Tipin Cas9-CKO Strategy

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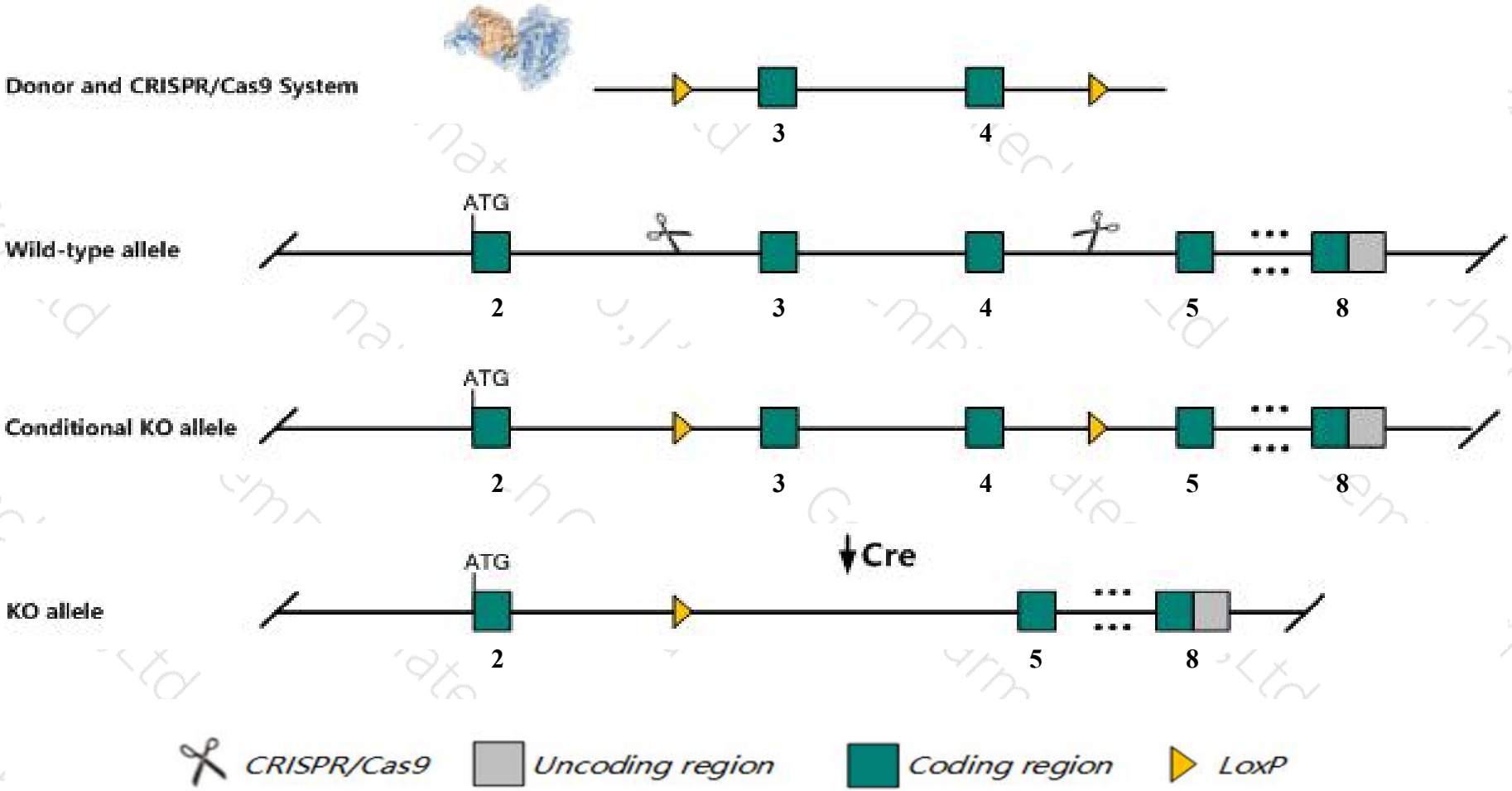
Project Overview

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

Conditional Knockout strategy

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



Technical routes

- The *Tipin* gene has 5 transcripts. According to the structure of *Tipin* gene, exon3-exon4 of *Tipin-205* (ENSMUST00000216594.1) transcript is recommended as the knockout region.The region contains 149bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Tipin* 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.

Notice

- The *Tipin* gene is located on the Chr9. 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.

- The effect on transcript *Tipin*-202&203&204 is unknown.

- The floxed region is near to the N-terminal of *Gm24289* gene,this strategy may influence the regulatory function of the N-terminal of *Gm24289* gene.

- 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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Tipin timeless interacting protein [Mus musculus (house mouse)]

Gene ID: 66131, updated on 13-Mar-2020

Summary

Official Symbol	Tipin provided by MGI
Official Full Name	timeless interacting protein provided by MGI
Primary source	MGI:MGI:1921571
See related	Ensembl:ENSMUSG00000032397
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	1110005A05Rik, 1110018P21Rik
Summary	The protein encoded by this gene is part of the replisome complex, a group of proteins that support DNA replication. It binds TIM and aids in protecting cells against DNA damage and stress. [provided by RefSeq, Feb 2014]
Expression	Biased expression in liver E14 (RPKM 28.2), CNS E11.5 (RPKM 20.3) and 13 other tissues See more
Orthologs	human all

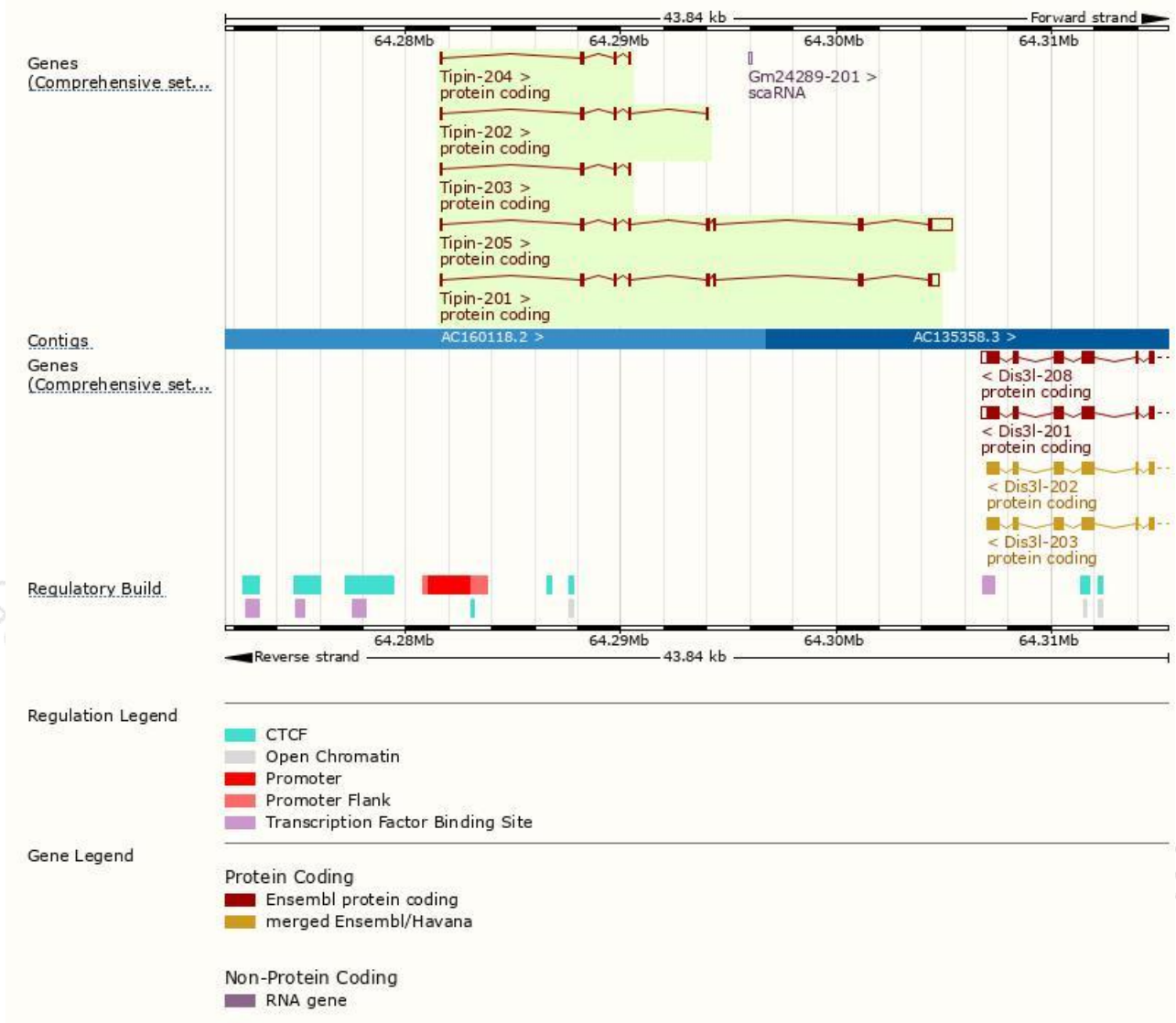
Transcript information（Ensembl）

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

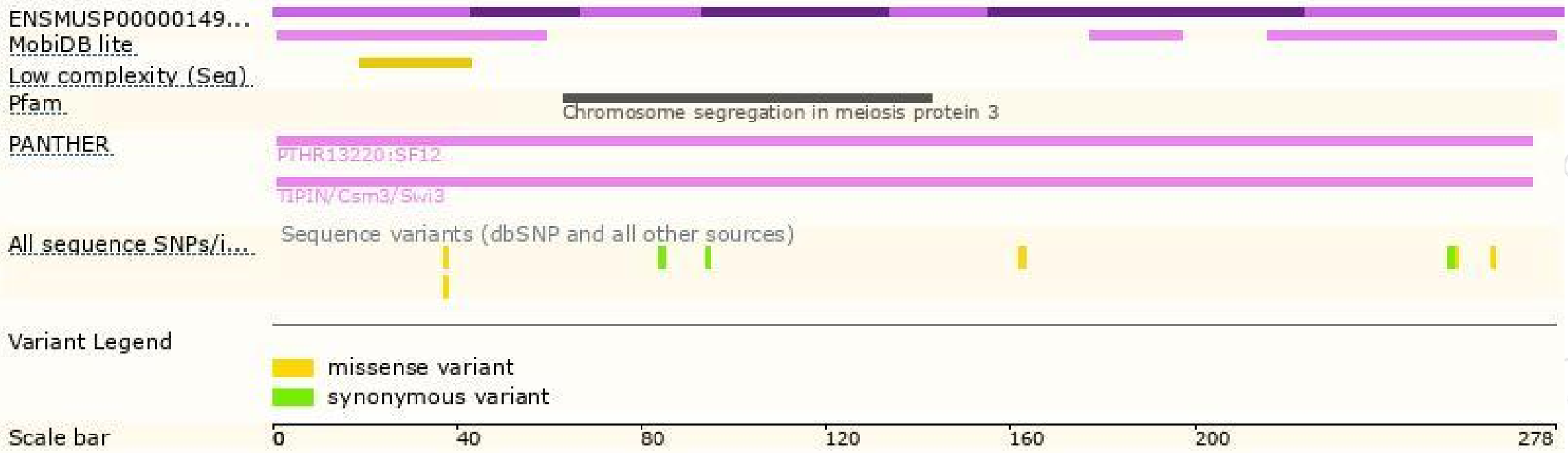
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Tipin-205	ENSMUST00000216594.1	1841	278aa	Protein coding	CCDS23279	Q91WA1	TSL:1 GENCODE basic APPRIS P1
Tipin-201	ENSMUST00000034964.6	1211	278aa	Protein coding	CCDS23279	Q91WA1	TSL:1 GENCODE basic APPRIS P1
Tipin-202	ENSMUST00000213165.1	400	111aa	Protein coding	-	A0A1L1SUJ5	CDS 3' incomplete TSL:2
Tipin-203	ENSMUST00000213289.1	353	93aa	Protein coding	-	A0A1L1STK1	CDS 3' incomplete TSL:2
Tipin-204	ENSMUST00000215031.1	352	93aa	Protein coding	-	A0A1L1SUT4	CDS 3' incomplete TSL:2

The strategy is based on the design of *Tipin-205* transcript,the transcription is shown below:

Genomic location distribution



Protein domain



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

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