

# Scml4 Cas9-CKO Strategy

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Reviewer: Rui Xiong

**Design Date: 2021-7-6** 

## **Project Overview**



Project Name Scml4

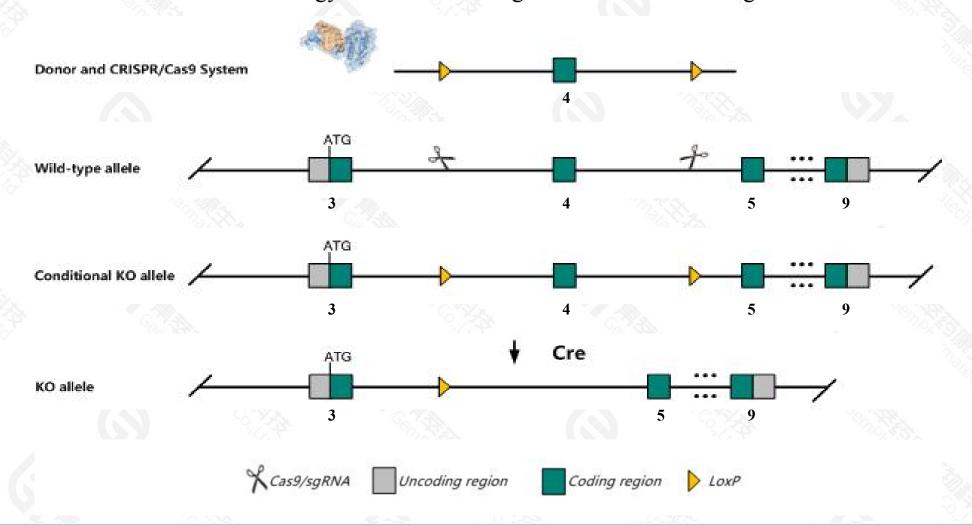
Project type Cas9-CKO

Strain background C57BL/6JGpt

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Scml4* gene. The schematic diagram is as follows:



### **Technical routes**



- > The *Scml4* gene has 9 transcripts. According to the structure of *Scml4* gene, exon4 of *Scml4*-201(ENSMUST00000063063.14) transcript is recommended as the knockout region. The region contains 130bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Scml4* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor 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 *Scml4* gene is located on the Chr10. 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)



#### Scml4 Scm polycomb group protein like 4 [Mus musculus (house mouse)]

Gene ID: 268297, updated on 17-Dec-2020

#### Summary

☆ ?

Official Symbol Scml4 provided by MGI

Official Full Name Scm polycomb group protein like 4 provided by MGI

Primary source MGI:MGI:2446140

See related Ensembl:ENSMUSG00000044770

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 9330161D17, mFLJ00197

Expression Broad expression in spleen adult (RPKM 3.2), thymus adult (RPKM 2.9) and 22 other tissuesSee more

Orthologs <u>human all</u>

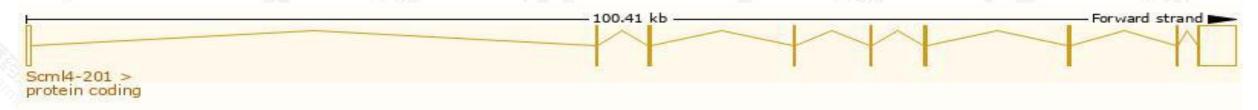
## Transcript information (Ensembl)



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

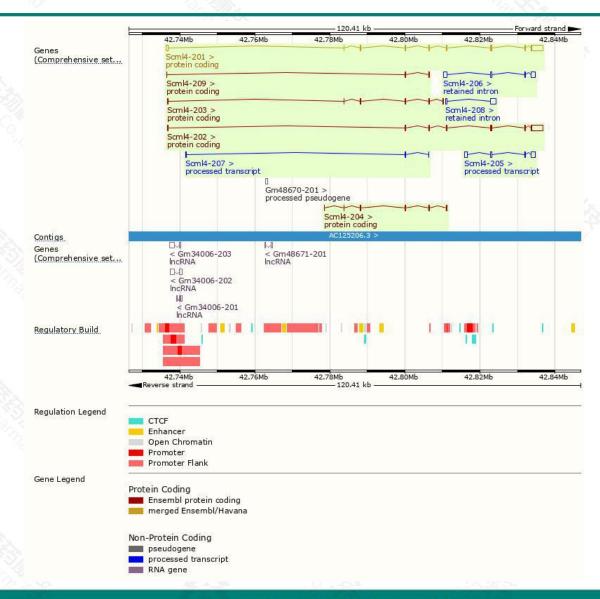
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Scml4-201	ENSMUST00000063063.14	4885	408aa	Protein coding	CCDS23816		TSL:1, GENCODE basic, APPRIS P1,
Scml4-202	ENSMUST00000105494.8	4166	350aa	Protein coding	-		TSL:1 , GENCODE basic ,
Scml4-203	ENSMUST00000105495.8	1033	<u>177aa</u>	Protein coding	826		TSL:1 , GENCODE basic ,
Scml4-204	ENSMUST00000125576.2	887	<u>224aa</u>	Protein coding			CDS 3' incomplete , TSL:3 ,
Scml4-209	ENSMUST00000157071.8	365	<u>94aa</u>	Protein coding	145		CDS 3' incomplete , TSL:3 ,
Scml4-205	ENSMUST00000127675.2	2214	No protein	Processed transcript	520		TSL:1,
Scml4-207	ENSMUST00000136841.2	349	No protein	Processed transcript	-		TSL:5,
Scml4-206	ENSMUST00000130511.8	2381	No protein	Retained intron	(20		TSL:1,
Scml4-208	ENSMUST00000153091.2	1563	No protein	Retained intron			TSL:1,

The strategy is based on the design of *Scml4-201* transcript, the transcription is shown below:



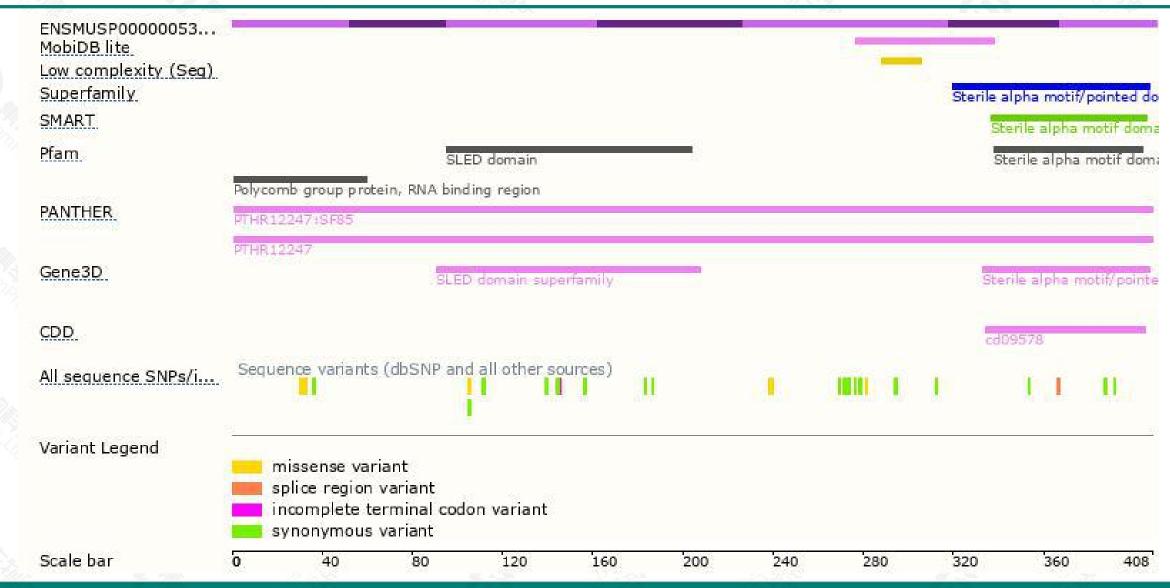
### Genomic location distribution





### Protein domain







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

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