集萃药康
GemPharmatech

Sec31b Cas9-CKO Strategy

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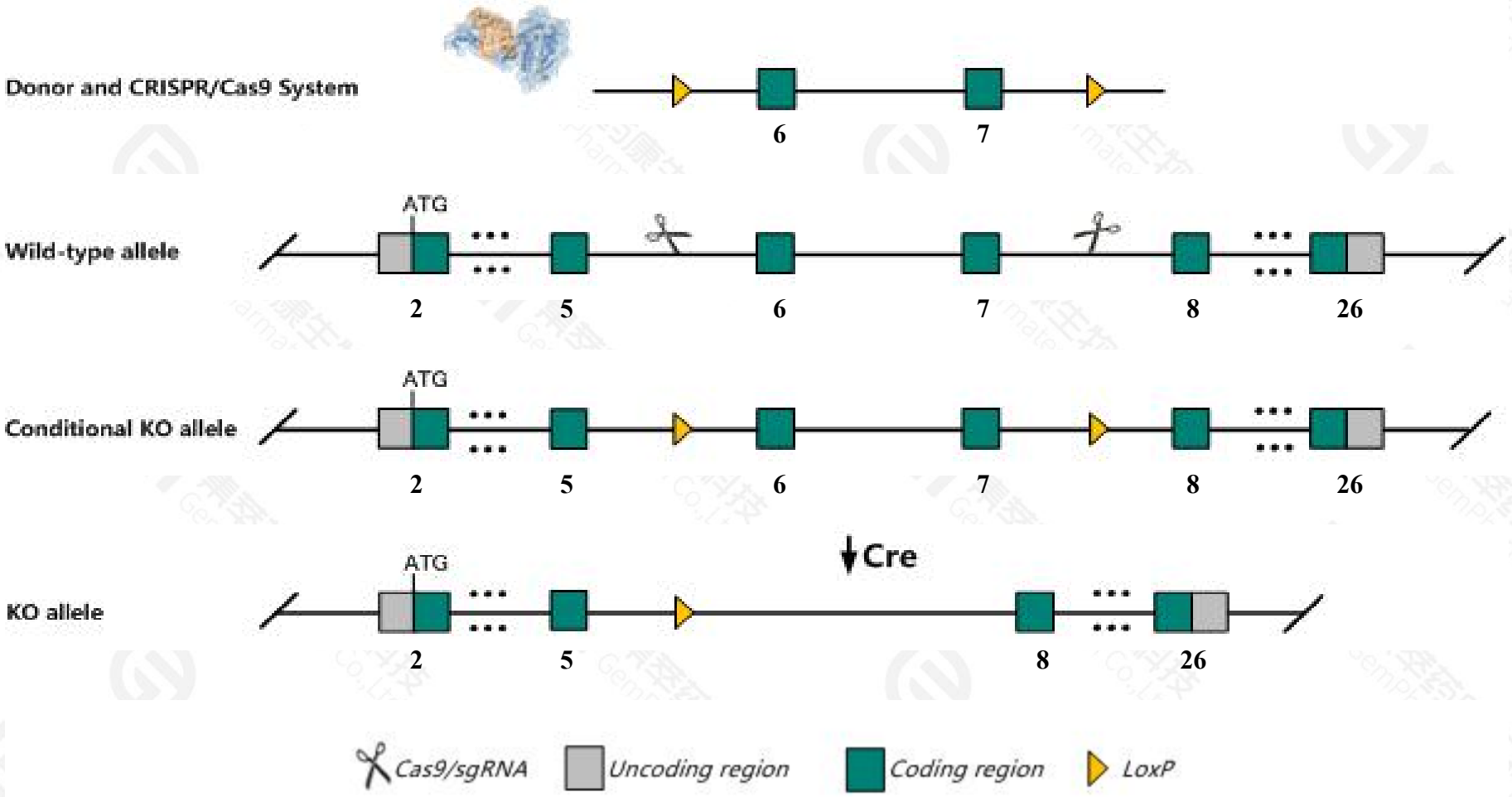
Project Overview

Project Name	***Sec31b***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Sec31b* gene. The schematic diagram is as follows:



Technical routes

➢ The *Sec31b* gene has 3 transcripts. According to the structure of *Sec31b* gene, exon6-exon7 of *Sec31b-201*(ENSMUST00000063632.14) transcript is recommended as the knockout region.The region contains 287bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Sec31b* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor 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

➢ The *Sec31b* gene is located on the Chr19. 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.

➢Transcript *Sec31b*-203 may not be affected.

➢ 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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Sec31b Sec31 homolog B (S. cerevisiae) [Mus musculus (house mouse)]

Gene ID: 240667, updated on 17-Nov-2020

Summary

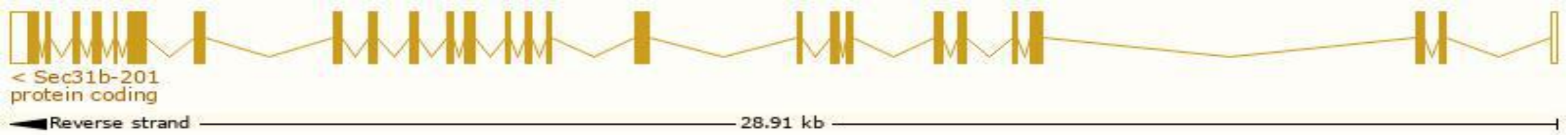
Official Symbol	Sec31b provided by MGI
Official Full Name	Sec31 homolog B (S. cerevisiae) provided byMGI
Primary source	MGI:MGI:2685187
See related	Ensembl:ENSMUSG00000051984
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Gm341, Sec31, Sec31l2
Expression	Broad expression in heart adult (RPKM 6.7), mammary gland adult (RPKM 5.3) and 19 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

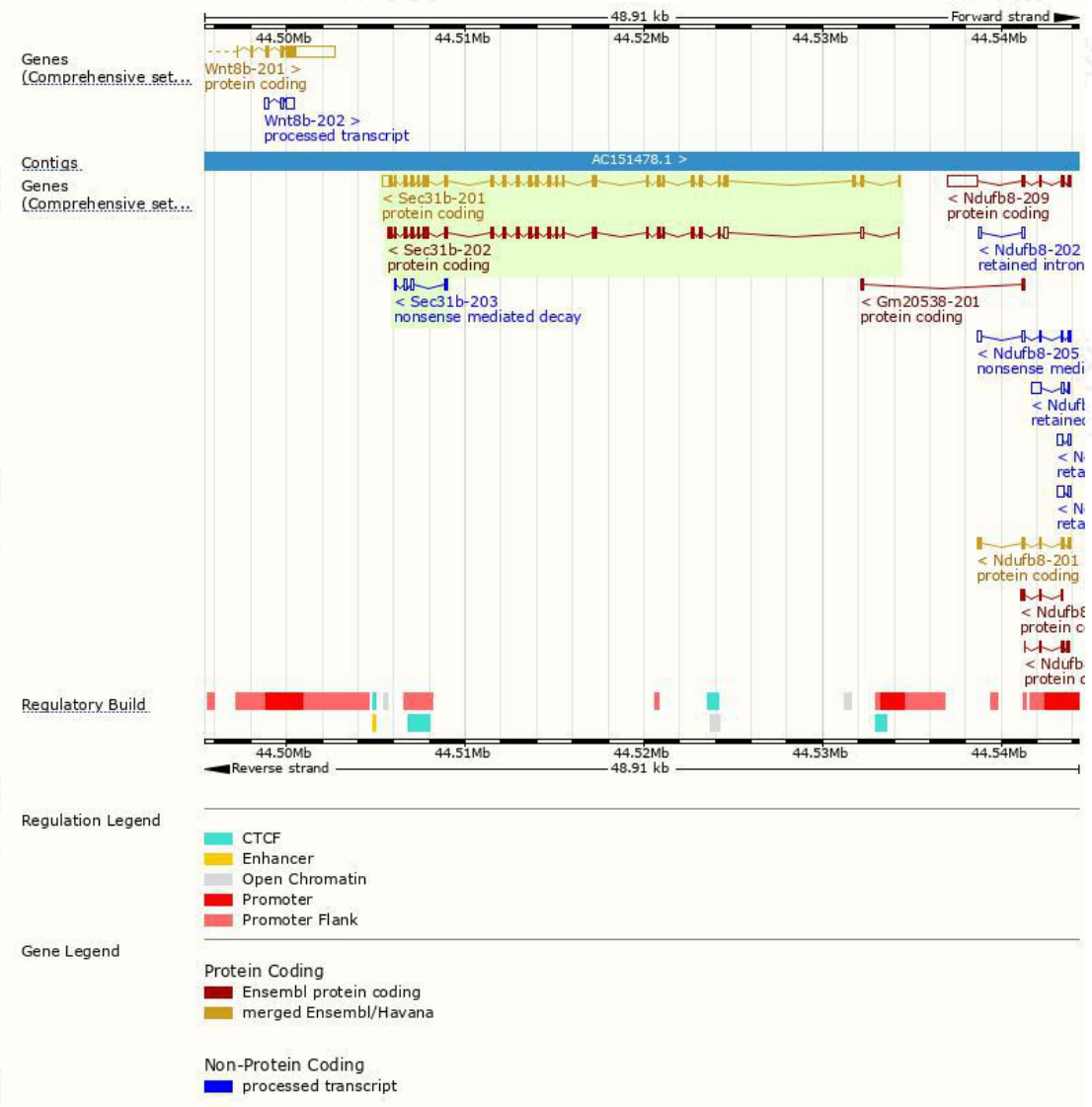
The gene has 3 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Sec31b-201	ENSMUST00000063632.14	3939	1158aa	Protein coding	CCDS37997		TSL:5 , GENCODE basic , APPRIS P1 ,
Sec31b-202	ENSMUST00000111985.2	3548	1001aa	Protein coding	-		TSL:1 , GENCODE basic ,
Sec31b-203	ENSMUST00000165758.2	503	59aa	Nonsense mediated decay	-		CDS 5' incomplete , TSL:5 ,

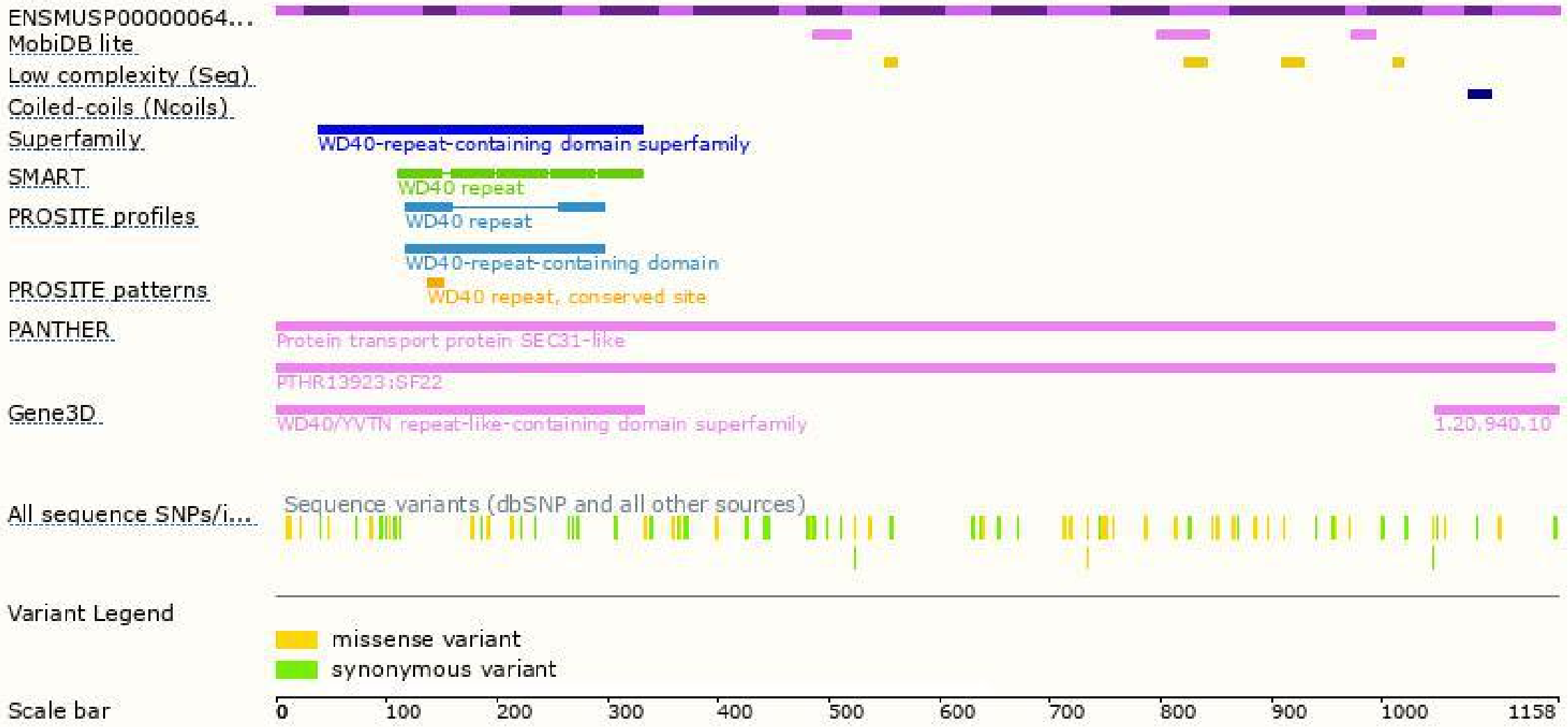
The strategy is based on the design of *Sec31b-201* transcript,the transcription is shown below:



Genomic location distribution



Protein domain



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

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