Pdcd4 Cas9-KO Strategy

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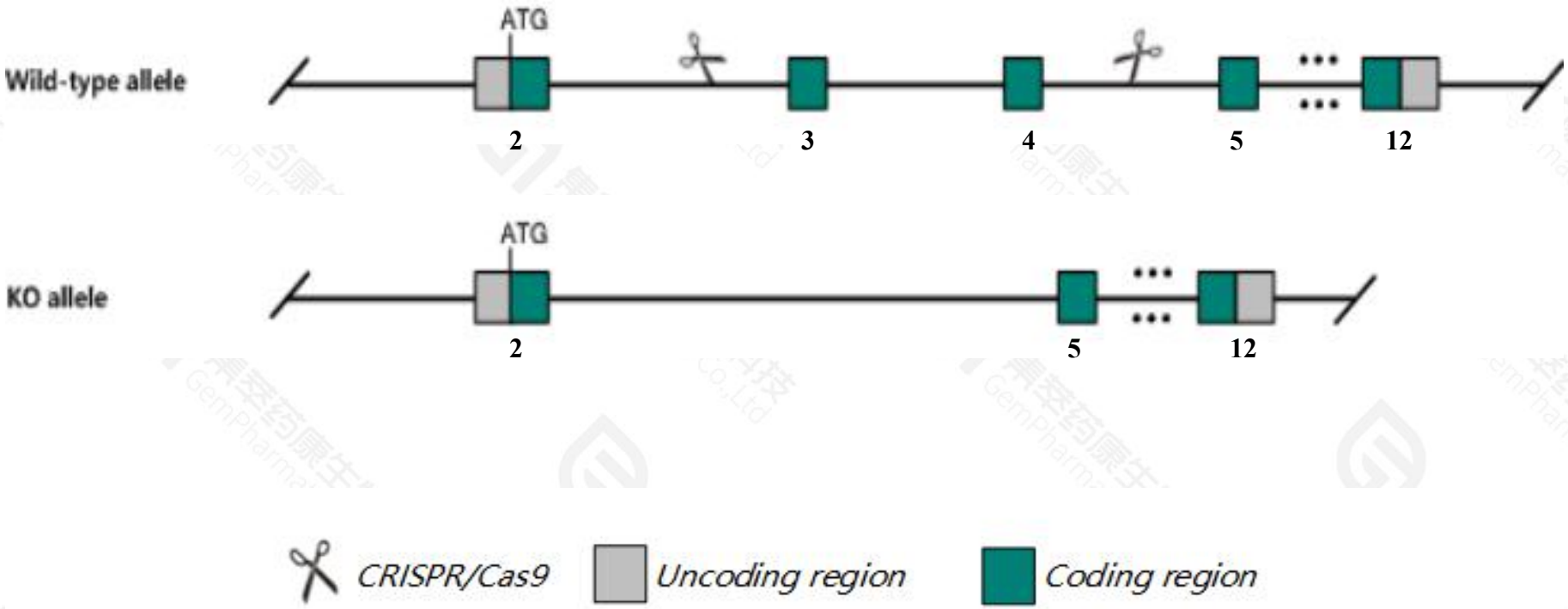
Project Overview

Project Name	***Pdcd4***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Pdcd4* gene. The schematic diagram is as follows:



Technical routes

➢ The *Pdcd4* gene has 6 transcripts. According to the structure of *Pdcd4* gene, exon3-exon4 of *Pdcd4-202*(ENSMUST00000074371.13) transcript is recommended as the knockout region.The region contains 398bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Pdcd4* gene. The brief process is as follows: CRISPR/Cas9 system 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.

Notice

➢ According to the existing MGI data,mice homozygous for a null allele have a higher prevalence of B cell derived lymphomas, multi-organ cysts and decreased susceptibility to experimentally induced autoimmune encephalomyelitis and type 1 diabetes.

➢ The *Pdcd4* gene is located on the Chr19. 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information （NCBI）

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Pdcd4 programmed cell death 4 [Mus musculus (house mouse)]

Gene ID: 18569, updated on 13-Mar-2020

Summary

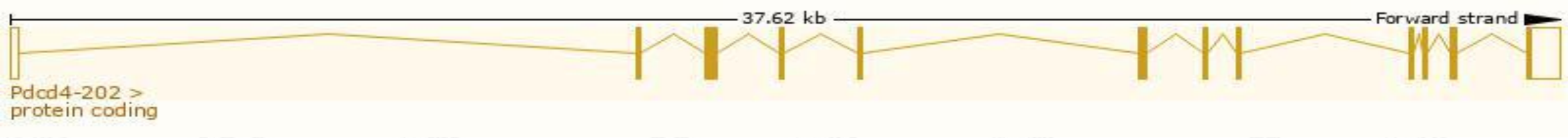
Official Symbol	Pdcd4 provided by MGI
Official Full Name	programmed cell death 4 provided byMGI
Primary source	MGI:MGI:107490
See related	Ensembl:ENSMUSG00000024975
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	D19Ucla1, Ma3, Tis
Expression	Broad expression in bladder adult (RPKM 26.5), placenta adult (RPKM 14.8) and 24 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

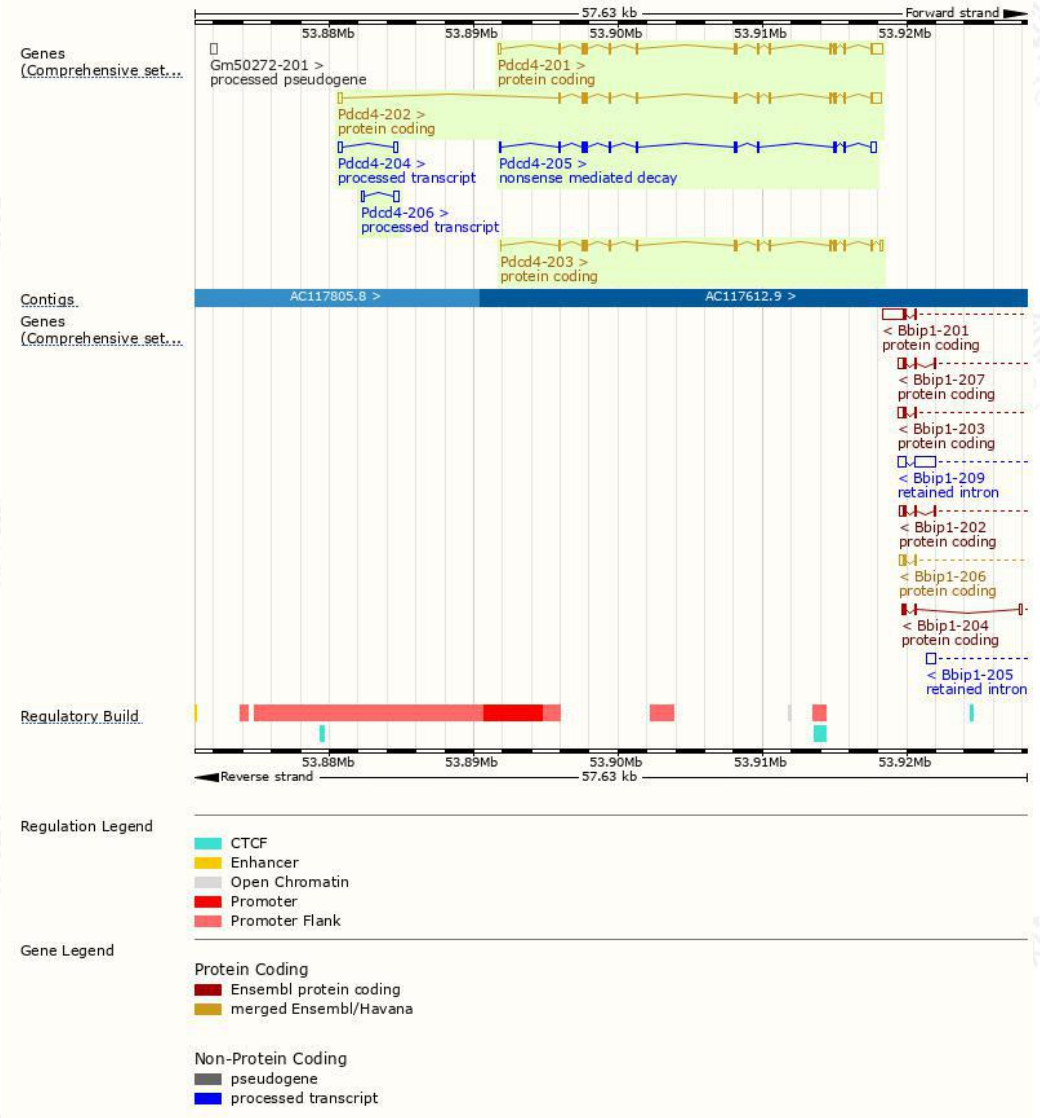
The gene has 6 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Pdcd4-202	ENSMUST00000074371.12	2408	469aa	Protein coding	CCDS29903	Q61823	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Pdcd4-201	ENSMUST00000025931.13	2381	469aa	Protein coding	CCDS29903	Q61823	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Pdcd4-203	ENSMUST00000165617.2	1733	469aa	Protein coding	CCDS29903	Q61823	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Pdcd4-205	ENSMUST00000237060.1	1623	319aa	Nonsense mediated decay	-	A0A494BA05	
Pdcd4-204	ENSMUST00000235854.1	538	No protein	Processed transcript	-	-	
Pdcd4-206	ENSMUST00000237699.1	528	No protein	Processed transcript	-	-	

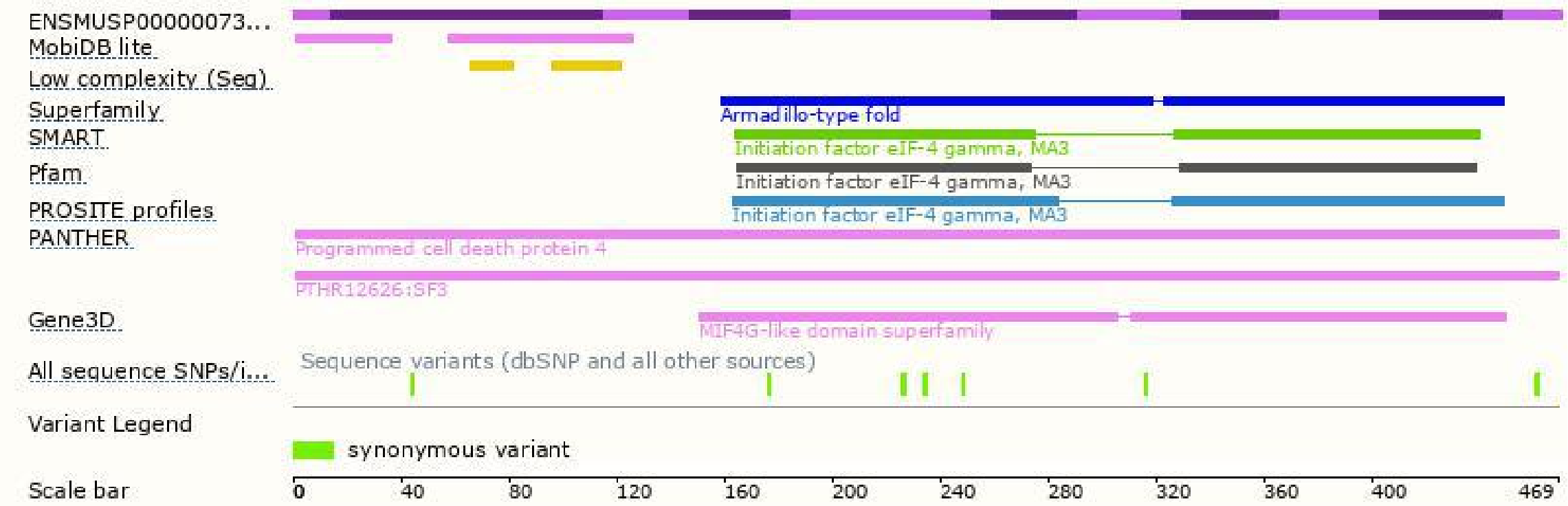
The strategy is based on the design of *Pdcd4-202* transcript,the transcription is shown below:



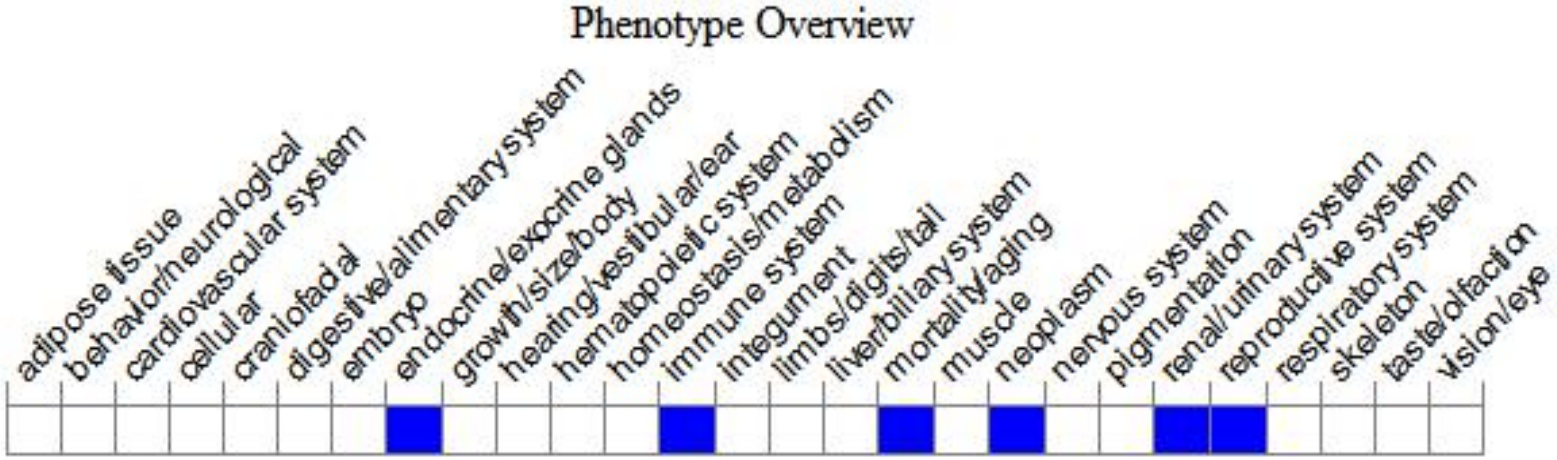
Genomic location distribution



Protein domain



Mouse phenotype description(MGI)



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

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