

Marchf8 Cas9-CKO Strategy

Designer: Huimin Su

Reviewer: Ruiuri Zhang

Design Date: 2020-8-10

Project Overview

Project Name

Marchf8

Project type

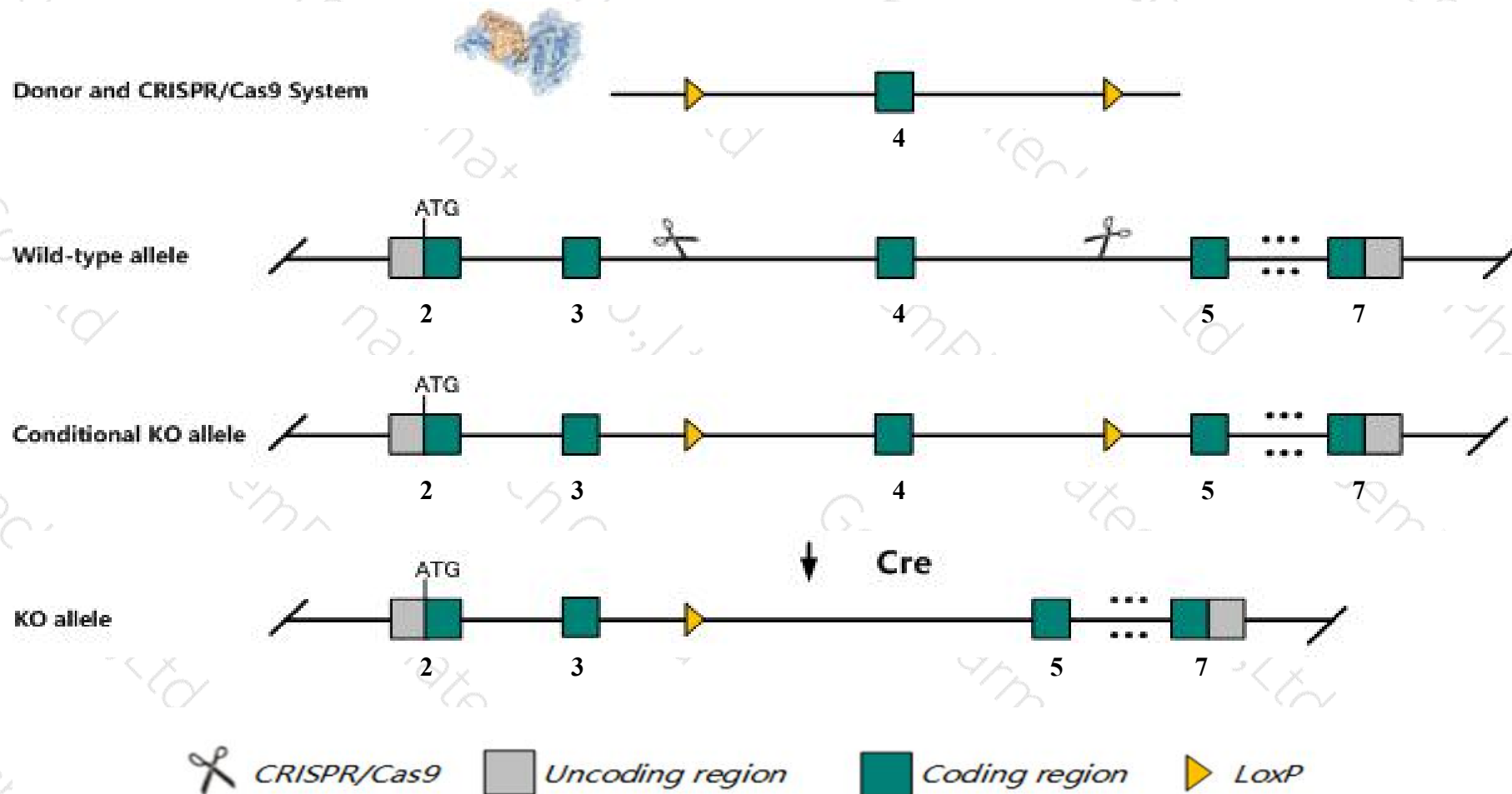
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Marchf8* gene. The schematic diagram is as follows:



- The *Marchf8* gene has 16 transcripts. According to the structure of *Marchf8* gene, exon4 of *March8-202*(ENSMUST00000101032.9) transcript is recommended as the knockout region. The region contains 86bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Marchf8* 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, mice homozygous for a knock-out allele exhibit a normal CD4⁺ T cell compartment in the thymus.
- Transcripts *Marchf8-212*, *Marchf8-205* and *Marchf8-207* are incomplete, so the effect on them are unknown.
- The *Marchf8* gene is located on the Chr6. 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)

Marchf8 membrane associated ring-CH-type finger 8 [Mus musculus (house mouse)]

Gene ID: 71779, updated on 13-Mar-2020

Summary



Official Symbol Marchf8 provided by [MGI](#)

Official Full Name membrane associated ring-CH-type finger 8 provided by [MGI](#)

Primary source [MGI:MGI:1919029](#)

See related [Ensembl:ENSMUSG00000025702](#)

Gene type protein coding

RefSeq status REVIEWED

Organism [Mus musculus](#)

Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus

Also known as 1300017E09Rik, MARCH-VIII, March8, Mir

Summary The protein encoded by this gene is a member of the membrane-associated really interesting new gene-CH family of proteins. These proteins are E3 ubiquitin-protein ligases that modulate antigen presentation by downregulating major histocompatibility complex class II surface expression through endocytosis. The transcript is primarily expressed by dendritic cells and macrophages. Overexpression of this gene in antigen presenting cells results in immune defective phenotypes, including resistance to autoimmune disease onset. Alternative splicing results in multiple transcript variants. [provided by RefSeq, Oct 2014]

Expression Ubiquitous expression in kidney adult (RPKM 18.7), liver E14 (RPKM 18.4) and 28 other tissues [See more](#)

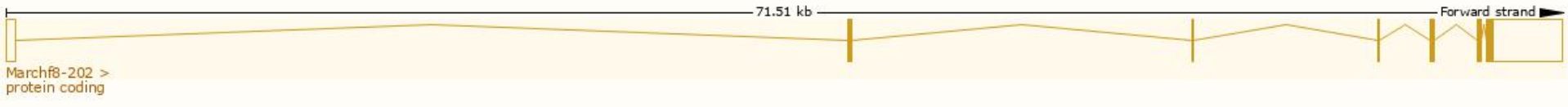
Orthologs [human](#) [all](#)

Transcript information (Ensembl)

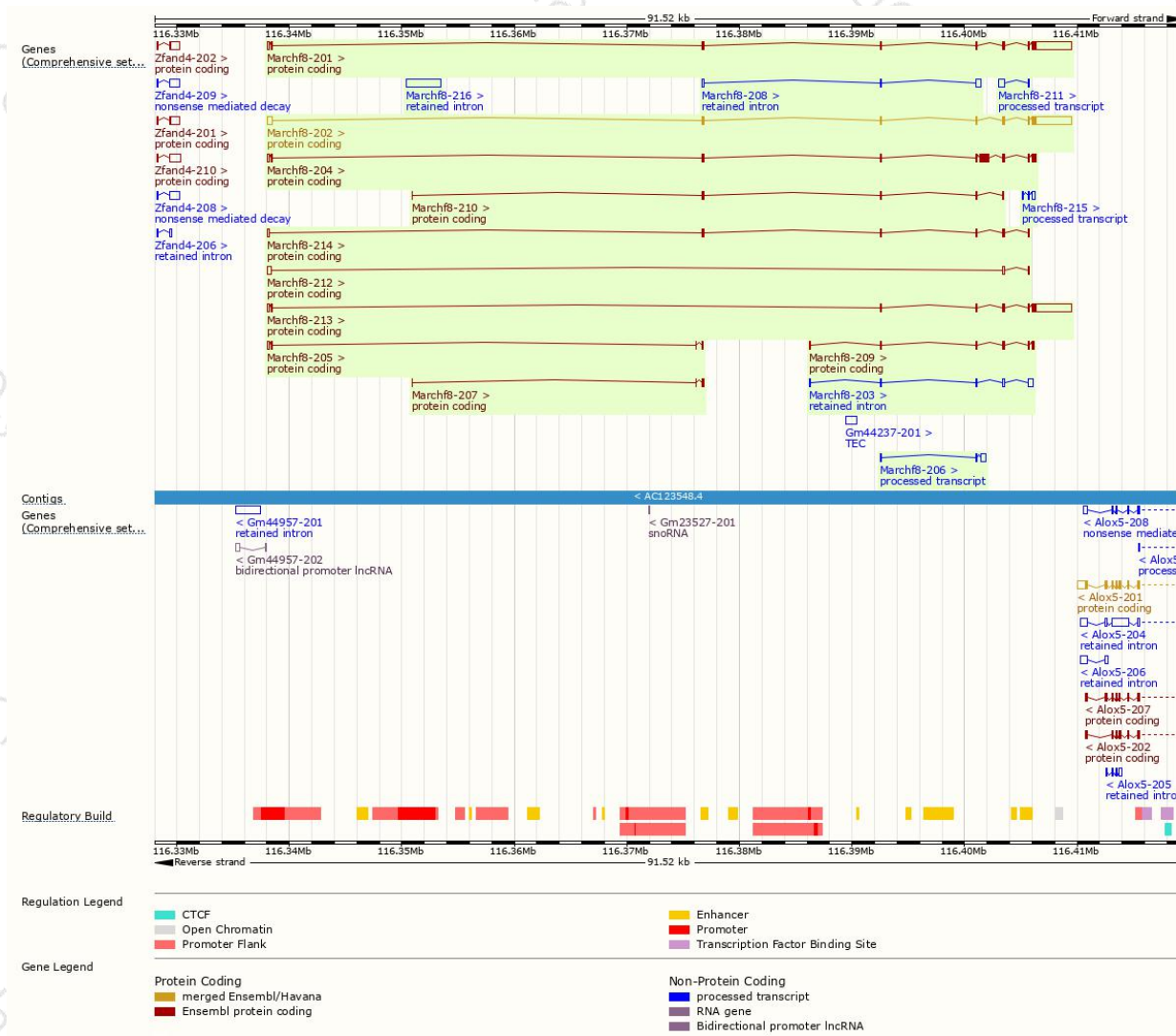
The gene has 16 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Marchf8-205	ENSMUST00000126376.7	407	10aa	Protein coding	-	A0A0N4SVW5	CDS 3' incomplete TSL:3
Marchf8-207	ENSMUST00000129170.7	235	31aa	Protein coding	-	A0A0N4SVD4	CDS 3' incomplete TSL:3
Marchf8-212	ENSMUST00000203116.1	648	45aa	Protein coding	-	A0A0N4SVT6	CDS 3' incomplete TSL:5
Marchf8-210	ENSMUST00000140884.7	382	79aa	Protein coding	-	A0A0N4SVD9	CDS 3' incomplete TSL:5
Marchf8-214	ENSMUST00000204657.2	780	176aa	Protein coding	-	A0A0N4SW32	CDS 3' incomplete TSL:3
Marchf8-209	ENSMUST00000135901.1	747	232aa	Protein coding	-	F6THH9	CDS 3' incomplete TSL:3
Marchf8-213	ENSMUST00000203193.2	4245	247aa	Protein coding	CCDS85138	A0A0N4SVK0	TSL:1 GENCODE basic APPRIS ALT2
Marchf8-202	ENSMUST00000101032.9	4611	286aa	Protein coding	CCDS20451	Q9DBD2	TSL:1 GENCODE basic APPRIS P3
Marchf8-201	ENSMUST00000079012.12	4450	286aa	Protein coding	CCDS20451	Q9DBD2	TSL:1 GENCODE basic APPRIS P3
Marchf8-204	ENSMUST00000123405.7	2102	568aa	Protein coding	-	A0A0N4SV35	TSL:5 GENCODE basic APPRIS ALT2
Marchf8-211	ENSMUST00000156014.3	757	No protein	Processed transcript	-	-	TSL:5
Marchf8-206	ENSMUST00000127992.6	579	No protein	Processed transcript	-	-	TSL:5
Marchf8-215	ENSMUST00000204918.1	485	No protein	Processed transcript	-	-	TSL:5
Marchf8-216	ENSMUST00000205066.1	3107	No protein	Retained intron	-	-	TSL:NA
Marchf8-203	ENSMUST00000123246.7	873	No protein	Retained intron	-	-	TSL:3
Marchf8-208	ENSMUST00000130445.7	617	No protein	Retained intron	-	-	TSL:3

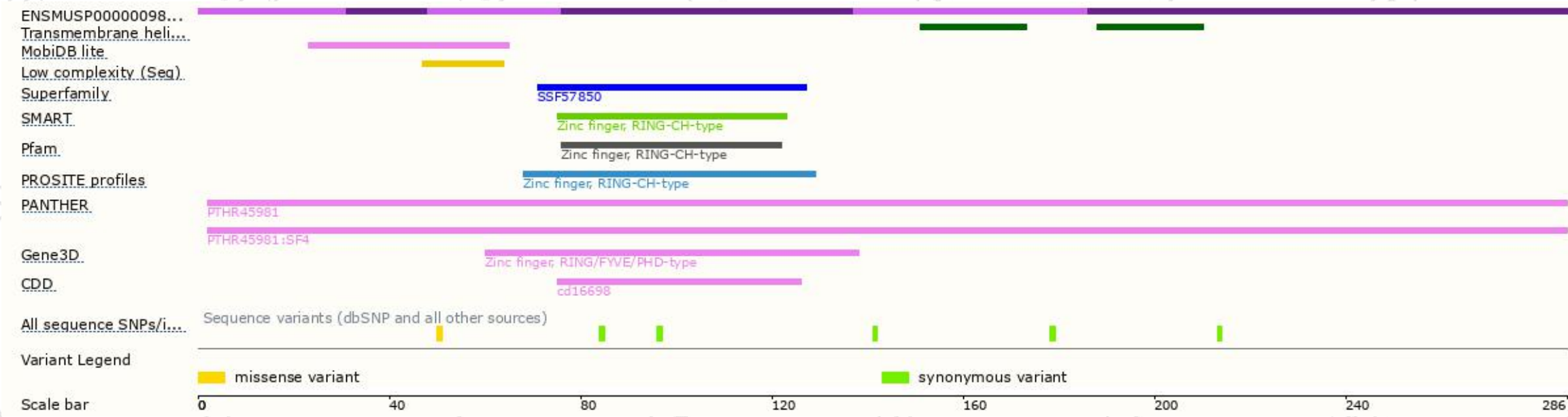
The strategy is based on the design of *March8-202* transcript,the transcription is shown below:



Genomic location distribution



Protein domain



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

Tel: 400-9660890

