

Sirt3 Cas9-CKO Strategy

Designer:

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Project Overview

Project Name

Sirt3

Project type

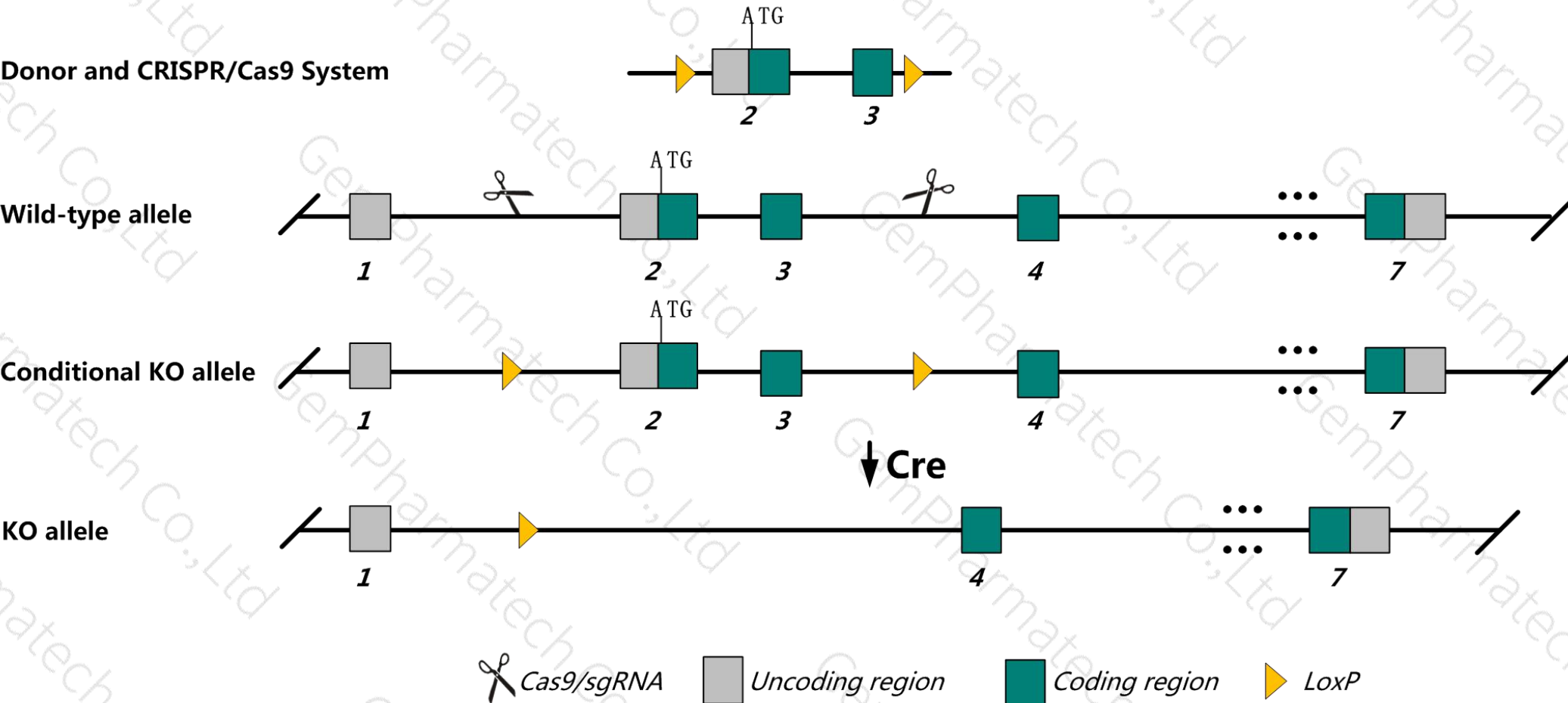
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Sirt3* gene. The schematic diagram is as follows:



- The *Sirt3* gene has 10 transcript. According to the structure of *Sirt3* gene, exon2-3 of *Sirt3*-201 transcript is recommended as the knockout region. The region contains start codon ATG coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Sirt3* gene. The brief process is as follows: gRNA was transcribed in vitro, donor was constructed. Cas9, gRNA 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 or cell types.

- According to the existing MGI data , Mice homozygous for one null allele exhibit increased systolic blood pressure while mice homozygous for another allele exhibit reduced ATP production.
- The *Sirt3* gene is located on the Chr7. 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)

Sirt3 sirtuin 3 [*Mus musculus* (house mouse)]

Gene ID: 64384, updated on 20-Jul-2019

Summary



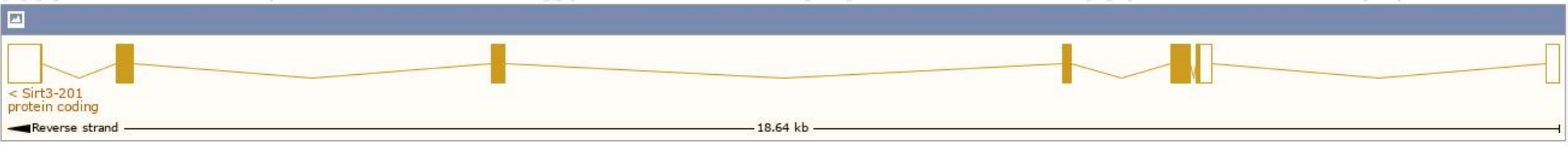
Official Symbol	Sirt3 provided by MGI
Official Full Name	sirtuin 3 provided by MGI
Primary source	MGI:MGI:1927665
See related	Ensembl:ENSMUSG00000025486
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Sir2l3; AI848213; 2310003L23Rik
Expression	Ubiquitous expression in kidney adult (RPKM 35.3), liver adult (RPKM 33.7) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

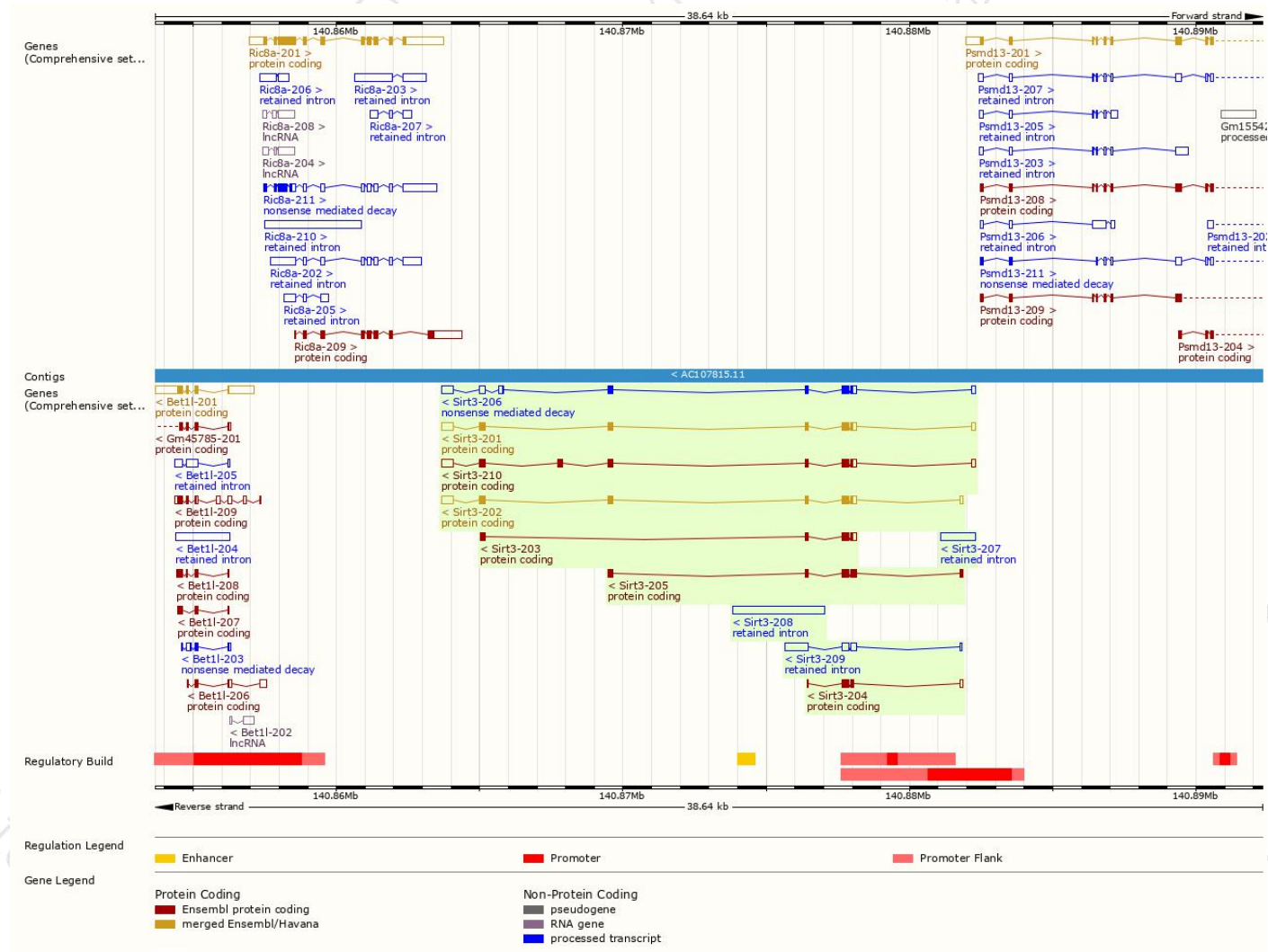
The gene has 10 transcripts, and all transcripts are shown below:

Show/hide columns (1 hidden)		Filter					
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Sirt3-201	ENSMUST00000026559.13	1461	257aa	Protein coding	CCDS21989	Q4FJK3 Q8R104	TSL:1 Gencode basic APPRIS P1
Sirt3-202	ENSMUST00000106048.9	1416	257aa	Protein coding	CCDS21989	Q4FJK3 Q8R104	TSL:1 Gencode basic APPRIS P1
Sirt3-210	ENSMUST00000211179.1	1606	309aa	Protein coding	-	A0A1B0GRY5	TSL:5 Gencode basic
Sirt3-205	ENSMUST00000147331.8	784	258aa	Protein coding	-	D3YTK6	CDS 3' incomplete TSL:3
Sirt3-203	ENSMUST00000137024.7	690	182aa	Protein coding	-	D3YVQ5	CDS 3' incomplete TSL:3
Sirt3-204	ENSMUST00000137710.1	456	100aa	Protein coding	-	D3YWD6	CDS 3' incomplete TSL:5
Sirt3-206	ENSMUST00000210296.1	1598	189aa	Nonsense mediated decay	-	A0A1B0GRF7	TSL:5
Sirt3-208	ENSMUST00000210706.1	3206	No protein	Retained intron	-	-	TSL:NA
Sirt3-209	ENSMUST00000210974.1	1272	No protein	Retained intron	-	-	TSL:2
Sirt3-207	ENSMUST00000210398.1	1223	No protein	Retained intron	-	-	TSL:NA

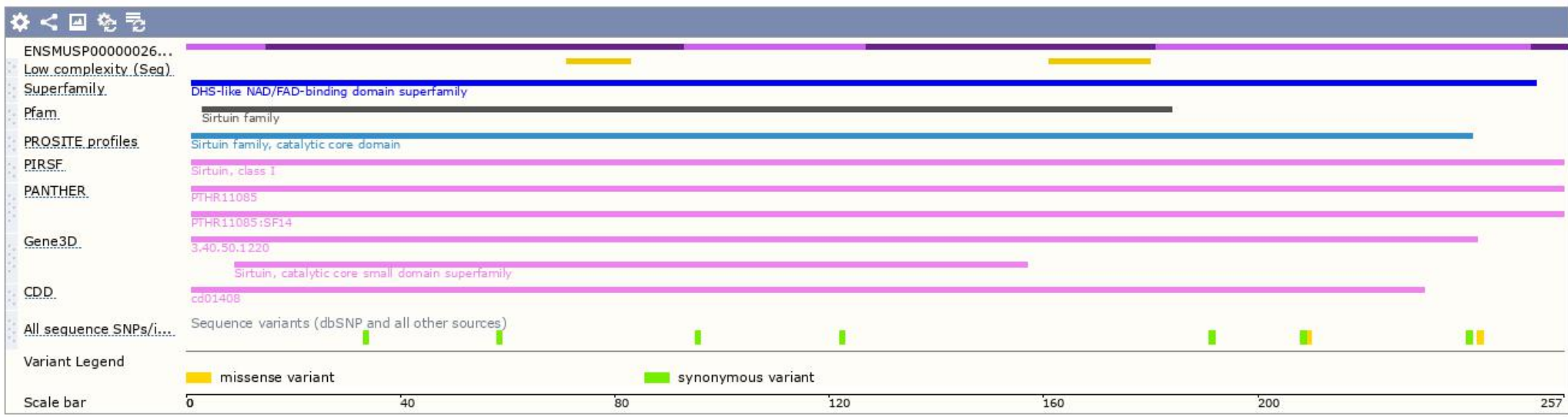
The strategy is based on the design of *Sirt3*-201 transcript, The transcription is shown below



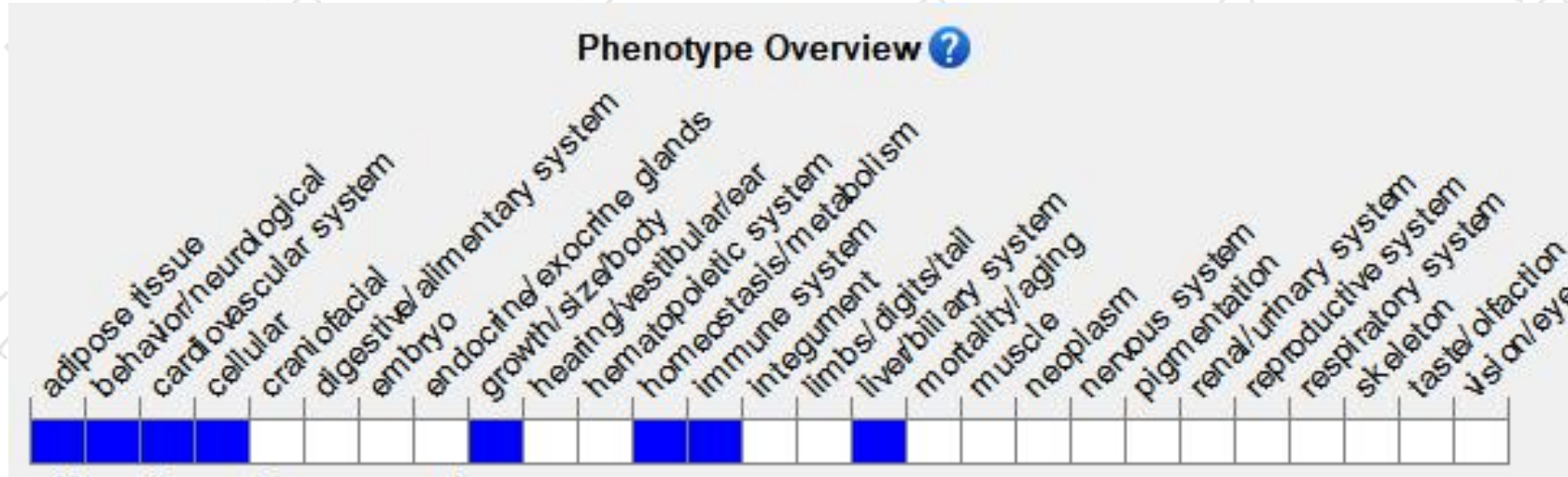
Genomic location distribution



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, Mutations in this locus affect cell-cycle regulation and apoptosis. Null homozygotes show high, early-onset tumor incidence; some have persistent hyaloid vasculature and cataracts. Truncated or temperature-sensitive alleles cause early aging phenotypes.

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