

***Trmt44* Cas9-CKO Strategy**

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Project Overview

Project Name

Trmt44

Project type

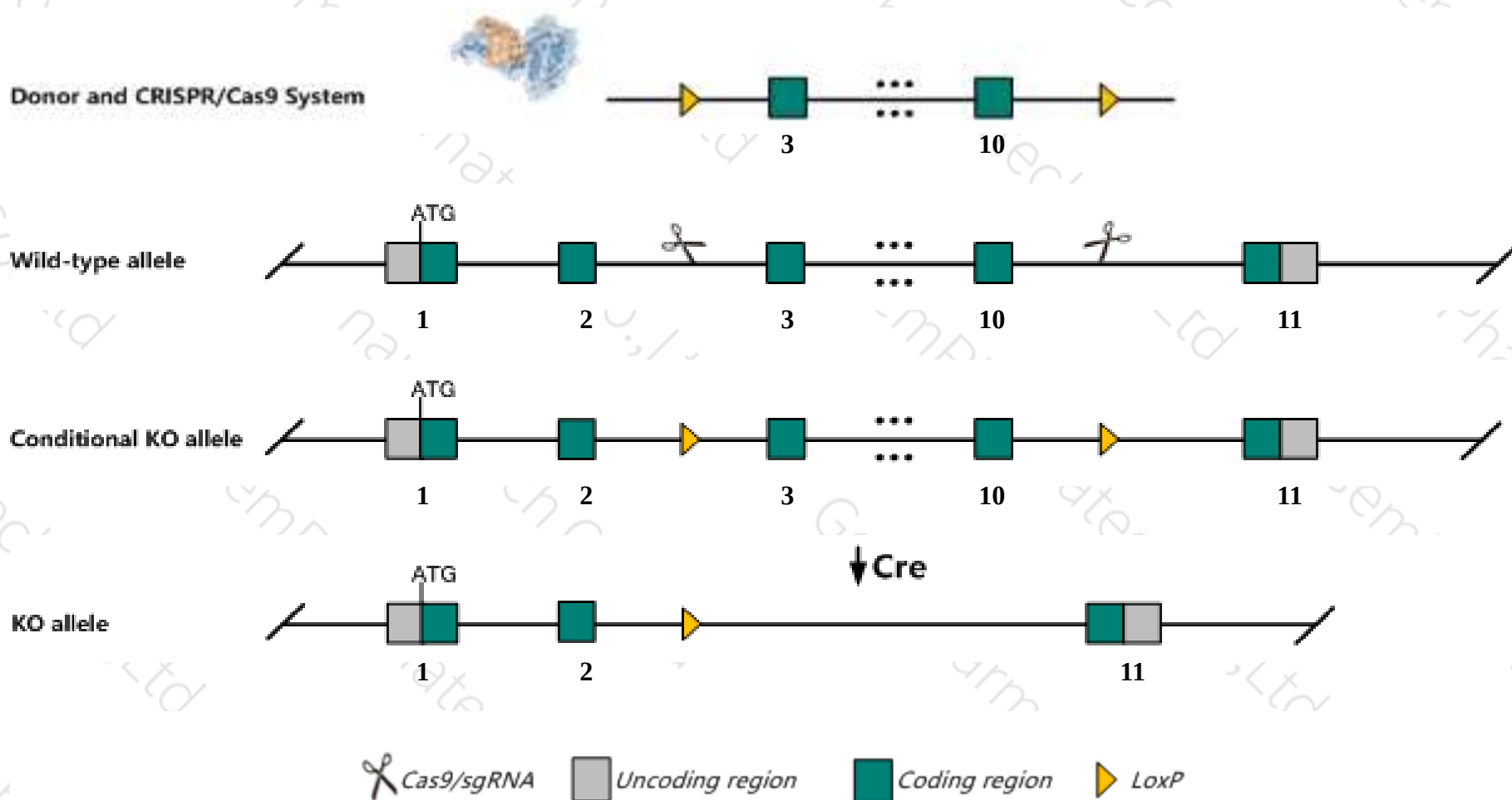
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Trmt44* gene. The schematic diagram is as follows:



- The *Trmt44* gene has 3 transcripts. According to the structure of *Trmt44* gene, exon3-exon10 of *Trmt44*-201(ENSMUST00000030980.11) transcript is recommended as the knockout region. The region contains 1310bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Trmt44* 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.

- The *Trmt44* gene is located on the Chr5. 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)

Trmt44 tRNA methyltransferase 44 [Mus musculus (house mouse)]

Gene ID: 78890, updated on 13-Mar-2020

Summary



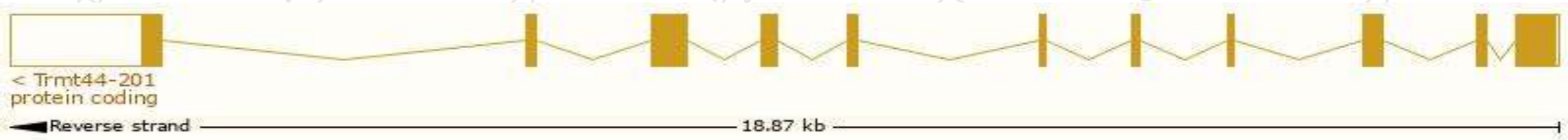
Official Symbol	Trmt44 provided by MGI
Official Full Name	tRNA methyltransferase 44 provided by MGI
Primary source	MGI:MGI:1926140
See related	Ensembl:ENSMUSG00000029097
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	2310079F23Rik, Mettl19
Expression	Ubiquitous expression in ovary adult (RPKM 3.7), thymus adult (RPKM 3.0) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

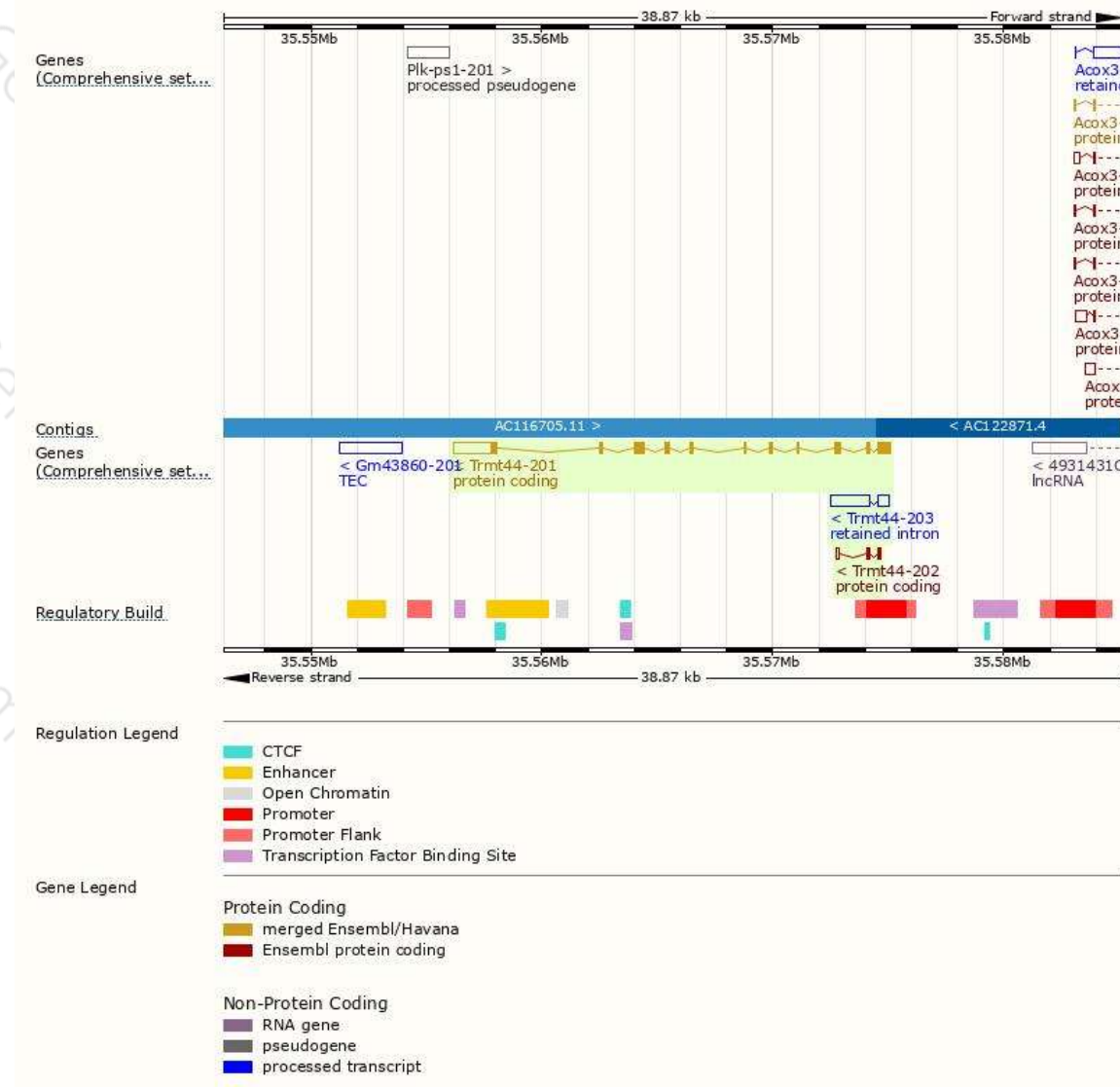
The gene has 3 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Trmt44-201	ENSMUST00000030980.11	3790	713aa	Protein coding	CCDS19230	Q9D2Q2	TSL:1 GENCODE basic APPRIS P1
Trmt44-202	ENSMUST00000128195.2	375	77aa	Protein coding	-	A0A0J9YTY4	CDS 5' incomplete TSL:2
Trmt44-203	ENSMUST00000145930.1	2192	No protein	Retained intron	-	-	TSL:1

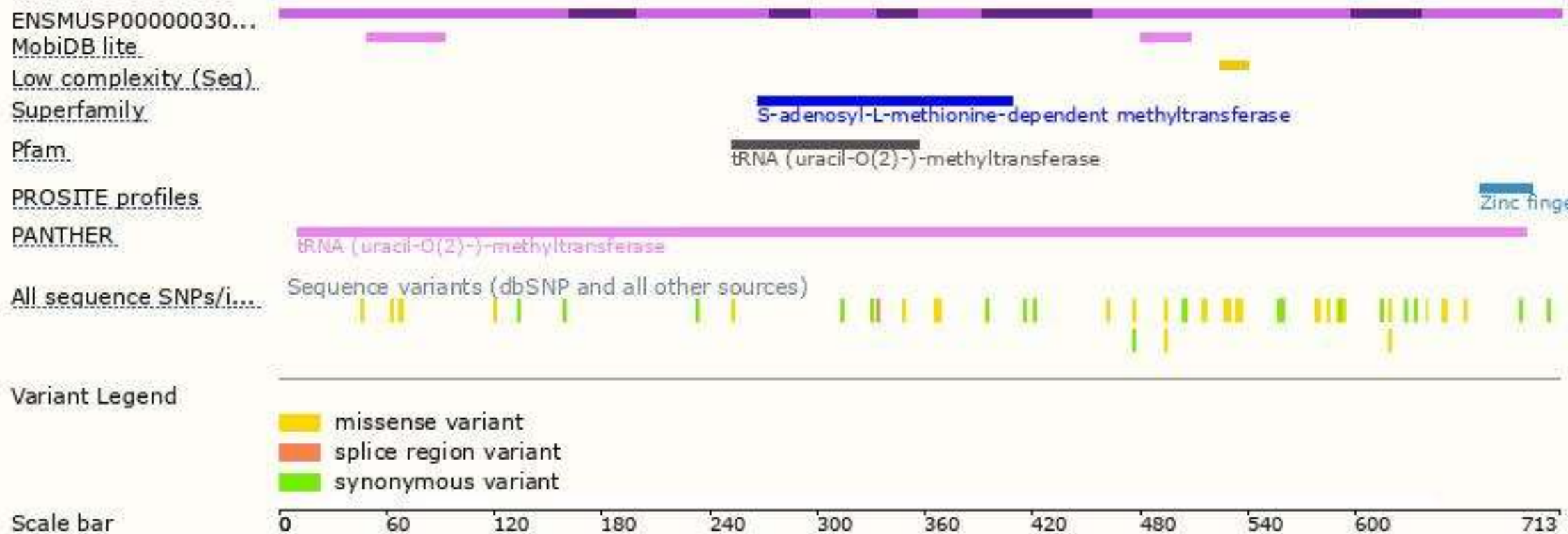
The strategy is based on the design of *Trmt44-201* transcript,the transcription is shown below:



Genomic location distribution



Protein domain



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

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