

Mfap3 Cas9-CKO Strategy

Designer: Jia Yu

Reviewer: Xiaojing Li

Design Date: 2020-10-12

Project Overview



Project Name Mfap3

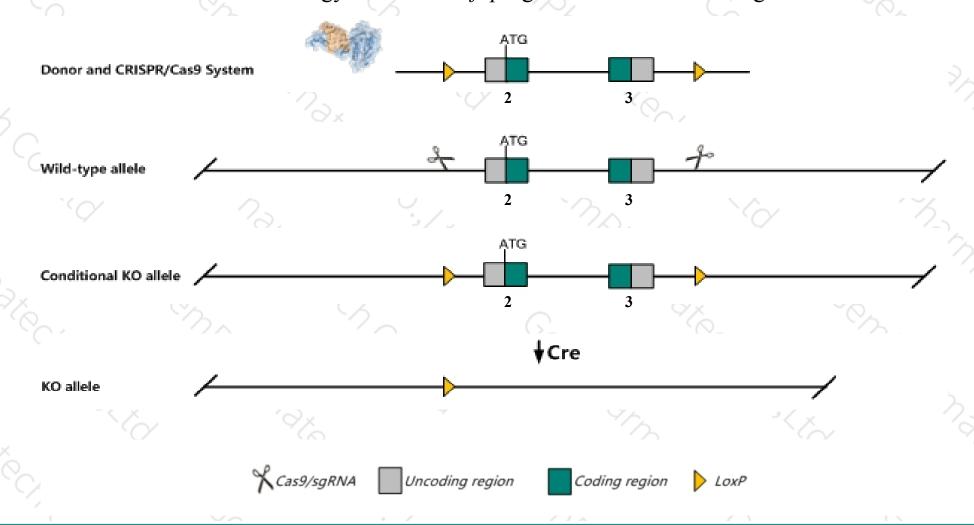
Project type Cas9-CKO

Strain background C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Mfap3* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Mfap3* gene has 4 transcripts. According to the structure of *Mfap3* gene, exon2-exon3 of *Mfap3*201(ENSMUST00000020830.13) 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 *Mfap3* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA 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 was 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



- > The *Mfap3* gene is located on the Chr11. 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)



Mfap3 microfibrillar-associated protein 3 [Mus musculus (house mouse)]

Gene ID: 216760, updated on 13-Mar-2020

Summary

☆ ?

Official Symbol Mfap3 provided by MGI

Official Full Name microfibrillar-associated protein 3 provided by MGI

Primary source MGI:MGI:1924068

See related Ensembl:ENSMUSG00000020522

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 2610509F16Rik, 2700079M14Rik, AW536327

Expression Ubiquitous expression in whole brain E14.5 (RPKM 10.8), CNS E14 (RPKM 10.2) and 28 other tissuesSee more

Orthologs <u>human</u> <u>all</u>

Transcript information (Ensembl)



The gene has 4 transcripts, all transcripts are shown below:

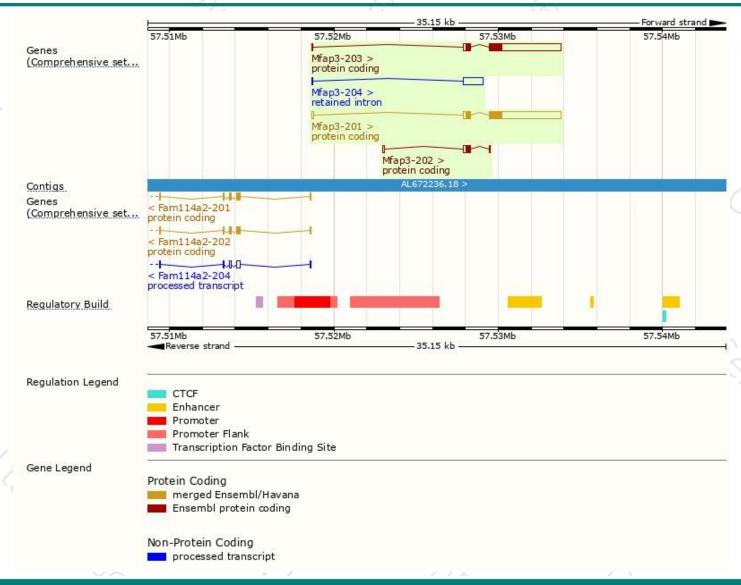
| Name | Transcript ID | bp | Protein | Biotype | CCDS | UniProt | Flags |
|-----------|-----------------------|------|--------------|-----------------|-----------|------------|-------------------------------|
| Mfap3-201 | ENSMUST00000020830.13 | 4839 | <u>349aa</u> | Protein coding | CCDS24718 | A0A0R4J1C7 | TSL:1 GENCODE basic APPRIS P1 |
| Mfap3-203 | ENSMUST00000108849.7 | 4827 | <u>349aa</u> | Protein coding | CCDS24718 | A0A0R4J1C7 | TSL:1 GENCODE basic APPRIS P1 |
| Mfap3-202 | ENSMUST00000108848.1 | 627 | <u>110aa</u> | Protein coding | - | D3Z5B4 | CDS 3' incomplete TSL:3 |
| Mfap3-204 | ENSMUST00000128971.1 | 1258 | No protein | Retained intron | - | - | TSL:1 |

The strategy is based on the design of *Mfap3-201* transcript, the transcription is shown below:



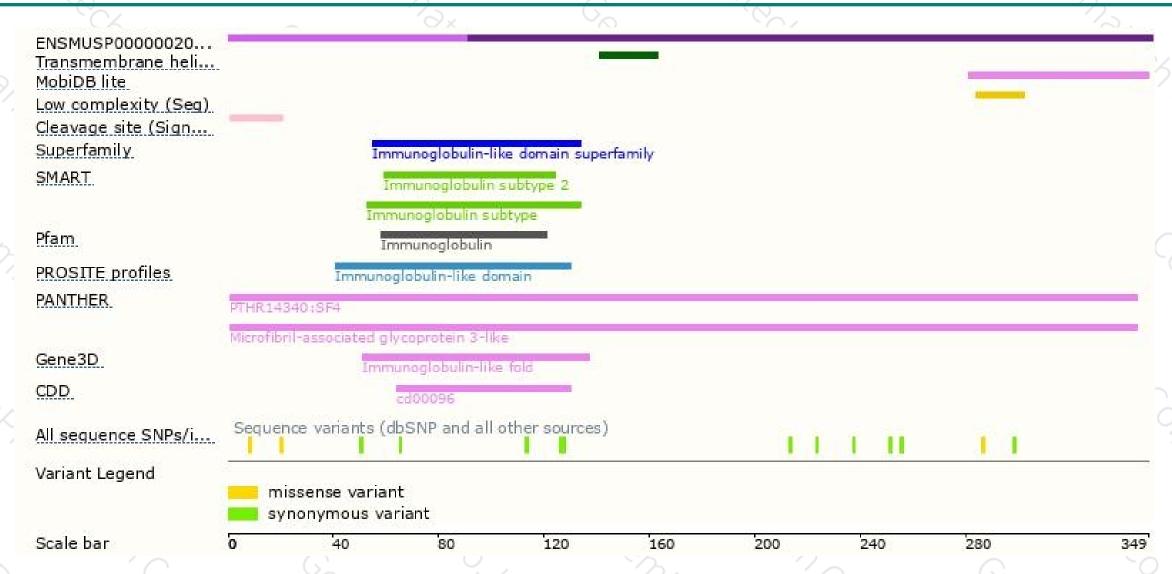
Genomic location distribution





Protein domain







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

Tel: 025-5864 1534





