

Piwil1 Cas9-CKO Strategy

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Design Date: 2020-5-26

Project Overview

Project Name

Piwill

Project type

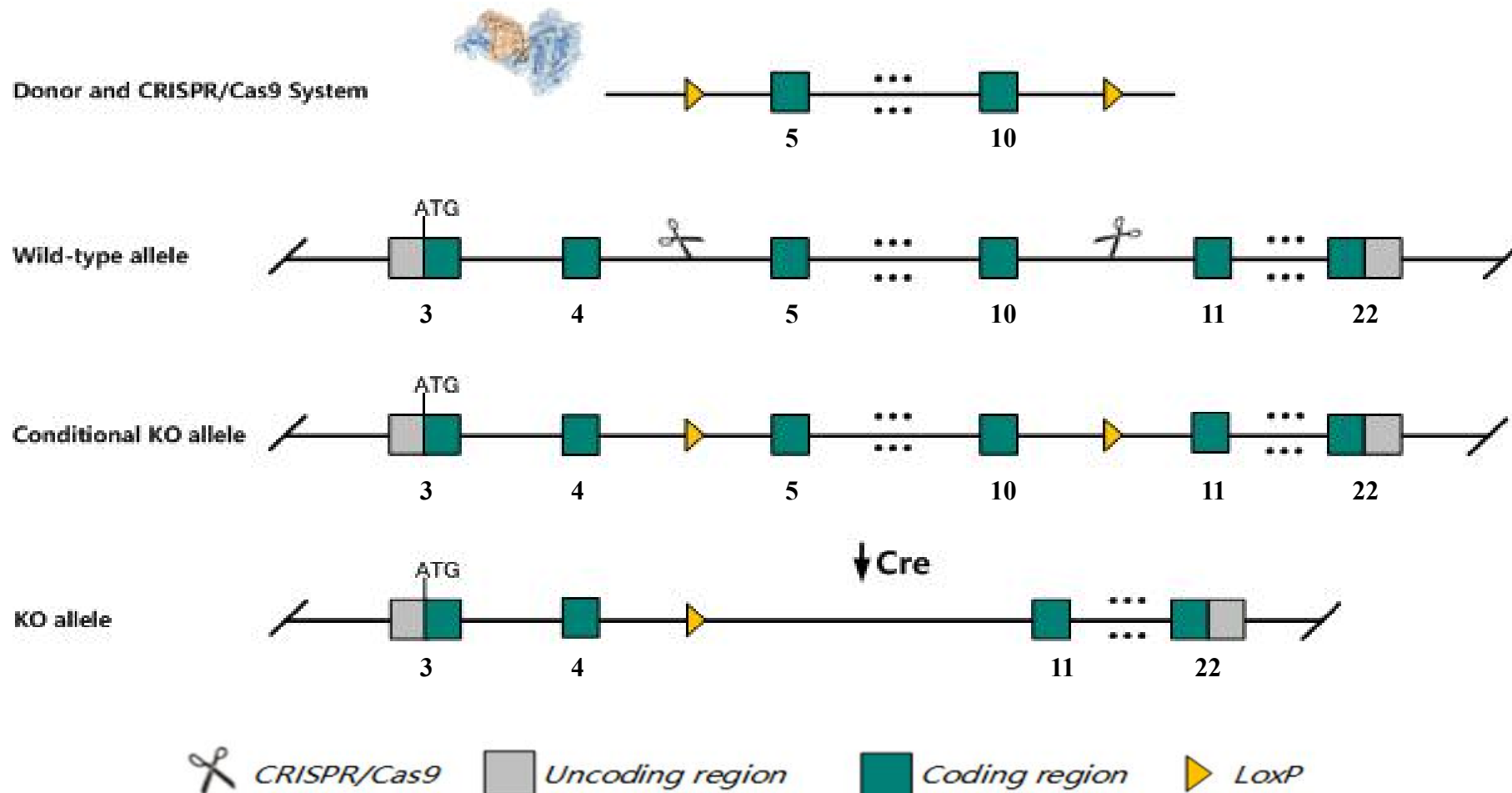
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



The *Piwill* gene has 3 transcripts. According to the structure of *Piwill* gene, exon5-exon10 of *Piwill*-201 (ENSMUST00000086056.7) transcript is recommended as the knockout region. The region contains 854bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Piwill* 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, homozygotes for a targeted null mutation exhibit male sterility due to a block in spermatogenesis beginning at the round spermatid stage.

The *Piwill* 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.

Piwil1 piwi-like RNA-mediated gene silencing 1 [Mus musculus (house mouse)]

Gene ID: 57749, updated on 13-Mar-2020

Summary

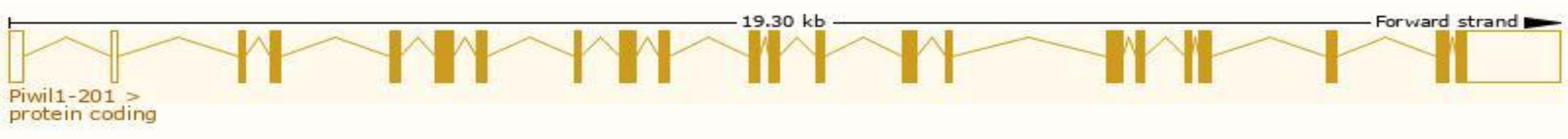
Official Symbol	Piwil1 provided by MGI
Official Full Name	piwi-like RNA-mediated gene silencing 1 provided by MGI
Primary source	MGI:MGI:1928897
See related	Ensembl:ENSMUSG00000029423
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	MIWI
Expression	Restricted expression toward testis adult (RPKM 111.5) See more
Orthologs	human all

Transcript information Ensembl

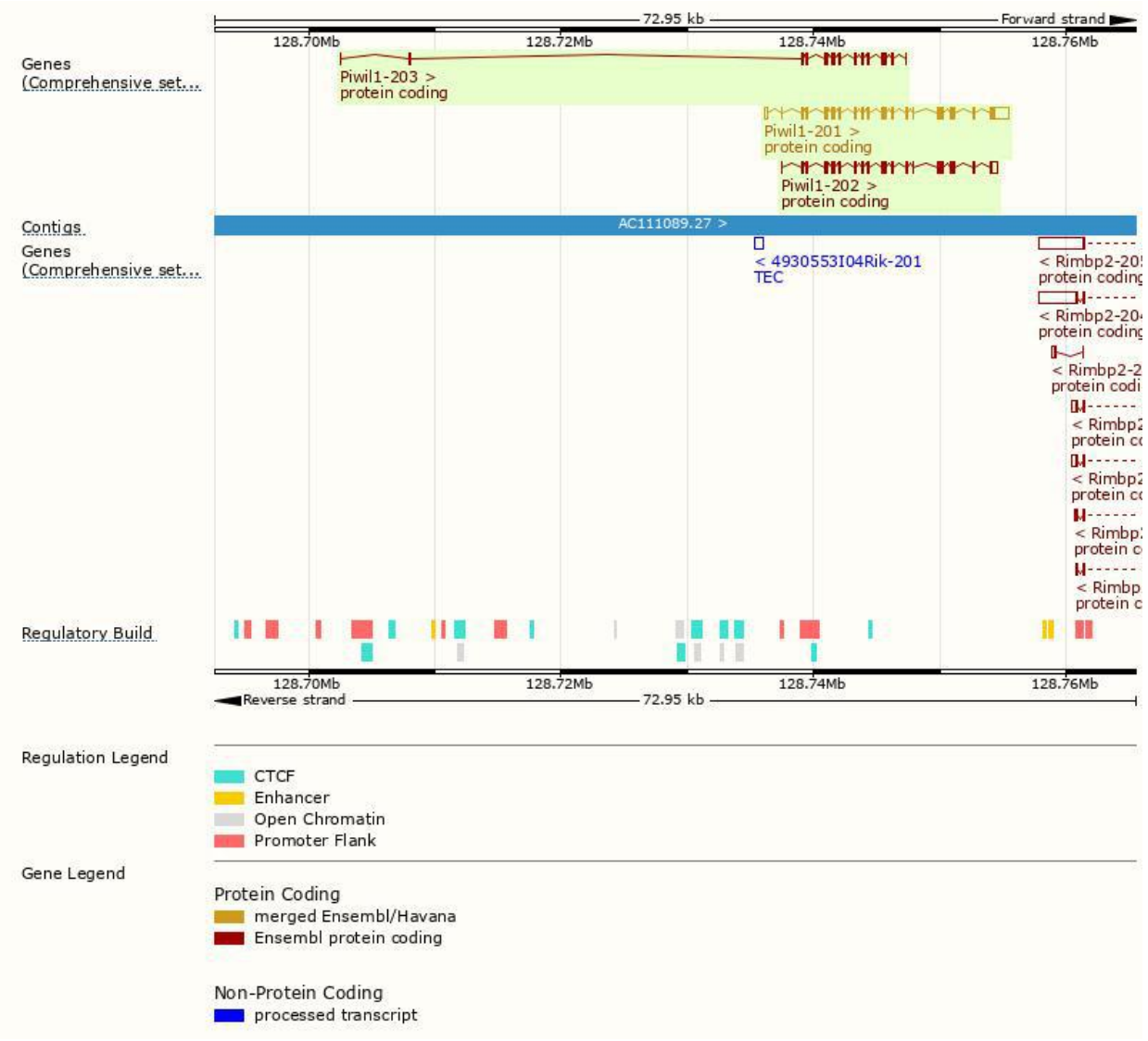
The gene has 3 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Piwil1-201	ENSMUST00000086056.7	4016	862aa	Protein coding	CCDS19690	Q9JMB7	TSL:1 GENCODE basic APPRIS P2
Piwil1-202	ENSMUST00000195959.1	3013	835aa	Protein coding	-	Q9JMB7	TSL:1 GENCODE basic APPRIS ALT2
Piwil1-203	ENSMUST00000200192.4	1691	493aa	Protein coding	-	A0A0G2JEK6	CDS 3' incomplete TSL:1

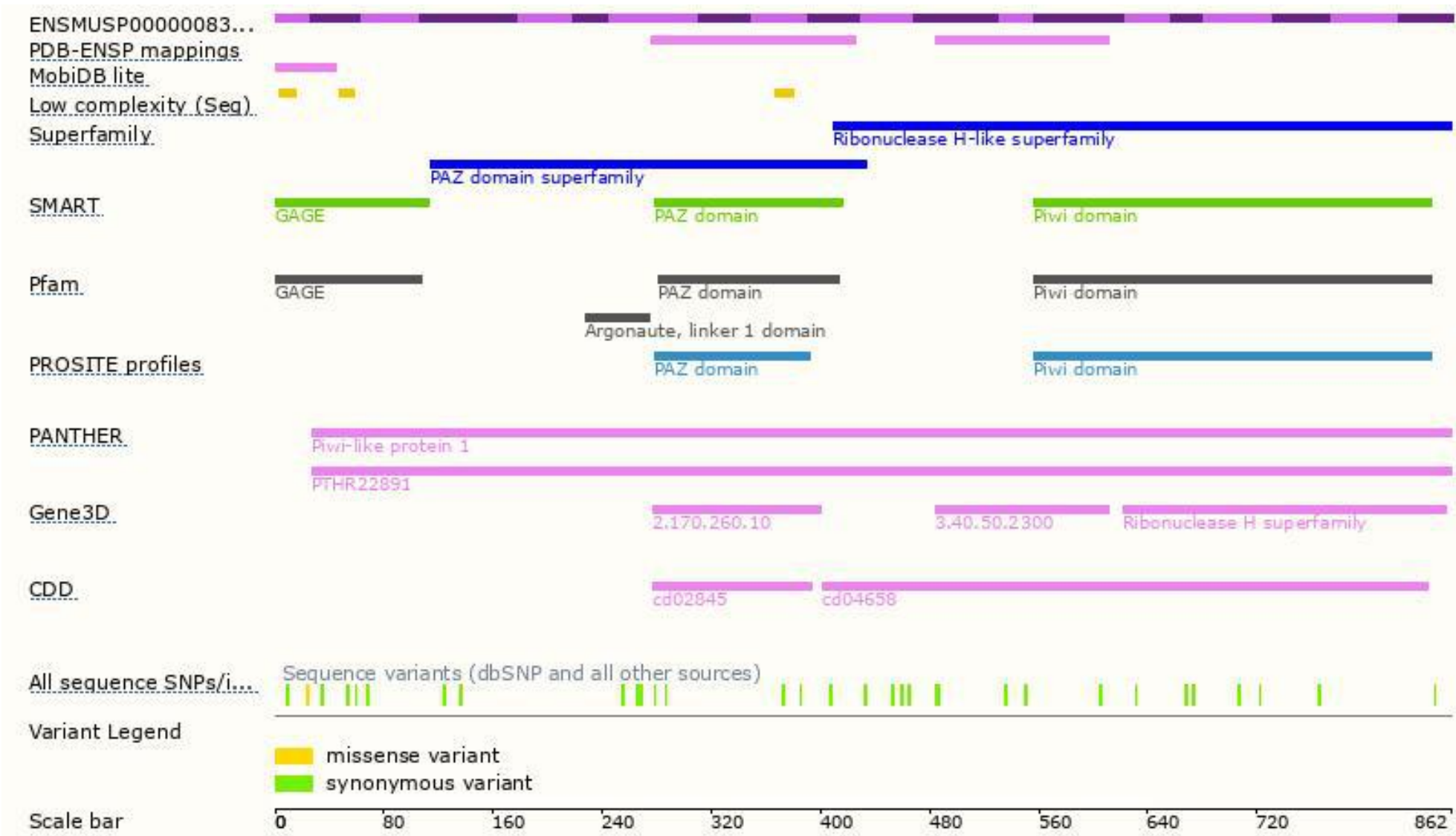
The strategy is based on the design of *Piwil1-201* transcript,the transcription is shown below:



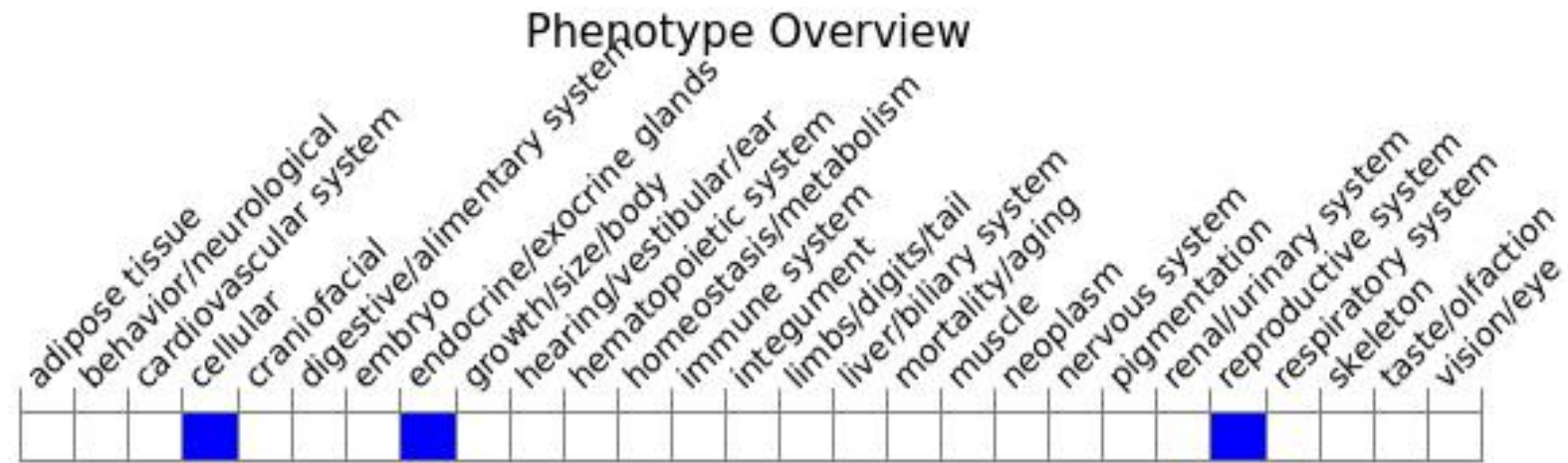
Genomic location distribution



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/>).

According to the existing MGI data, homozygotes for a targeted null mutation exhibit male sterility due to a block in spermatogenesis beginning at the round spermatid stage.

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