Pam16 Cas9-CKO Strategy

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Project Overview

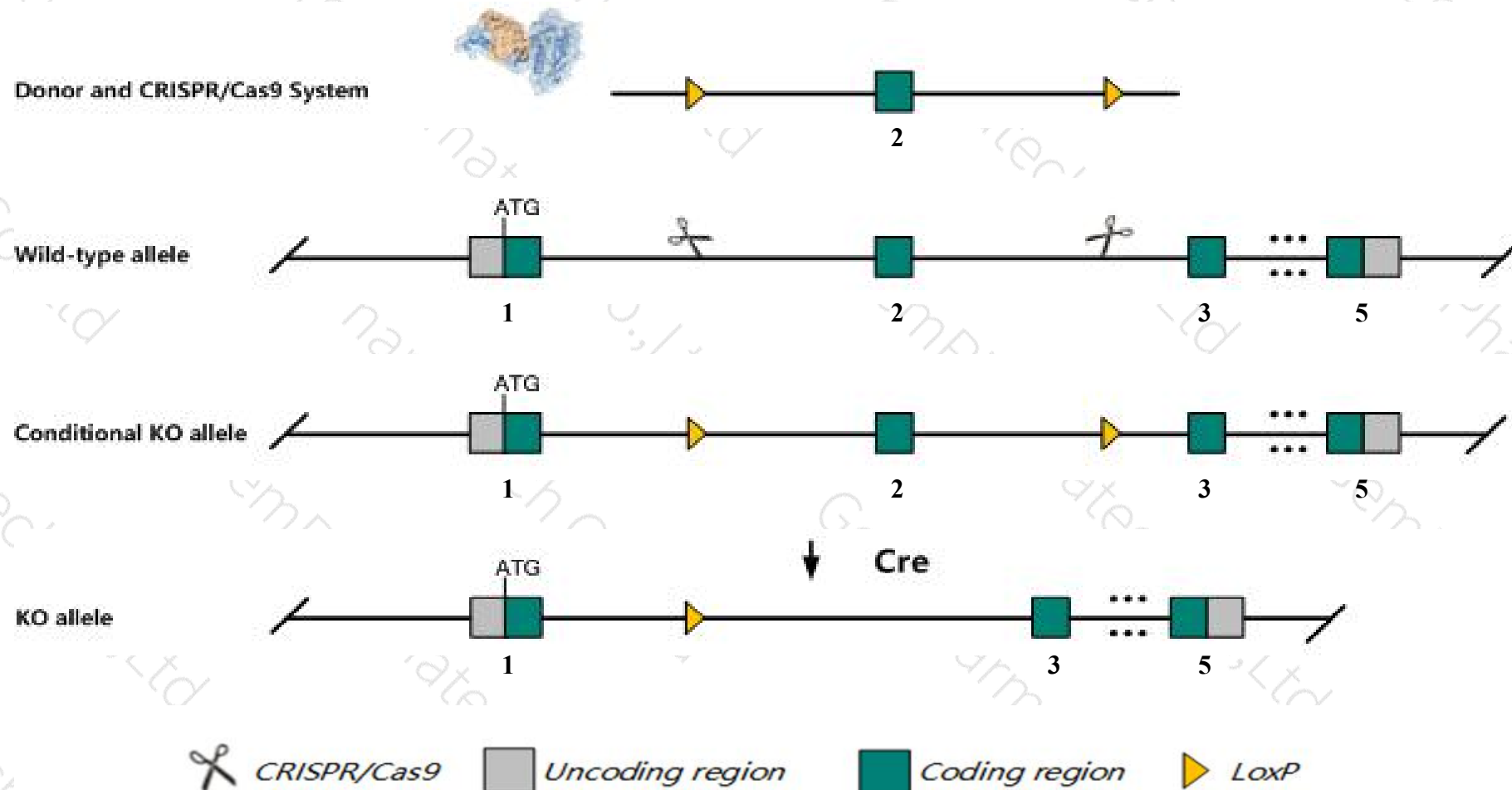|	
Project Name	*Pam16*
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Pam16* gene. The schematic diagram is as follows:

Technical routes

➤ The *Pam16* gene has 5 transcripts. According to the structure of *Pam16* gene, exon2 of *Pam16-201* (ENSMUST00000014445.6) transcript is recommended as the knockout region.The region contains Ens 85bp coding sequence. Knock out the region will result in disruption of protein function.

➤ In this project we use CRISPR/Cas9 technology to modify *Pam16* 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 *Pam16* gene is located on the Chr16. 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）

Pam16 presequence translocase-asssociated motor 16 homolog (S. cerevisiae) [Mus musculus (house mouse)]

Gene ID: 66449, updated on 13-Mar-2020

▲ Summary

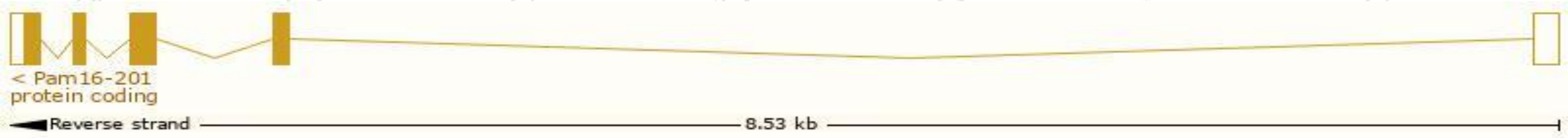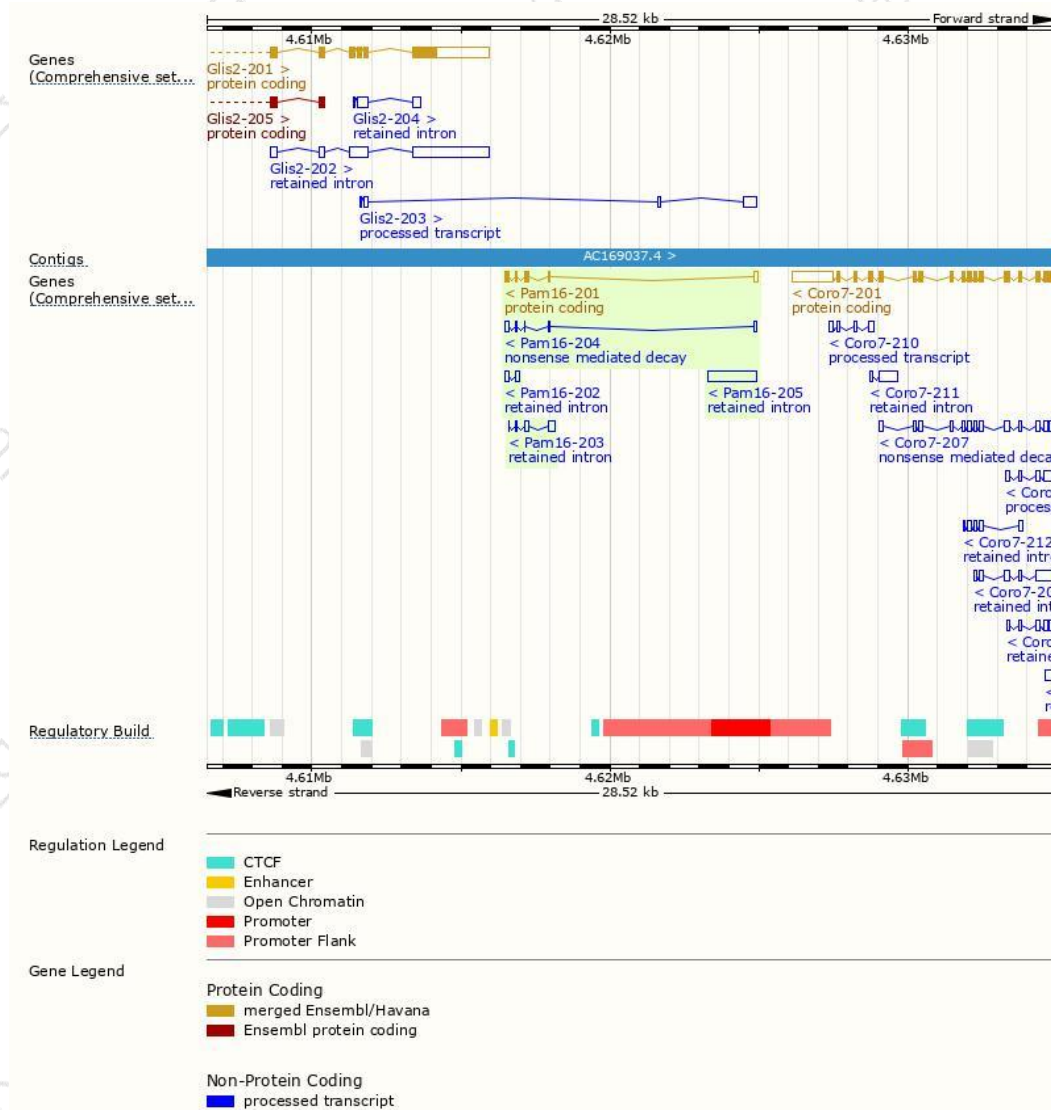|	
Official Symbol	Pam16 provided by MGI
Official Full Name	presequence translocase-asssociated motor 16 homolog (S. cerevisiae) provided by MGI
Primary source	MGI:MGI:1913699
See related	Ensembl:ENSMUSG00000014301
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	2010110I09Rik, AV006767, CGI-136, Magmas, Tim16, Timm16
Expression	Ubiquitous expression in testis adult (RPKM 60.5), thymus adult (RPKM 48.1) and 28 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Pam16-201	ENSMUST00000014445.6	596	125aa	Protein coding	CCDS37241	Q9CQV1	TSL:1 GENCODE basic APPRIS P1
Pam16-204	ENSMUST00000156889.7	431	39aa	Nonsense mediated decay	-	D6RI93	TSL:3
Pam16-205	ENSMUST00000229767.1	1634	No protein	Retained intron	-	-	-
Pam16-203	ENSMUST00000139534.1	471	No protein	Retained intron	-	-	TSL:2
Pam16-202	ENSMUST00000131250.7	346	No protein	Retained intron	-	-	TSL:2

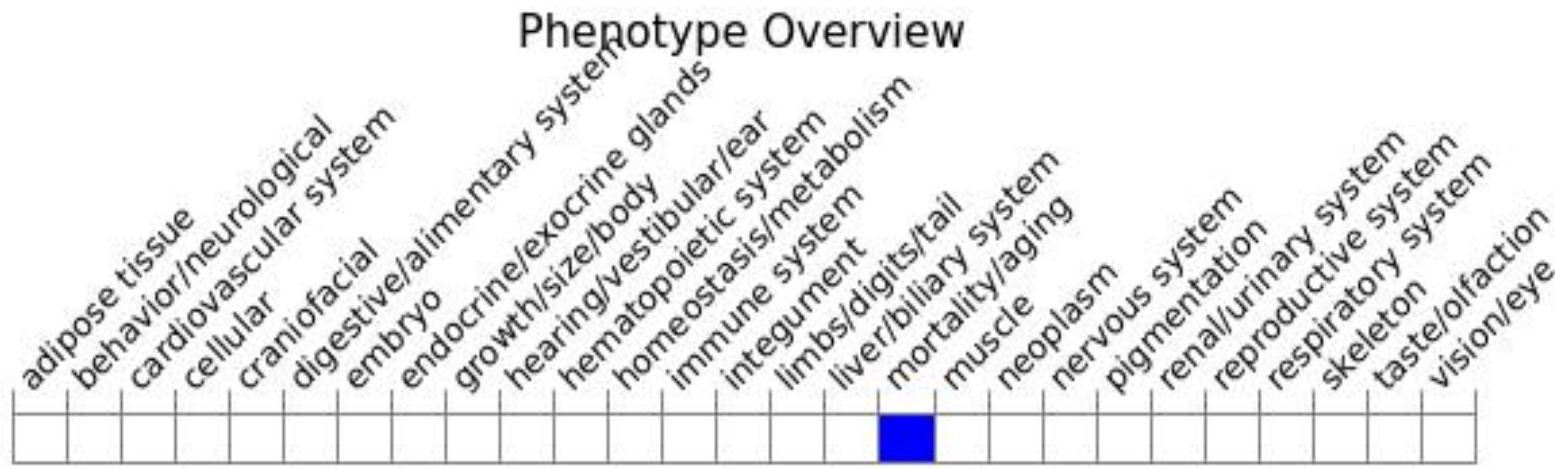The strategy is based on the design of *Pam16-201* transcript,the transcription is shown below:

< Pam16-201
protein coding

◄ Reverse strand   8.53 kb

Genomic location distribution

Protein domain

ENSMUSP00000014...	
Low complexity (Seg)	
Pfam	PF03656
PANTHER	Mitochondrial import inner membrane translocase subunit Tim16
	PTHR12388:SF0
Gene3D	Chaperone J-domain superfamily
All sequence SNPs/i...	Sequence variants (dbSNP and all other sources)

Variant Legend

- 🟨 missense variant
- 🟧 splice region variant
- 🟩 synonymous variant

Scale bar: 0   20   40   60   80   100   125
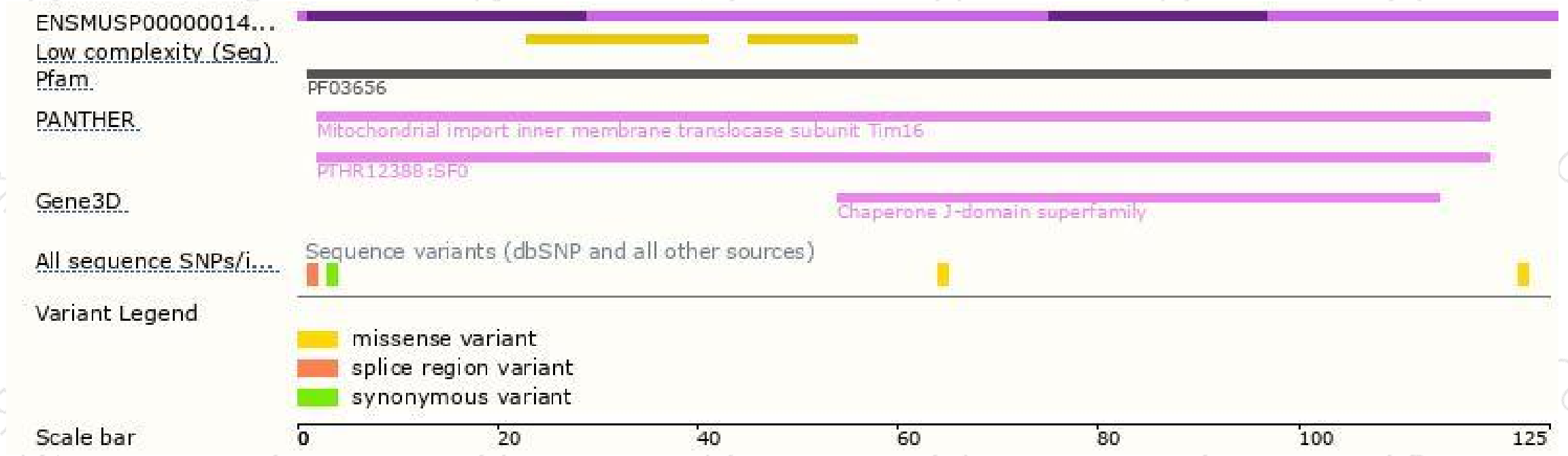
Mouse phenotype description(MGI)

Phenotypes affected by the gene are marked in blue.Data quoted from MGI database(http://www.informatics.jax.org/).

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