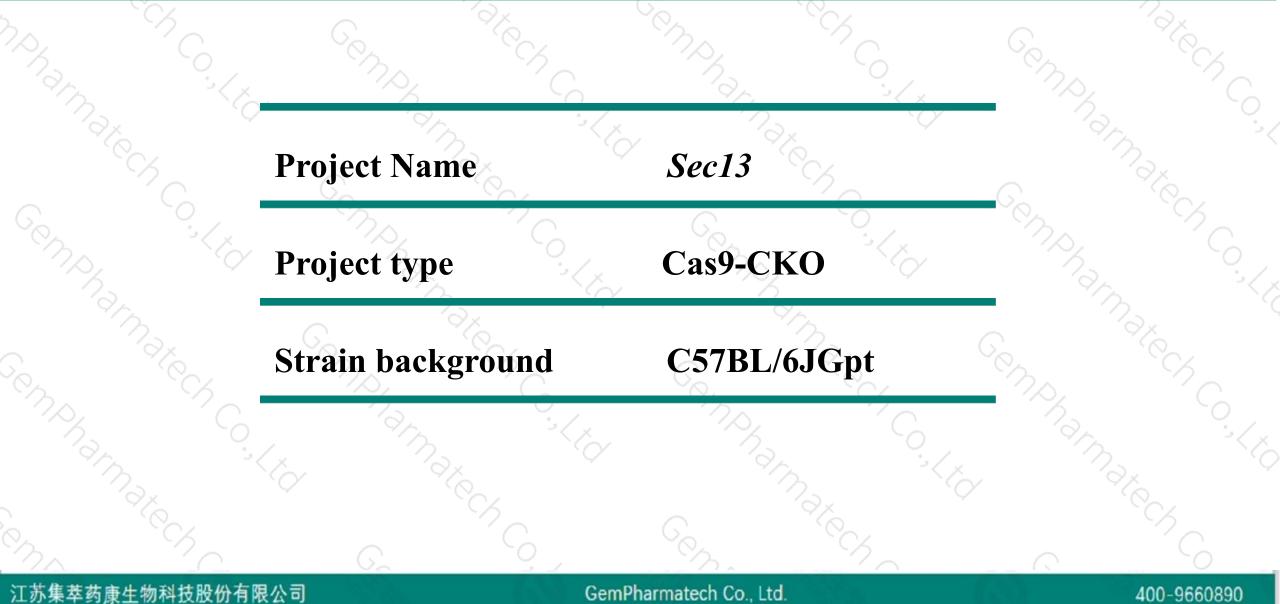


Sec13 Cas9-CKO Strategy

Designer: Xueting Zhang Reviewer:Yanhua Shen Date:2020-02-25

Project Overview



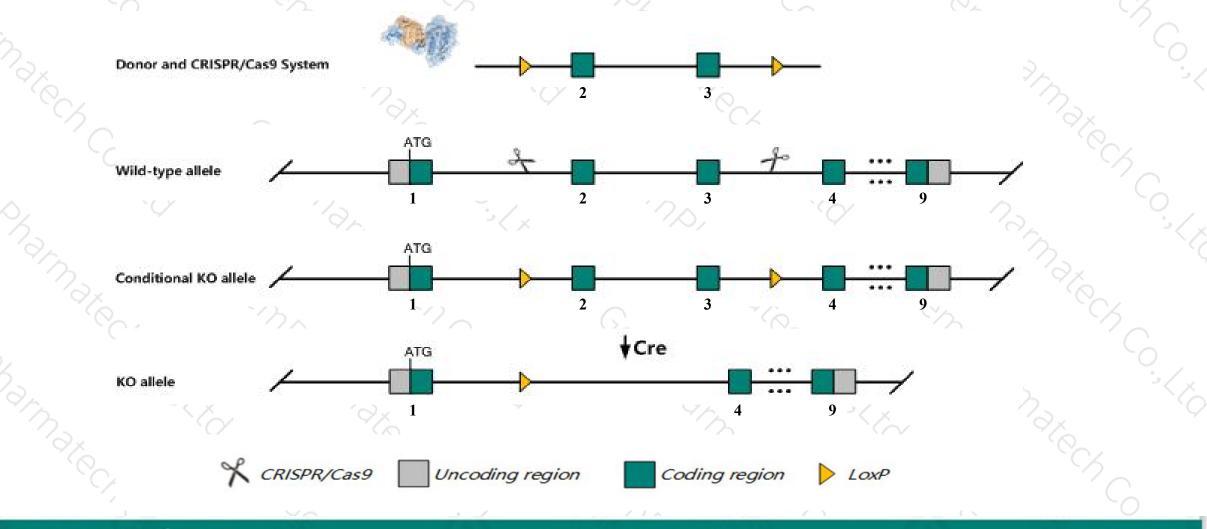


Conditional Knockout strategy



400-9660890

This model will use CRISPR/Cas9 technology to edit the Sec13 gene. The schematic diagram is as follows:



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The Sec13 gene has 4 transcripts. According to the structure of Sec13 gene, exon2-exon3 of Sec13-201 (ENSMUST00000032440.5) transcript is recommended as the knockout region. The region contains 161bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify Sec13 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



- ➤ Transcript *Sec13*-202&203&204 may not be affected.
- The Sec13 gene is located on the Chr6. 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)



\$?

2 ?

Sec13 SEC13 homolog, nuclear pore and COPII coat complex component [Mus musculus (house mouse)]

Gene ID: 110379, updated on 27-Aug-2019

Summary

| Official Symbol | Sec13 provided by MGI |
|--------------------|--|
| Official Full Name | SEC13 homolog, nuclear pore and COPII coat complex component provided by MGI |
| Primary source | MGI:MGI:99832 |
| See related | Ensembl:ENSMUSG00000030298 |
| 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; Mus; Mus |
| Also known as | Sec13r; Sec13l1; 1110003H02Rik |
| Expression | Ubiquitous expression in large intestine adult (RPKM 114.3), limb E14.5 (RPKM 92.9) and 28 other tissues See more |
| Orthologs | human all |

Genomic context

Location: 6 E3; 6 52.84 cM

Exon count: 9

See Sec13 in Genome Data Viewer

| Annotation relea | se Status | Assembly | Chr | Location |
|------------------|-------------------|------------------------------|-----|--|
| 108 | current | GRCm38.p6 (GCF_000001635.26) | 6 | NC_000072.6 (113728052113740681, complement) |
| Build 37.2 | previous assembly | MGSCv37 (GCF_000001635.18) | 6 | NC_000072.5 (113678046113690675, complement) |

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

| Name | Transcript ID | bp | Protein | Biotype | CCDS | UniProt | Flags |
|-----------|---------------------------|------|--------------|-----------------|-----------|---------------|-------------------------------|
| Sec13-201 | ENSMUST0000032440.5 | 1353 | <u>322aa</u> | Protein coding | CCDS39596 | Q9D1M0 | TSL:1 GENCODE basic APPRIS P1 |
| Sec13-204 | ENSMUST00000205135.1 | 1599 | No protein | Retained intron | | | TSL:NA |
| Sec13-202 | ENSMUST00000138204.7 | 492 | No protein | Retained intron | 2 | 83 2 5 | TSL:5 |
| Sec13-203 | -203 ENSMUST00000145979.1 | | No protein | Retained intron | - | 1225 | TSL:3 |

The strategy is based on the design of Sec13-201 transcript, The transcription is shown below

< Sec13-201 protein coding

Reverse strand

- 12.68 kb -

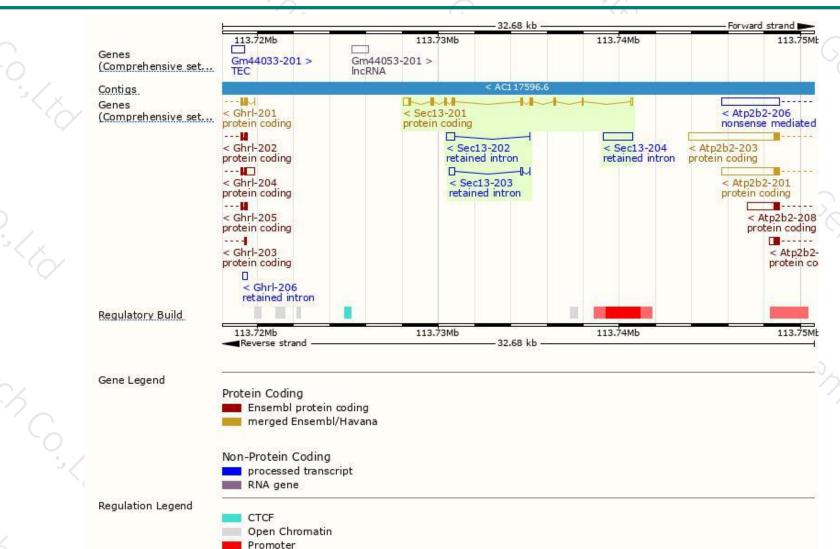
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Genomic location distribution

Promoter Flank





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Protein domain



| <u>`</u> 0 | | | | | ×. | | | р Л | |
|--------------------------------|-----------------------|--|-------------------|-------------|-----|-----|------------|----------|-----|
| ENSMUSP00000032 Superfamily | WD40- | epeat-contai | ning domain sup | erfamily | | | | | 9 |
| SMART | WD40 repeat | | - | | | | | | |
| <u>Pfam</u> | WD40 re | peat | 0 <mark>-0</mark> | | | | | 1 | |
| PROSITE profiles | WD40 rep | eat | | | | | | | |
| PANTHER. | WD40-rej Sec13/Seh | peat-containir 1 family | ig domain | | | | | - | 2 |
| | Protein Sec | 13 | | | | | | | |
| Gene3D | WD40/YVTN | repeat-like-c | ontaining domair | superfamily | | | | | |
| All sequence SNPs/i | Sequence v | variants (dbS | SNP and all oth | er sources) | | | E 4 | | |
| Variant Legend | splice | nse variant region varia ymous varia | | | | | | | 0, |
| Scale bar | 0 | 40 | 80 | 120 | 160 | 200 | 240 | 280 | 322 |
| a tech | | | | | | | | | × |



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If you have any questions, you are welcome to inquire. Tel: 400-9660890



