Q8BMJ8	TSL:1 GENCODE basic APPRIS P1
Sp8-202	ENSMUST00000223305.1	726	181aa	Protein coding	-	A0A1Y7VJL2	CDS 3' incomplete TSL:3

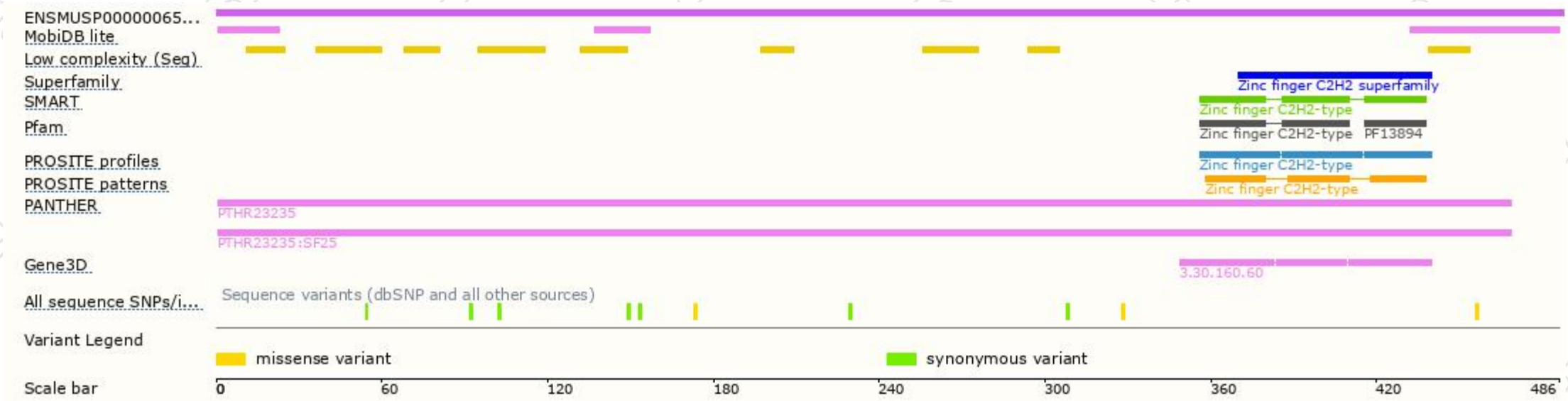
The strategy is based on the design of *Sp8-201* transcript,The transcription is shown below



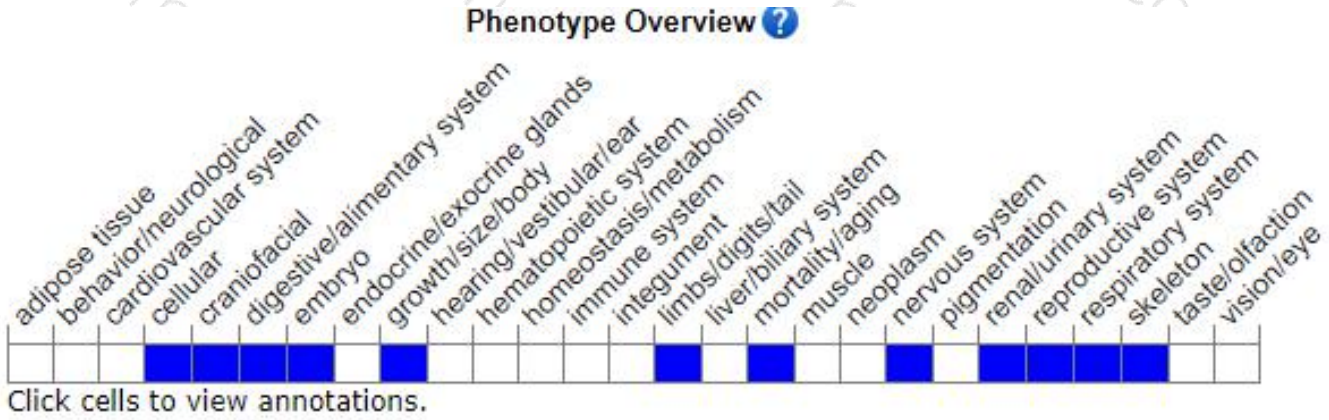
Genomic location distribution



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,Homozygous mutant fetuses are characterized by truncated limbs, the lack of a tail, and neural tube defects.

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