

***Fmn1* Cas9-CKO Strategy**

Designer:

Yang Zeng

Reviewer:

Xueting Zhang

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Project Overview

Project Name

Fmn1

Project type

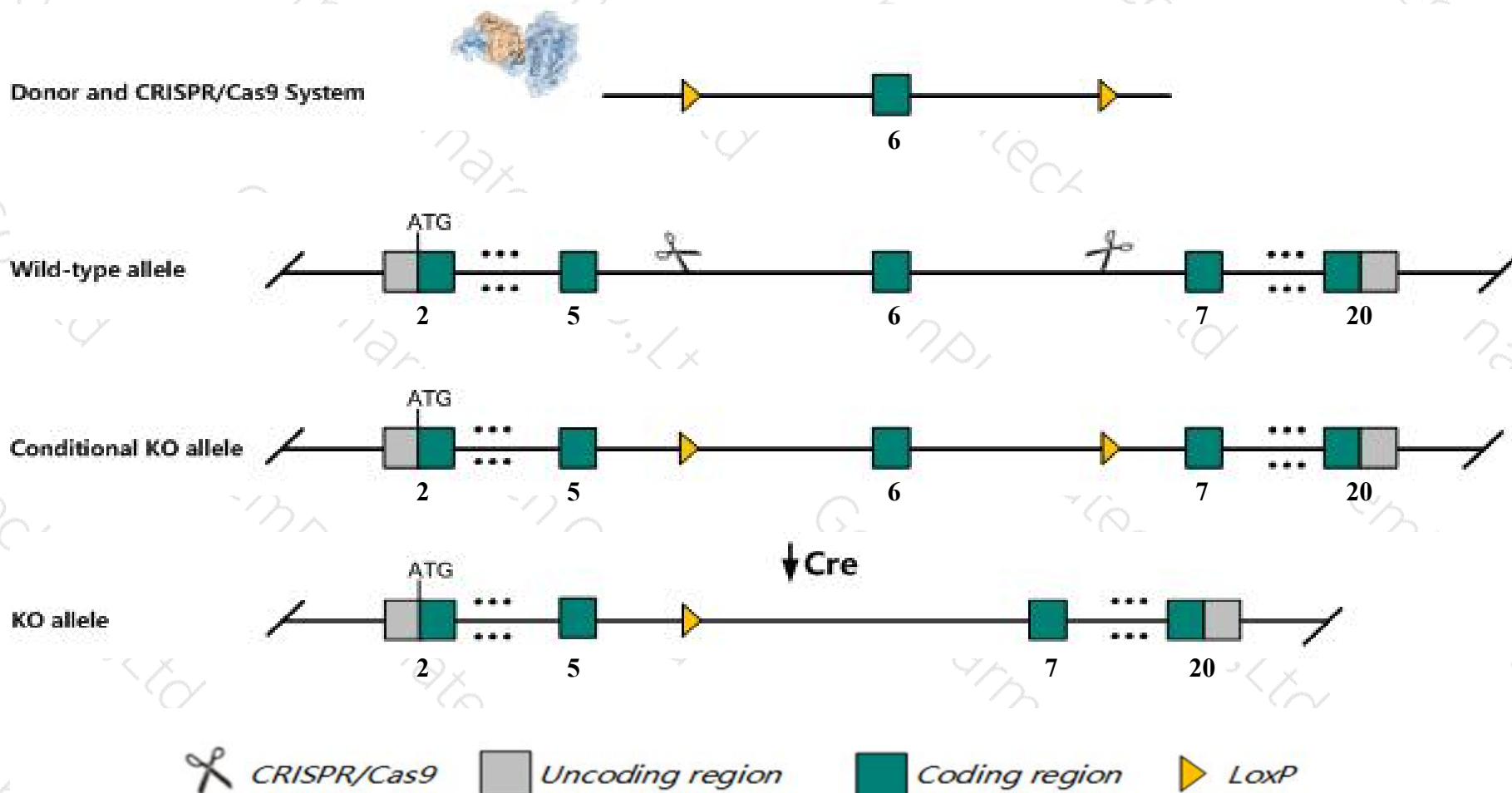
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Fmn1* gene. The schematic diagram is as follows:



Technical routes

- The *Fmn1* gene has 9 transcripts. According to the structure of *Fmn1* gene, exon6 of *Fmn1*-203 (ENSMUST00000102547.9) transcript is recommended as the knockout region. The region contains 791bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Fmn1* 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.

- According to the existing MGI data, Homozygotes for spontaneous, irradiation-induced, and transgene-insertional mutations show severe syndactyly and oligodactyly of the feet, abnormal long bones (including radius-ulna fusions), and reduced or absent kidneys. Many mutants survive and breed.
- Transcript *Fmn1*-204/205/206/207/208/209 lncRNA may not be affected.
- The *Fmn1* gene is located on the Chr2. 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.

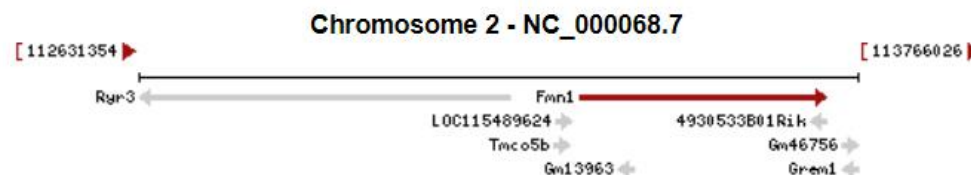
Gene information (NCBI)

Fmn1 formin 1 [*Mus musculus* (house mouse)]

Gene ID: 14260, updated on 10-Oct-2019

Summary

Official Symbol	Fmn1 provided by MGI
Official Full Name	formin 1 provided by MGI
Primary source	MGI:MGI:101815
See related	Ensembl:ENSMUSG00000044042
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	Id; Fmn; formin-1
Expression	Broad expression in testis adult (RPKM 2.9), kidney adult (RPKM 1.9) and 19 other tissues See more
Orthologs	human all

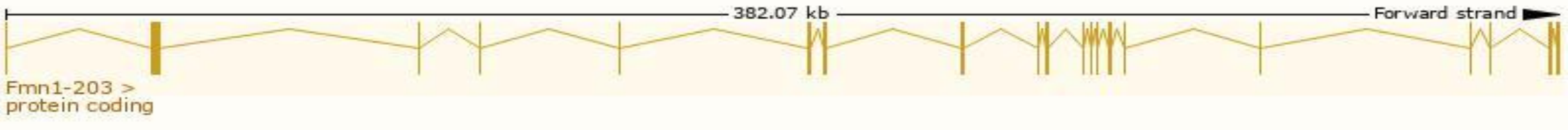


Transcript information (Ensembl)

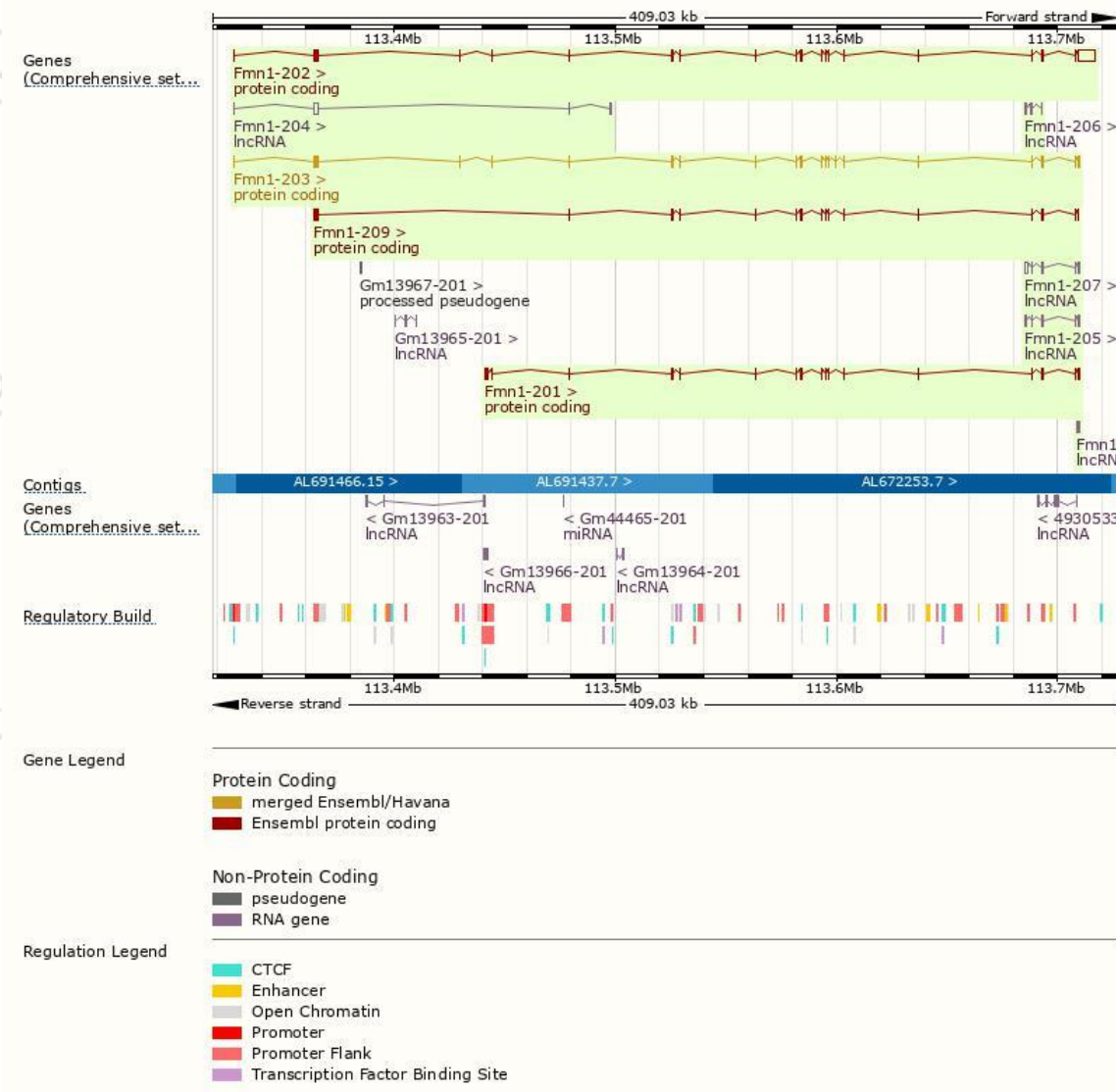
The gene has 9 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Translation ID	Biotype	CCDS	UniProt	Flags
Fmn1-203	ENSMUST00000102547.9	4967	1466aa	ENSMUSP00000099606.3	Protein coding	CCDS16559	Q05860	TSL:1 GENCODE basic
Fmn1-210	ENSMUST00000161731.4	4262	1332aa	ENSMUSP00000125052.1	Protein coding	CCDS71112	E9Q7P6	TSL:1 GENCODE basic APPRIS P4
Fmn1-201	ENSMUST00000081349.8	4235	1204aa	ENSMUSP00000080093.6	Protein coding	CCDS71113	Q05860	TSL:1 GENCODE basic APPRIS ALT2
Fmn1-202	ENSMUST00000099576.8	11817	1430aa	ENSMUSP00000097171.2	Protein coding	-	Q05860	TSL:5 GENCODE basic APPRIS ALT2
Fmn1-211	ENSMUST00000238883.1	2644	850aa	ENSMUSP00000159010.1	Protein coding	-	-	GENCODE basic APPRIS ALT2
Fmn1-204	ENSMUST00000110954.6	2384	No protein	-	lncRNA	-	-	TSL:1
Fmn1-207	ENSMUST00000152255.1	2097	No protein	-	lncRNA	-	-	TSL:1
Fmn1-205	ENSMUST00000145891.7	719	No protein	-	lncRNA	-	-	TSL:2
Fmn1-209	ENSMUST00000154834.1	576	No protein	-	lncRNA	-	-	TSL:3
Fmn1-208	ENSMUST00000153151.1	474	No protein	-	lncRNA	-	-	TSL:1
Fmn1-206	ENSMUST00000150510.7	385	No protein	-	lncRNA	-	-	TSL:5

The strategy is based on the design of *Fmn1-203* transcript,The transcription is shown below



Genomic location distribution

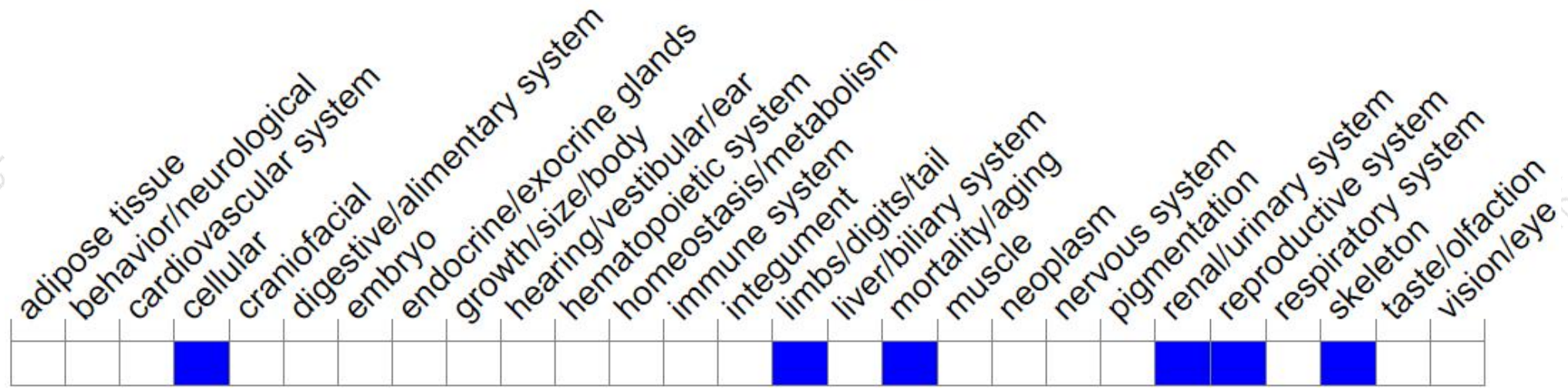


Protein domain



Mouse phenotype description(MGI)

Phenotype Overview ?



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.

Tel: 400-9660890

