Letmd1 Cas9-CKO Strategy

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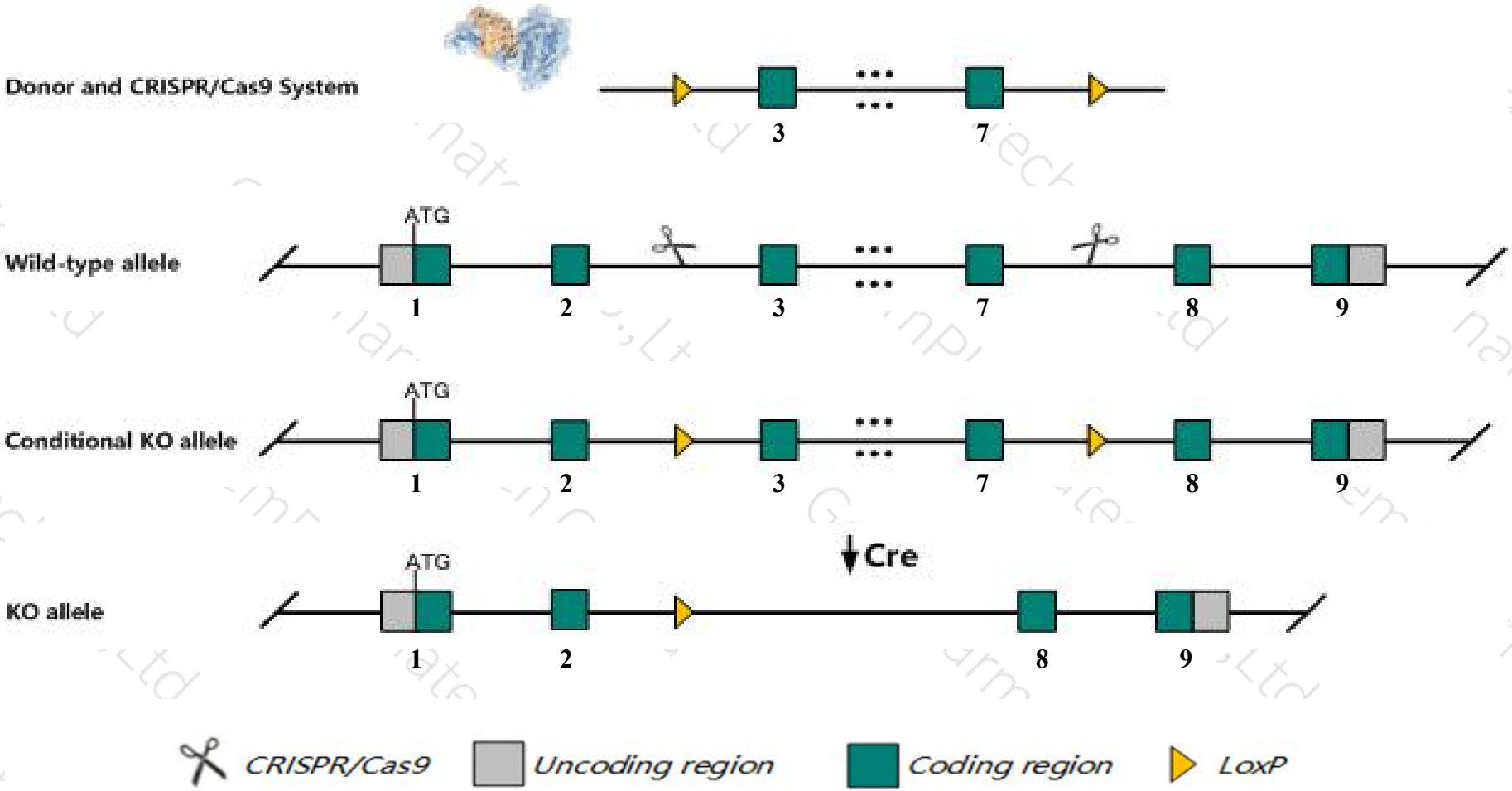
Project Overview

Project Name	***Letmd1***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Letmd1* gene. The schematic diagram is as follows:



Technical routes

- The *Letmd1* gene has 10 transcripts. According to the structure of *Letmd1* gene, exon3-exon7 of *Letmd1-201* (ENSMUST00000037001.9) transcript is recommended as the knockout region.The region contains 641bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Letmd1* 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.

Notice

➢ The *Letmd1* gene is located on the Chr15. 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）

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Letmd1 LETM1 domain containing 1 [Mus musculus (house mouse)]

Gene ID: 68614, updated on 31-Jan-2019

Summary

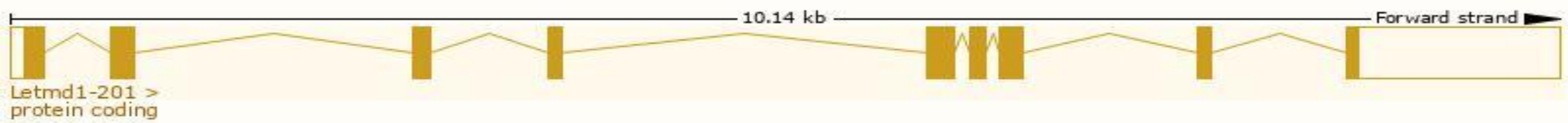
Official Symbol	Letmd1 provided by MGI
Official Full Name	LETM1 domain containing 1 provided by MGI
Primary source	MGI:MGI:1915864
See related	Ensembl:ENSMUSG00000037353
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	1110019O13Rik, AI593524, BB130465, BB235638, HCCR-2, HCCR1, MCC-32, Mccr
Expression	Ubiquitous expression in adrenal adult (RPKM 33.3), mammary gland adult (RPKM 24.6) and 28 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

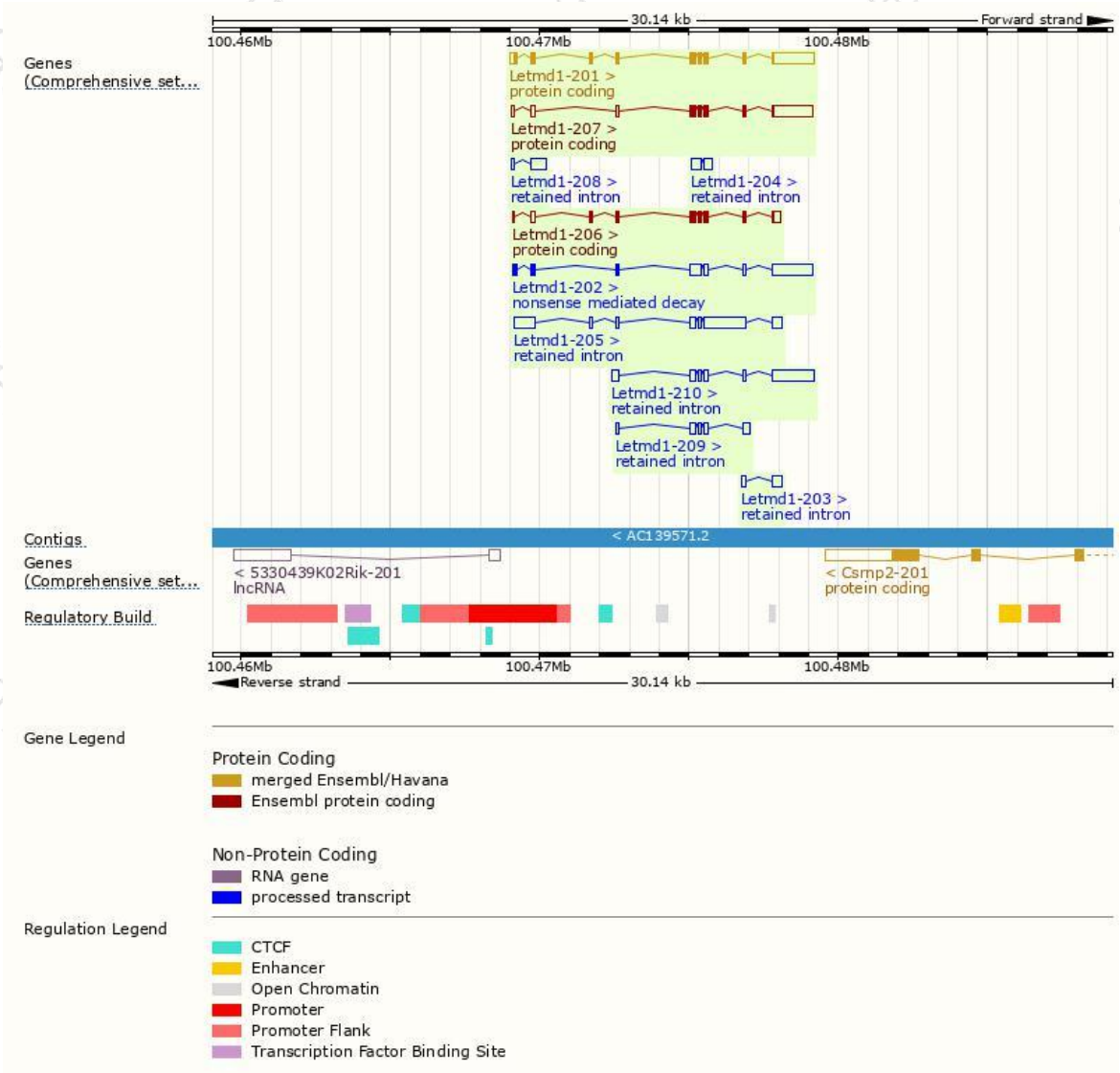
The gene has 10 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Letmd1-201	ENSMUST00000037001.9	2502	360aa	Protein coding	CCDS27838	Q924L1	TSL:1 GENCODE basic APPRIS P2
Letmd1-207	ENSMUST00000230294.1	2266	203aa	Protein coding	-	Q924L1	GENCODE basic
Letmd1-206	ENSMUST00000229648.1	1222	271aa	Protein coding	-	Q924L1	GENCODE basic APPRIS ALT2
Letmd1-202	ENSMUST00000229012.1	2389	123aa	Nonsense mediated decay	-	A0A2R8VK53	
Letmd1-205	ENSMUST00000229596.1	2917	No protein	Retained intron	-	-	
Letmd1-210	ENSMUST00000231001.1	2153	No protein	Retained intron	-	-	
Letmd1-209	ENSMUST00000230579.1	747	No protein	Retained intron	-	-	
Letmd1-204	ENSMUST00000229457.1	630	No protein	Retained intron	-	-	
Letmd1-208	ENSMUST00000230339.1	630	No protein	Retained intron	-	-	
Letmd1-203	ENSMUST00000229372.1	497	No protein	Retained intron	-	-	

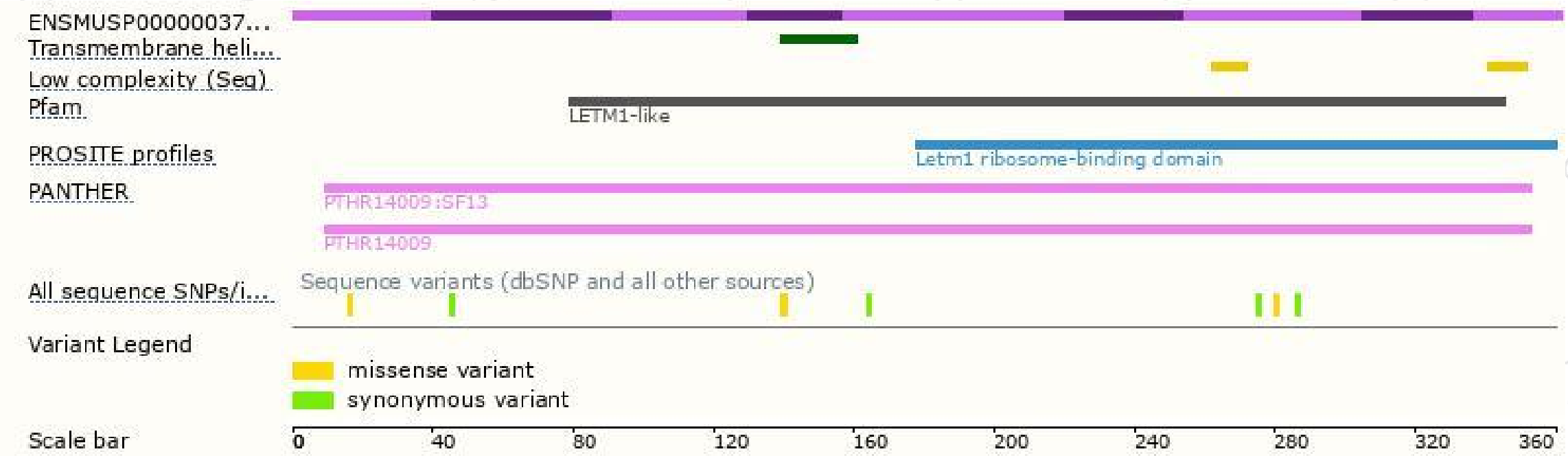
The strategy is based on the design of *Letmd1-201* transcript,The transcription is shown below



Genomic location distribution



Protein domain



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

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