Plrg1 Cas9-KO Strategy

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Reviewer: Wenjing Li

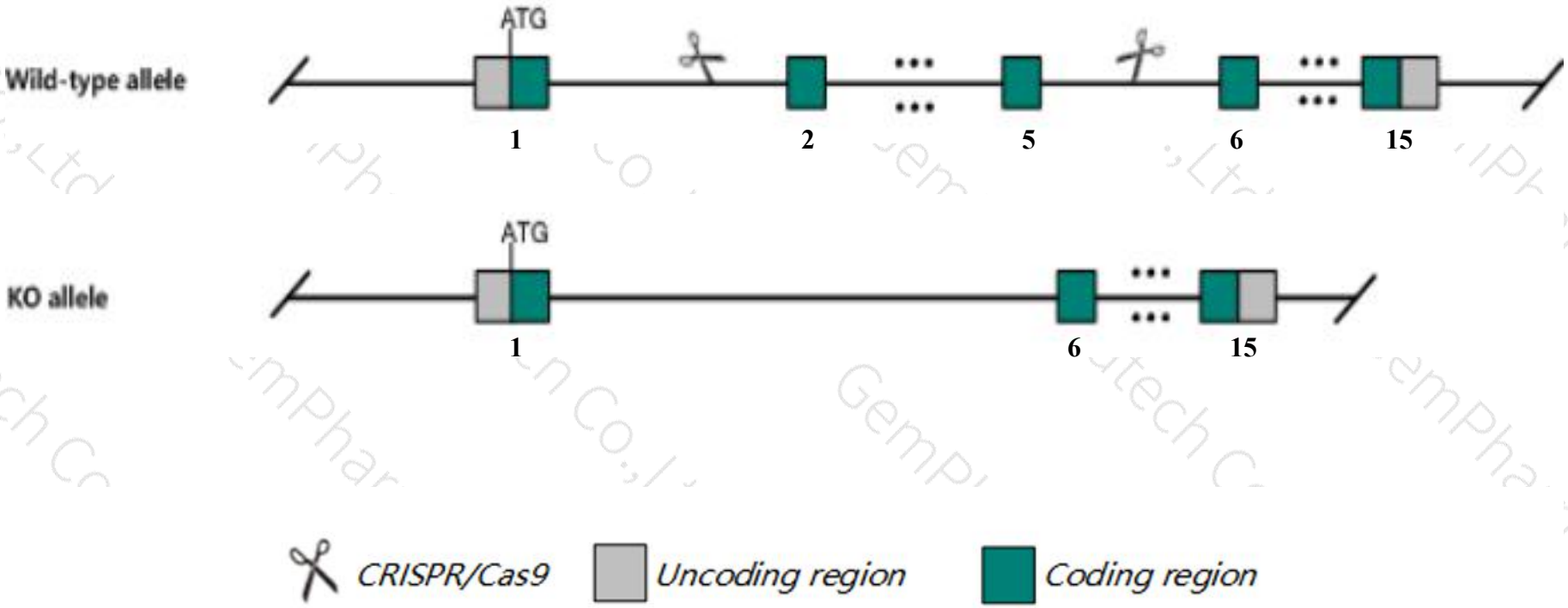
Design Date: 2020-8-5

Project Overview

Project Name	***Plrg1***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Plrg1* gene. The schematic diagram is as follows:



Technical routes

➢ The *Plrg1* gene has 5 transcripts. According to the structure of *Plrg1* gene, exon2-exon5 of *Plrg1-204*(ENSMUST00000150268.7) transcript is recommended as the knockout region.The region contains 395bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Plrg1* 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 knock-out allele exhibit embryonic lethality by E1.5.

➢ The *Plrg1* gene is located on the Chr3. 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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Plrg1 pleiotropic regulator 1 [Mus musculus (house mouse)]

Gene ID: 53317, updated on 13-Mar-2020

Summary

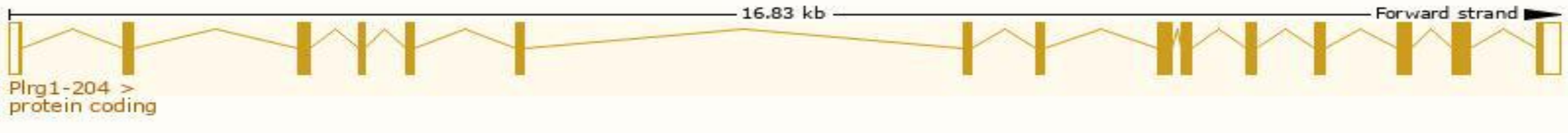
Official Symbol	Plrg1 provided by MGI
Official Full Name	pleiotropic regulator 1 provided byMGI
Primary source	MGI:MGI:1858197
See related	Ensembl:ENSMUSG00000027998
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AA958940, C80566, Tango4
Expression	Ubiquitous expression in CNS E11.5 (RPKM 34.5), CNS E14 (RPKM 30.5) and 28 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

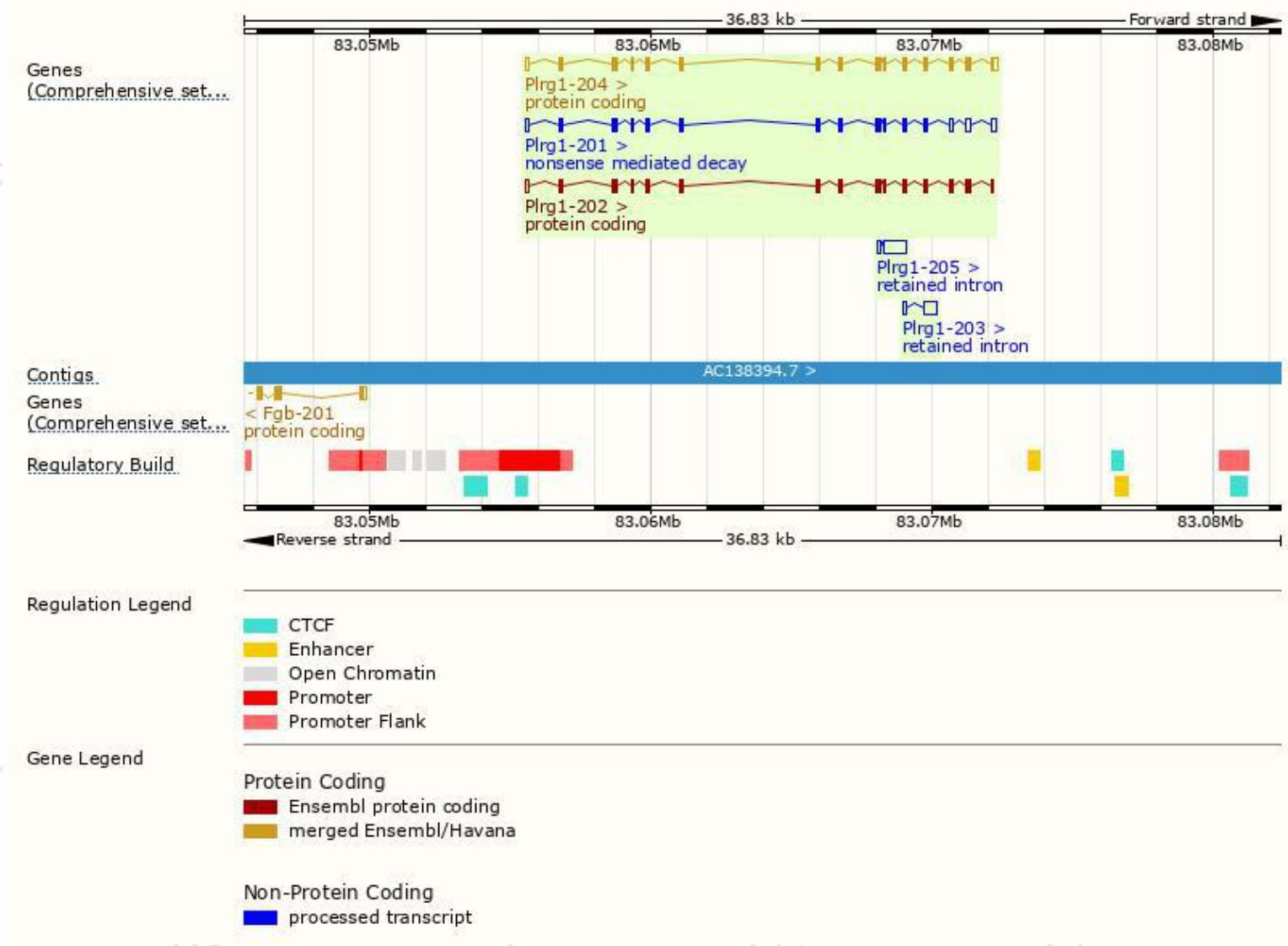
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Plrg1-204	ENSMUST00000150268.7	1851	513aa	Protein coding	CCDS17433	Q922V4	TSL:1 GENCODE basic APPRIS P1
Plrg1-202	ENSMUST00000122128.1	1608	504aa	Protein coding	-	D3Z4V1	TSL:5 GENCODE basic
Plrg1-201	ENSMUST00000029628.10	1749	383aa	Nonsense mediated decay	-	F8WI31	TSL:1
Plrg1-205	ENSMUST00000151915.1	911	No protein	Retained intron	-	-	TSL:3
Plrg1-203	ENSMUST00000135813.1	564	No protein	Retained intron	-	-	TSL:3

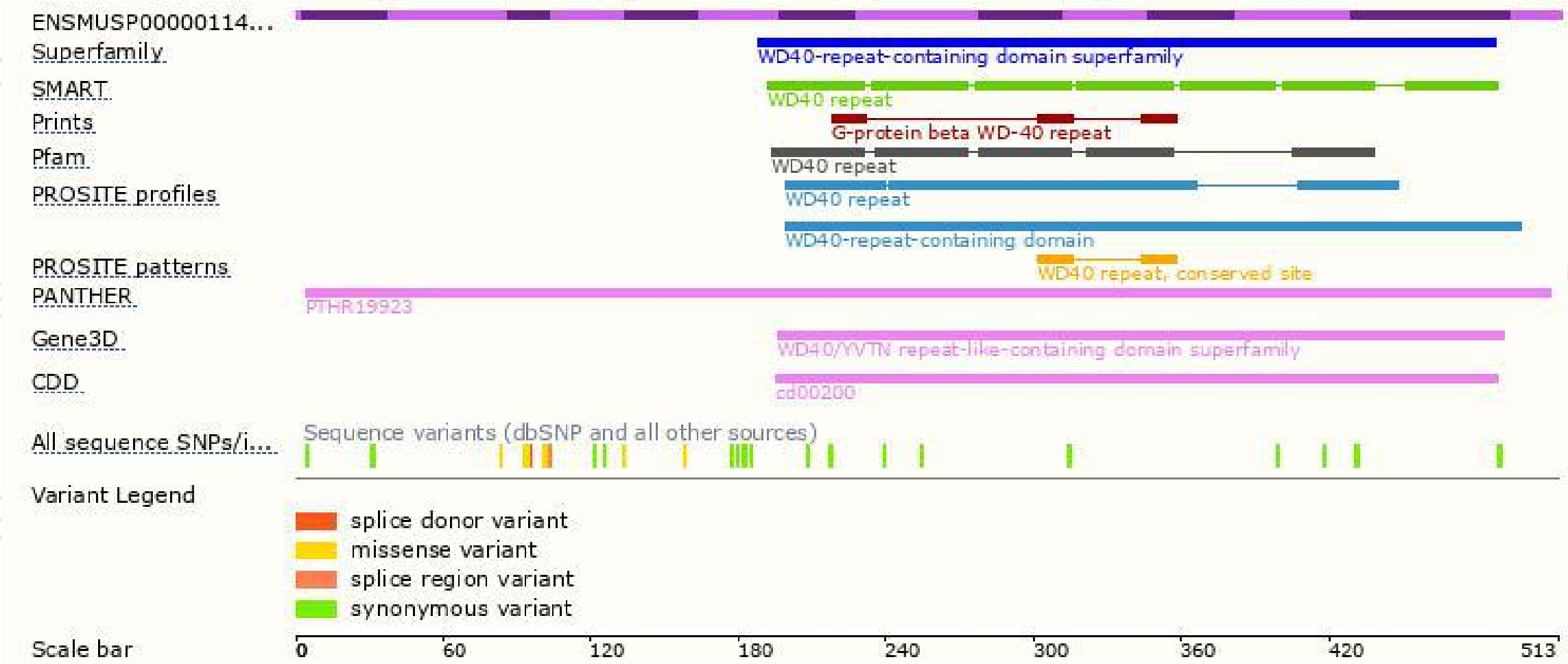
The strategy is based on the design of *Plrg1-204* transcript,the transcription is shown below:



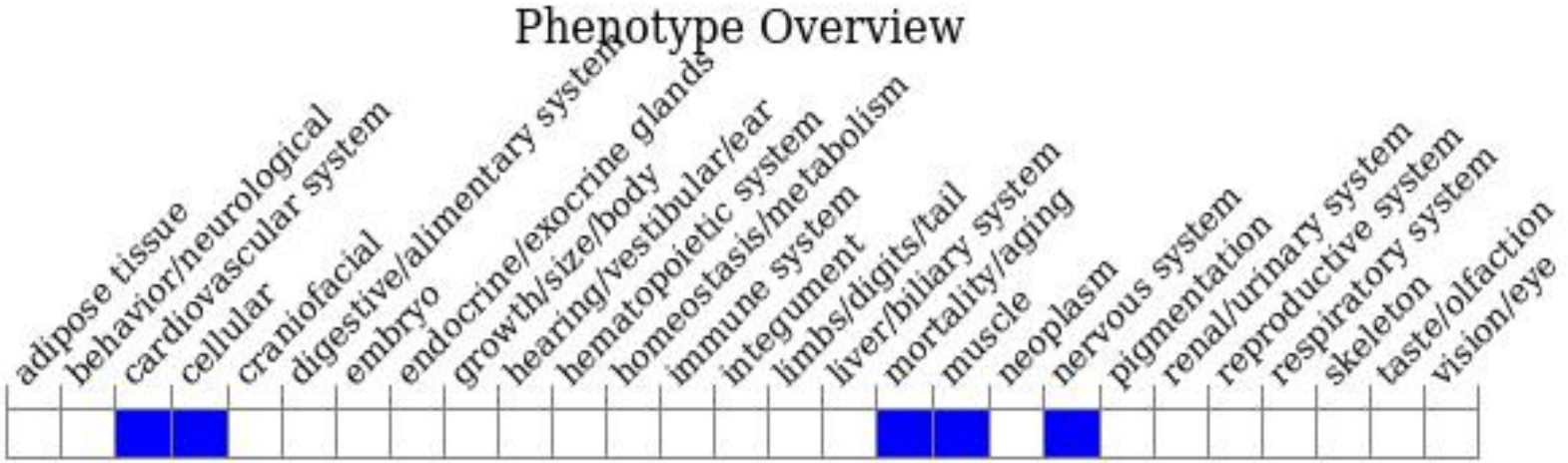
Genomic location distribution



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,mice homozygous for a knock-out allele exhibit embryonic lethality by E1.5.

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

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