Shc4 Cas9-CKO Strategy

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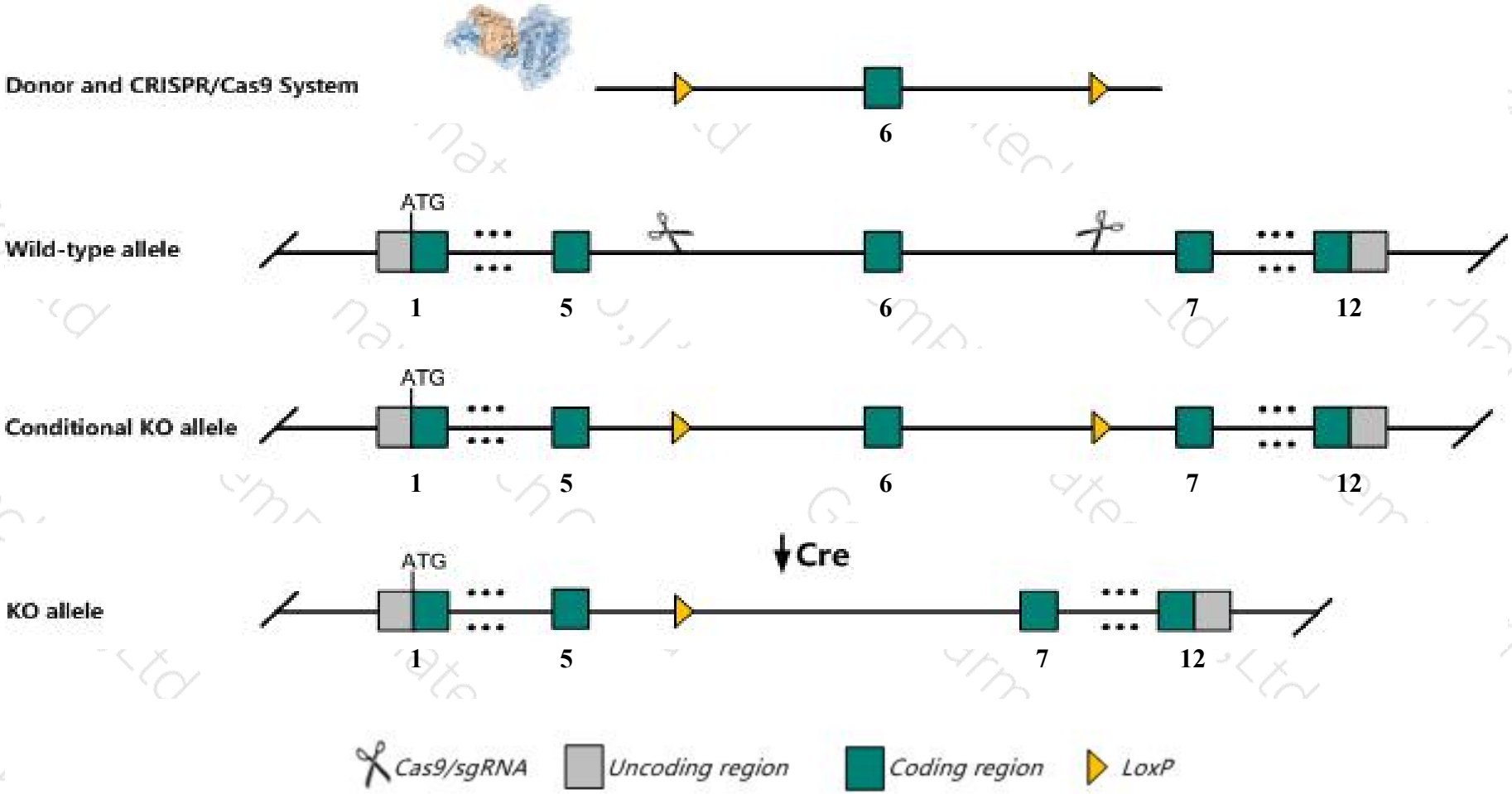
Project Overview

Project Name	***Shc4***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Shc4* gene. The schematic diagram is as follows:



Technical routes

➢ The *Shc4* gene has 4 transcripts. According to the structure of *Shc4* gene, exon6 of *Shc4-201*(ENSMUST00000042246.13) transcript is recommended as the knockout region.The region contains 52bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Shc4* 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

➢ The *Shc4* gene is located on the Chr2. 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 floxed region is near to the N-terminal of *Eid1* gene,this strategy may influence the regulatory function of the N-terminal of *Eid1* gene.

➢ Transcript *Shc4*-204 may not be affected.

➢ The N-terminal of *Shc4* gene will remain several amino acids ,it may remain the partial function of *Shc4* 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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Shc4 SHC (Src homology 2 domain containing) family, member 4 [Mus musculus (house mouse)]

Gene ID: 271849, updated on 22-Mar-2020

Summary

Official Symbol	Shc4 provided by MGI
Official Full Name	SHC (Src homology 2 domain containing) family, member 4 provided by MGI
Primary source	MGI:MGI:2655364
See related	Ensembl:ENSMUSG00000035109
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	6230417E10Rik, 9930029B02Rik, Gm685, RaLP
Expression	Broad expression in genital fat pad adult (RPKM 3.4), testis adult (RPKM 2.5) and 23 other tissues See more
Orthologs	human all

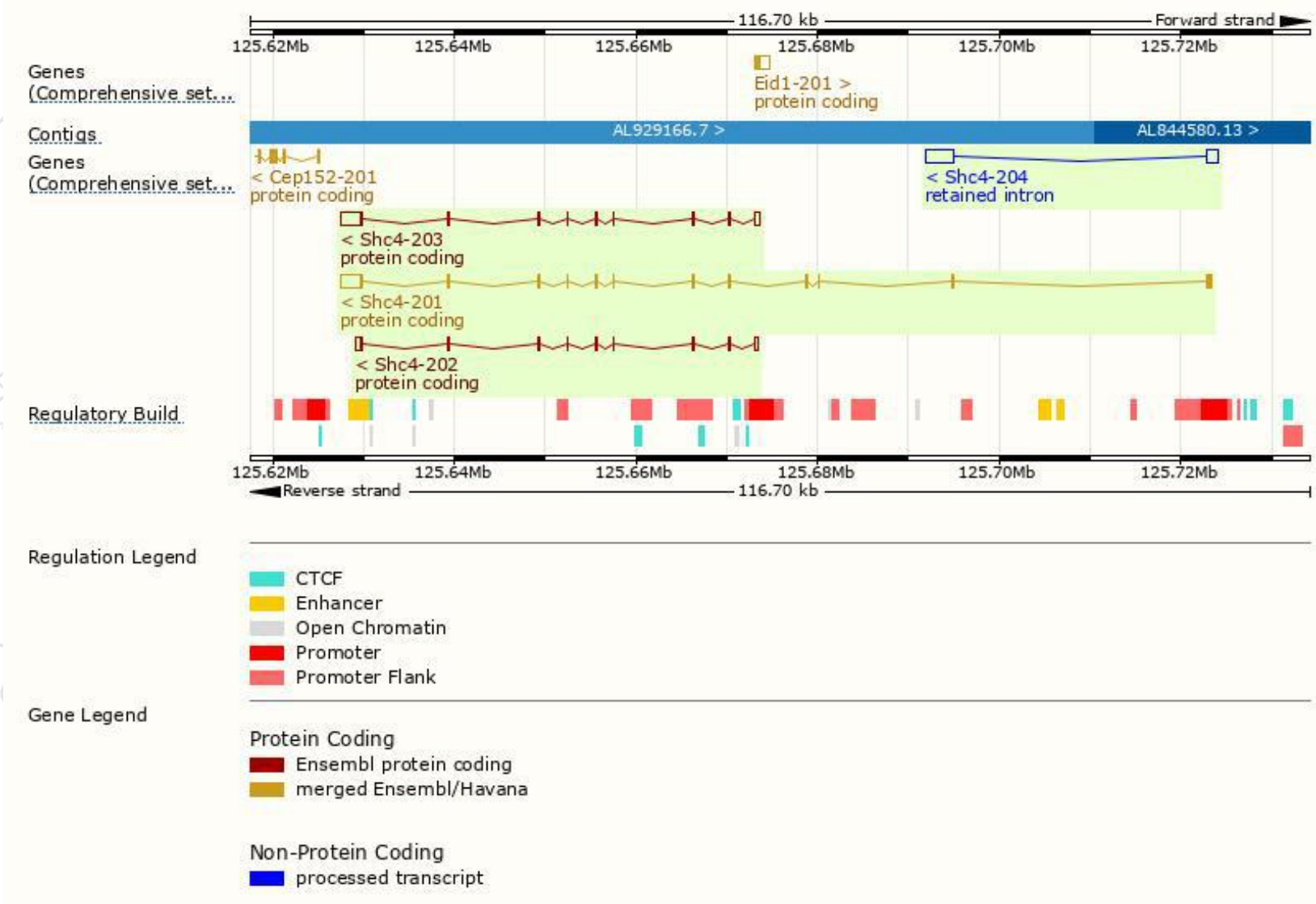
Transcript information（Ensembl）

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

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Shc4-201	ENSMUST00000042246.13	4059	626aa	Protein coding	CCDS16678	Q6S5L9	TSL:1 GENCODE basic APPRIS P1
Shc4-203	ENSMUST00000110480.7	3671	340aa	Protein coding	-	A2AUN0	TSL:1 GENCODE basic
Shc4-202	ENSMUST00000110477.1	1988	340aa	Protein coding	-	A2AUN0	TSL:1 GENCODE basic
Shc4-204	ENSMUST00000157002.1	4343	No protein	Retained intron	-	-	TSL:1

The strategy is based on the design of *Shc4-201* 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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