Ccnyl1 Cas9-CKO Strategy

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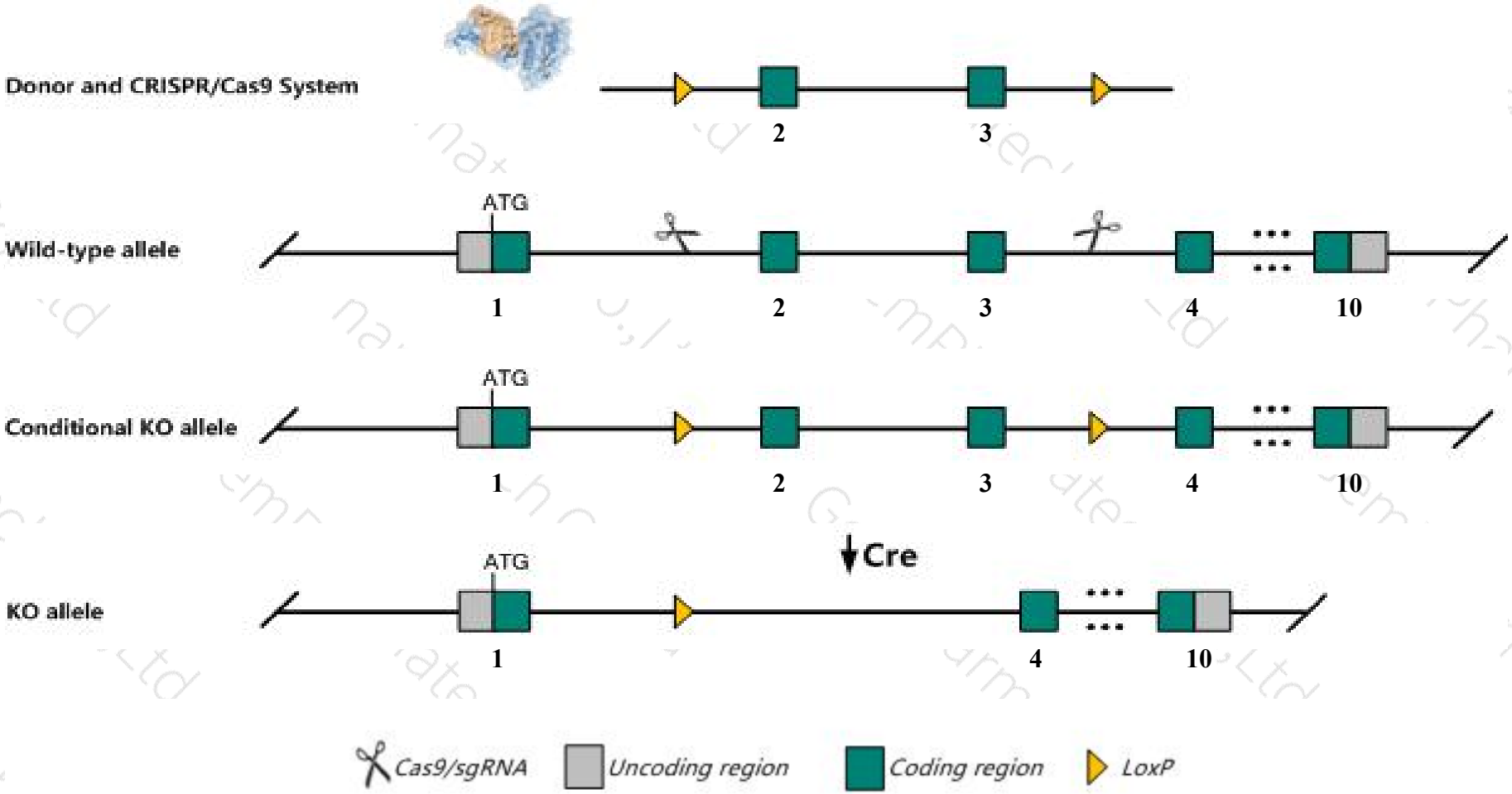
Project Overview

Project Name	***Ccnyl1***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Ccnyl1* gene. The schematic diagram is as follows:



Technical routes

➢ The *Ccnyl1* gene has 4 transcripts. According to the structure of *Ccnyl1* gene, exon2-exon3 of *Ccnyl1-201*(ENSMUST00000094898.4) transcript is recommended as the knockout region.The region contains 110bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Ccnyl1* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA 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 was 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

➢ According to the existing MGI data,mice homozygous for a knock-out allele exhibit male infertility associated with asthenozoospermia and teratozoospermia.

➢ The *Ccnyl1* gene is located on the Chr1. 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）

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Ccnyl1 cyclin Y-like 1 [Mus musculus (house mouse)]

Gene ID: 227210, updated on 13-Mar-2020

Summary

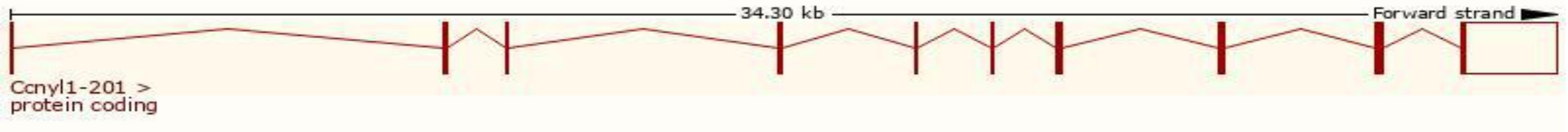
Official Symbol	Ccnyl1 provided by MGI
Official Full Name	cyclin Y-like 1 provided byMGI
Primary source	MGI:MGI:2138614
See related	Ensembl:ENSMUSG00000070871
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	9630037P07Rik, AW554339
Expression	Ubiquitous expression in testis adult (RPKM 21.2), limb E14.5 (RPKM 6.5) and 26 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

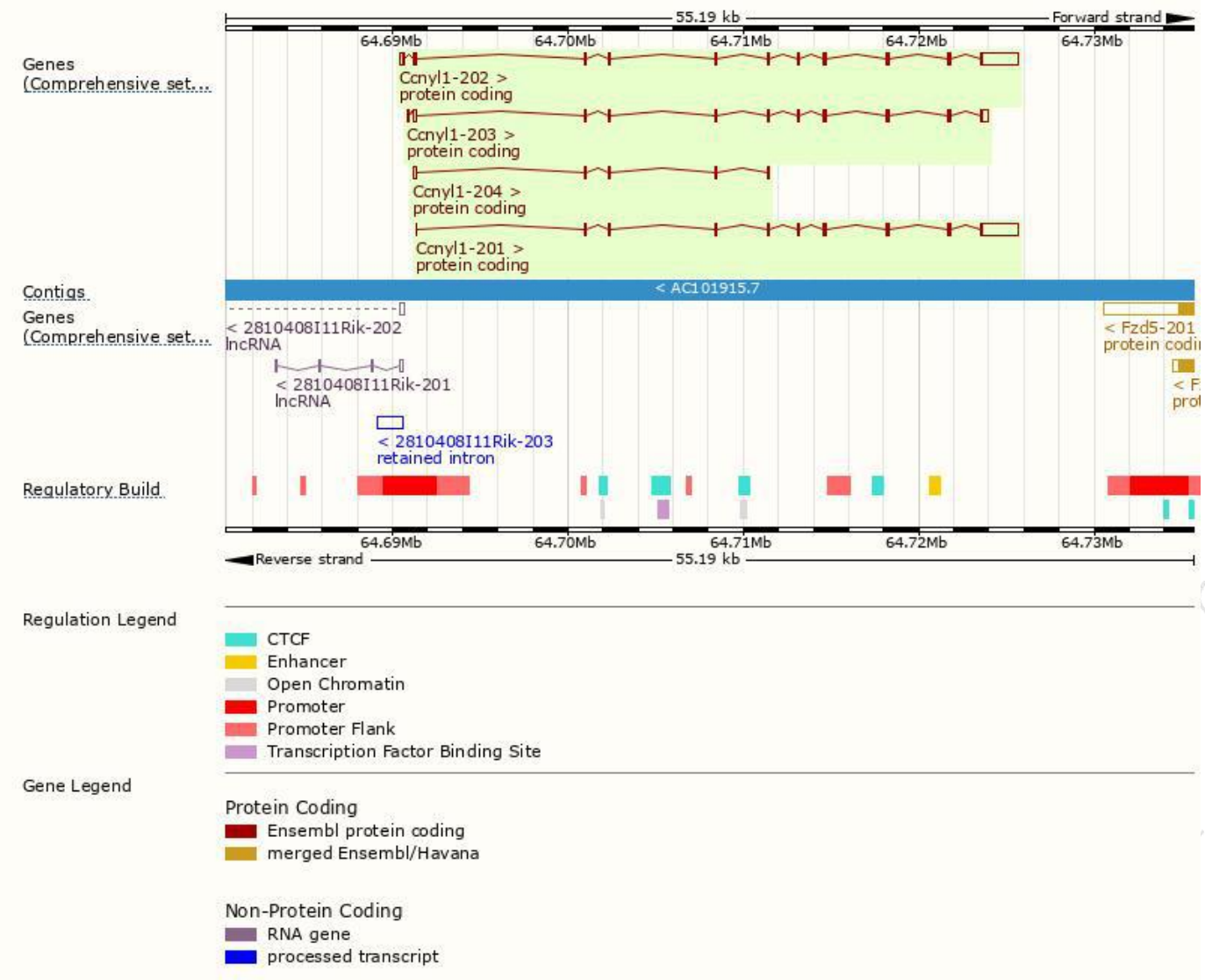
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ccnyl1-201	ENSMUST00000094898.4	2923	291aa	Protein coding	CCDS48281	E9Q226	TSL:5 GENCODE basic APPRIS P2
Ccnyl1-203	ENSMUST00000187170.6	1506	291aa	Protein coding	CCDS48281	E9Q226	TSL:5 GENCODE basic APPRIS P2
Ccnyl1-202	ENSMUST00000114077.7	3281	367aa	Protein coding	-	D3YUJ3	TSL:1 GENCODE basic APPRIS ALT2
Ccnyl1-204	ENSMUST00000187638.6	465	85aa	Protein coding	-	A0A087WPI4	CDS 3' incomplete TSL:5

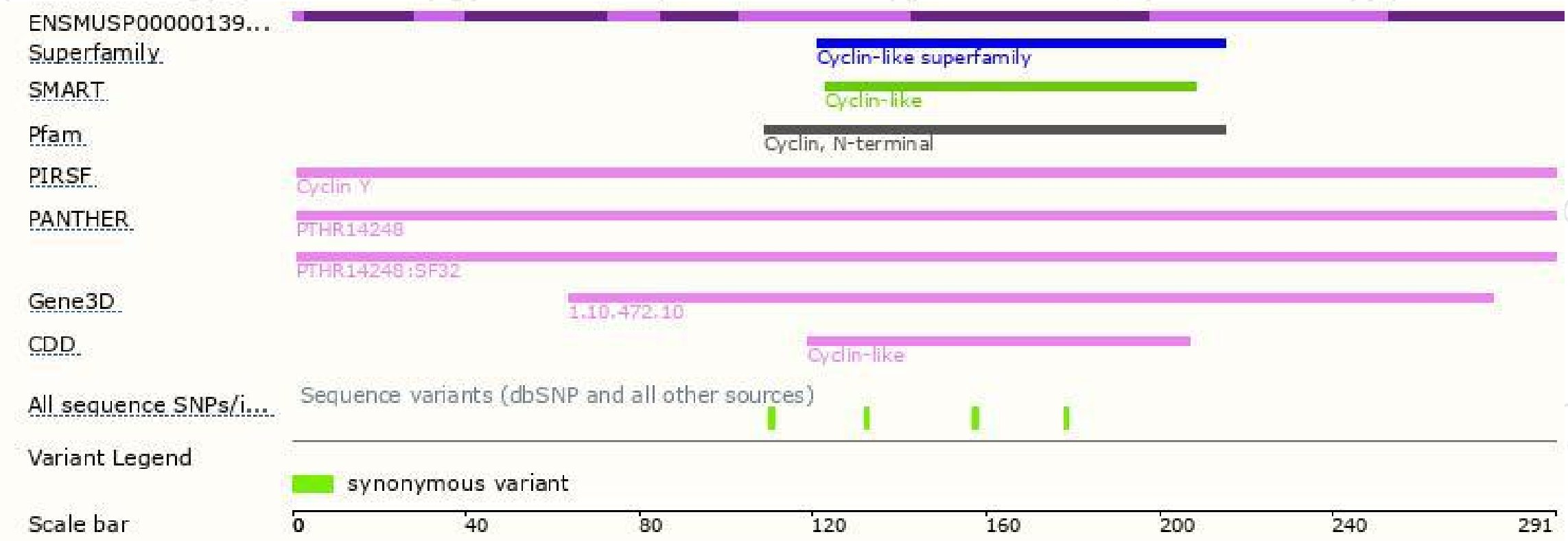
The strategy is based on the design of *Ccnyl1-201* transcript,the transcription is shown below:



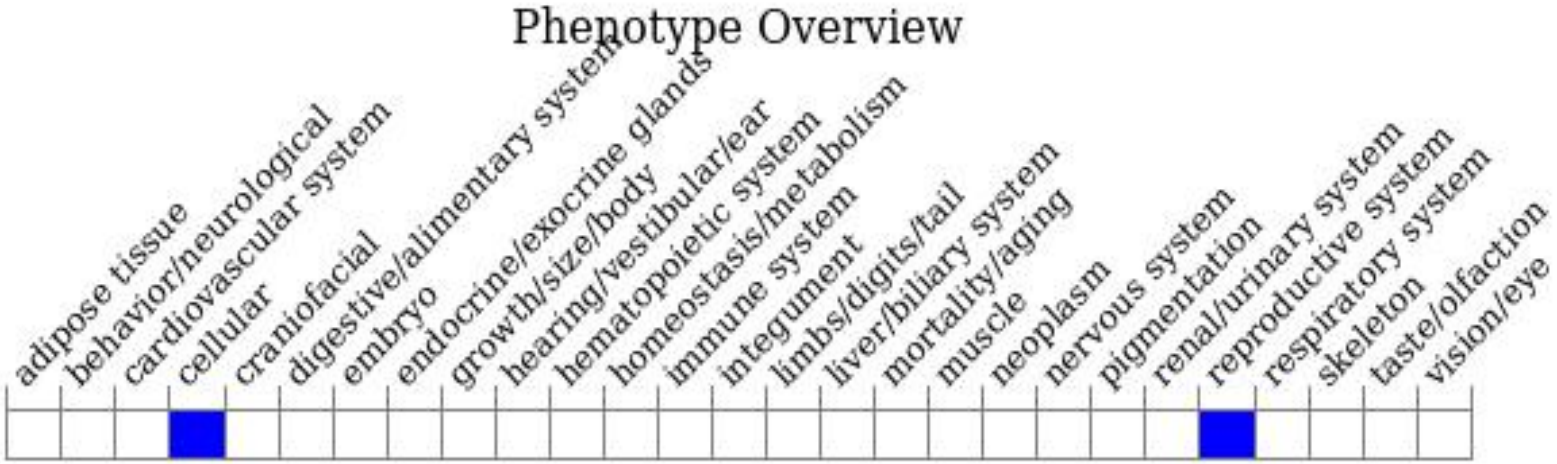
Genomic location distribution



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,mice homozygous for a knock-out allele exhibit male infertility associated with asthenozoospermia and teratozoospermia.

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

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