

# *Prdm9* Cas9-CKO Strategy

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# Project Overview

**Project Name**

***Prdm9***

**Project type**

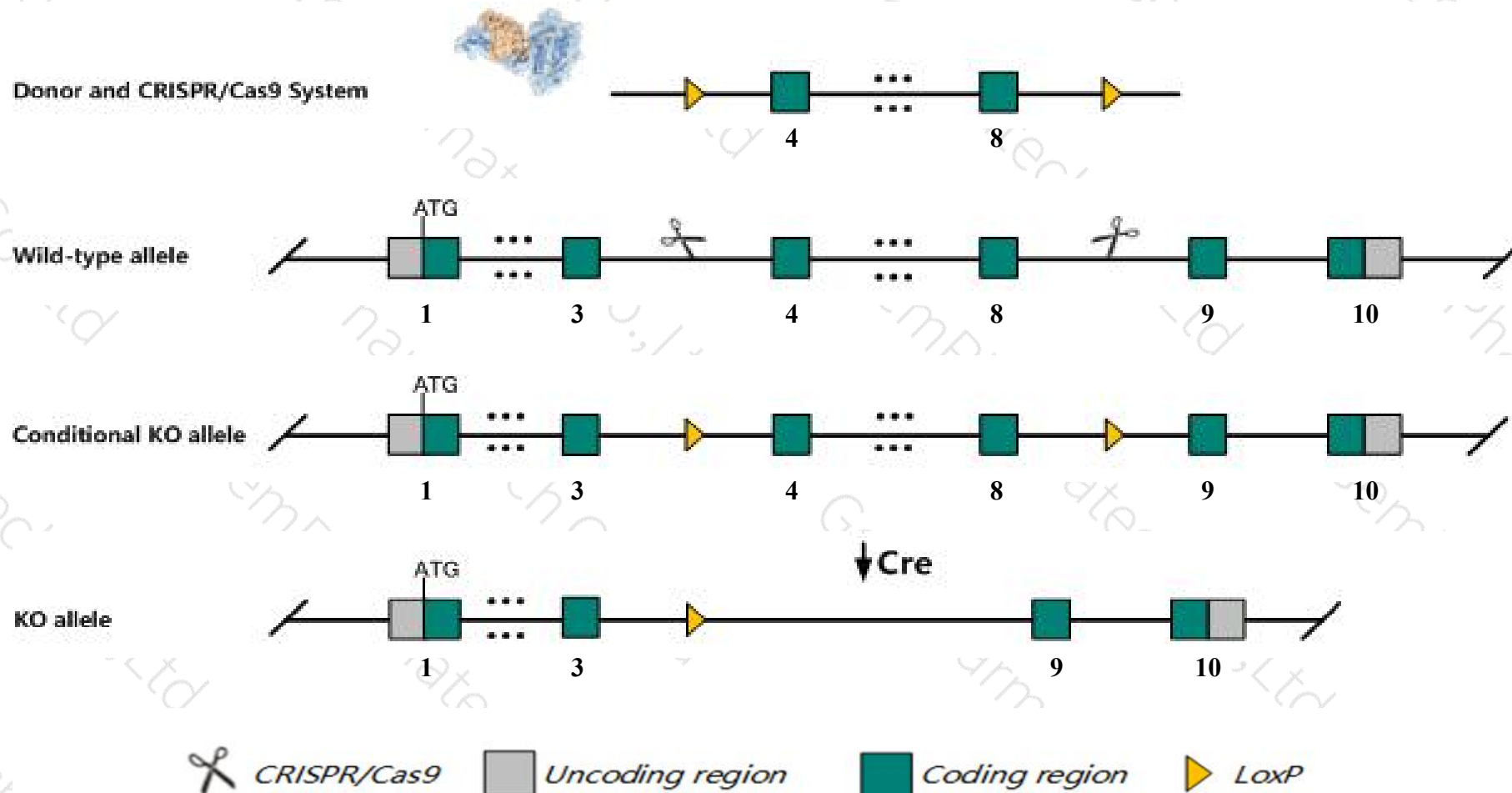
**Cas9-CKO**

**Strain background**

**C57BL/6JGpt**

# Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Prdm9* gene. The schematic diagram is as follows:



# Technical routes

- The *Prdm9* gene has 7 transcripts. According to the structure of *Prdm9* gene, exon4-exon8 of *Prdm9*-207 (ENSMUST00000233295.1) transcript is recommended as the knockout region. The region contains 649bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Prdm9* 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.

- According to the existing MGI data, mice homozygous for a knock-out allele show decreased oocyte number, azoospermia, and sterility in both sexes due to severe impairment of the double-stranded break repair pathway, deficient pairing of homologous chromosomes, and impaired sex body formation.
- Transcript *Prdm9*-203 may not be affected.
- The N-terminal of *Prdm9* gene will remain several amino acids, it may remain the partial function of *Prdm9* gene.
- The *Prdm9* gene is located on the Chr17. 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)

## Prdm9 PR domain containing 9 [Mus musculus (house mouse)]

Gene ID: 213389, updated on 13-Mar-2020

### Summary



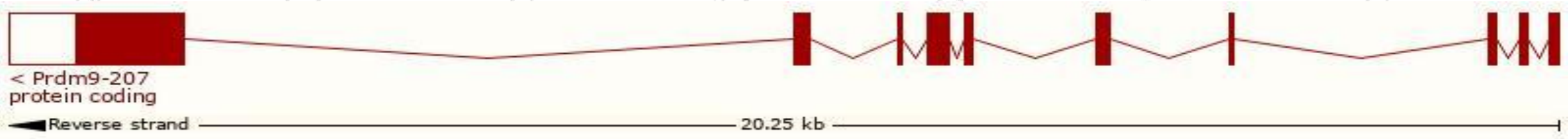
<b>Official Symbol</b>	Prdm9 provided by <a href="#">MGI</a>
<b>Official Full Name</b>	PR domain containing 9 provided by <a href="#">MGI</a>
<b>Primary source</b>	<a href="#">MGI:MGI:2384854</a>
<b>See related</b>	<a href="#">Ensembl:ENSMUSG00000051977</a>
<b>Gene type</b>	protein coding
<b>RefSeq status</b>	VALIDATED
<b>Organism</b>	<a href="#">Mus musculus</a>
<b>Lineage</b>	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
<b>Also known as</b>	BC012016, Dsbc1, G1-419-29, Meisetz, PRDM9-B, Rcr1, repro7
<b>Expression</b>	Ubiquitous expression in stomach adult (RPKM 2.6), spleen adult (RPKM 1.6) and 28 other tissues <a href="#">See more</a>
<b>Orthologs</b>	<a href="#">human</a> <a href="#">all</a>

# Transcript information（Ensembl）

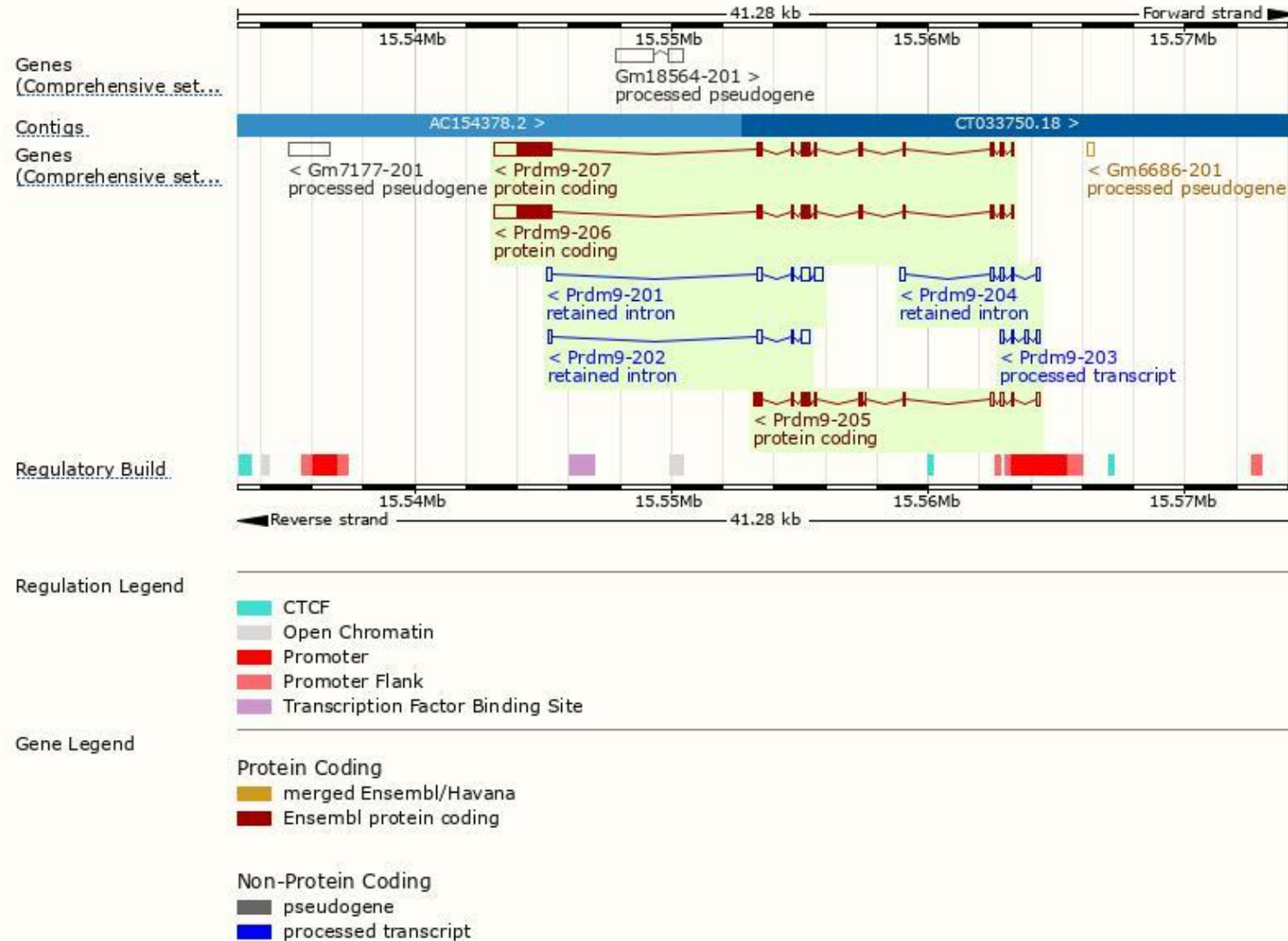
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Prdm9-207	<a href="#">ENSMUST00000233295.1</a>	3460	<a href="#">847aa</a>	Protein coding	<a href="#">CCDS49963</a>	<a href="#">E9Q4V2</a>	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 P2
Prdm9-206	<a href="#">ENSMUST00000167994.9</a>	3438	<a href="#">843aa</a>	Protein coding	-	<a href="#">E9Q4V2 Q96EQ9</a>	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 ALT2
Prdm9-205	<a href="#">ENSMUST00000147532.7</a>	1447	<a href="#">283aa</a>	Protein coding	-	<a href="#">Q96EQ9</a>	TSL:1 GENCODE basic
Prdm9-203	<a href="#">ENSMUST00000132675.1</a>	474	No protein	Processed transcript	-	-	TSL:3
Prdm9-201	<a href="#">ENSMUST00000128267.7</a>	1082	No protein	Retained intron	-	-	TSL:1
Prdm9-202	<a href="#">ENSMUST00000130297.1</a>	708	No protein	Retained intron	-	-	TSL:3
Prdm9-204	<a href="#">ENSMUST00000139911.1</a>	666	No protein	Retained intron	-	-	TSL:3

The strategy is based on the design of *Prdm9-207* transcript,the transcription is shown below:



# Genomic location distribution



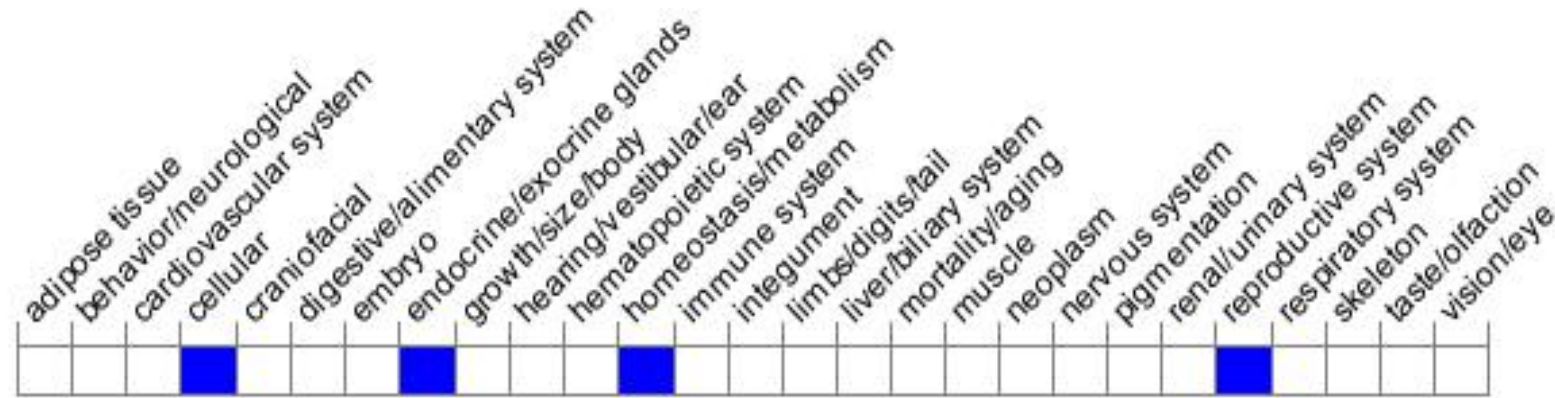


# Protein domain



# Mouse phenotype description(MGI)

## Phenotype Overview



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

According to the existing MGI data, mice homozygous for a knock-out allele show decreased oocyte number, azoospermia, and sterility in both sexes due to severe impairment of the double-stranded break repair pathway, deficient pairing of homologous chromosomes, and impaired sex body formation.

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

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