Stard8 Cas9-CKO Strategy

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Project Overview

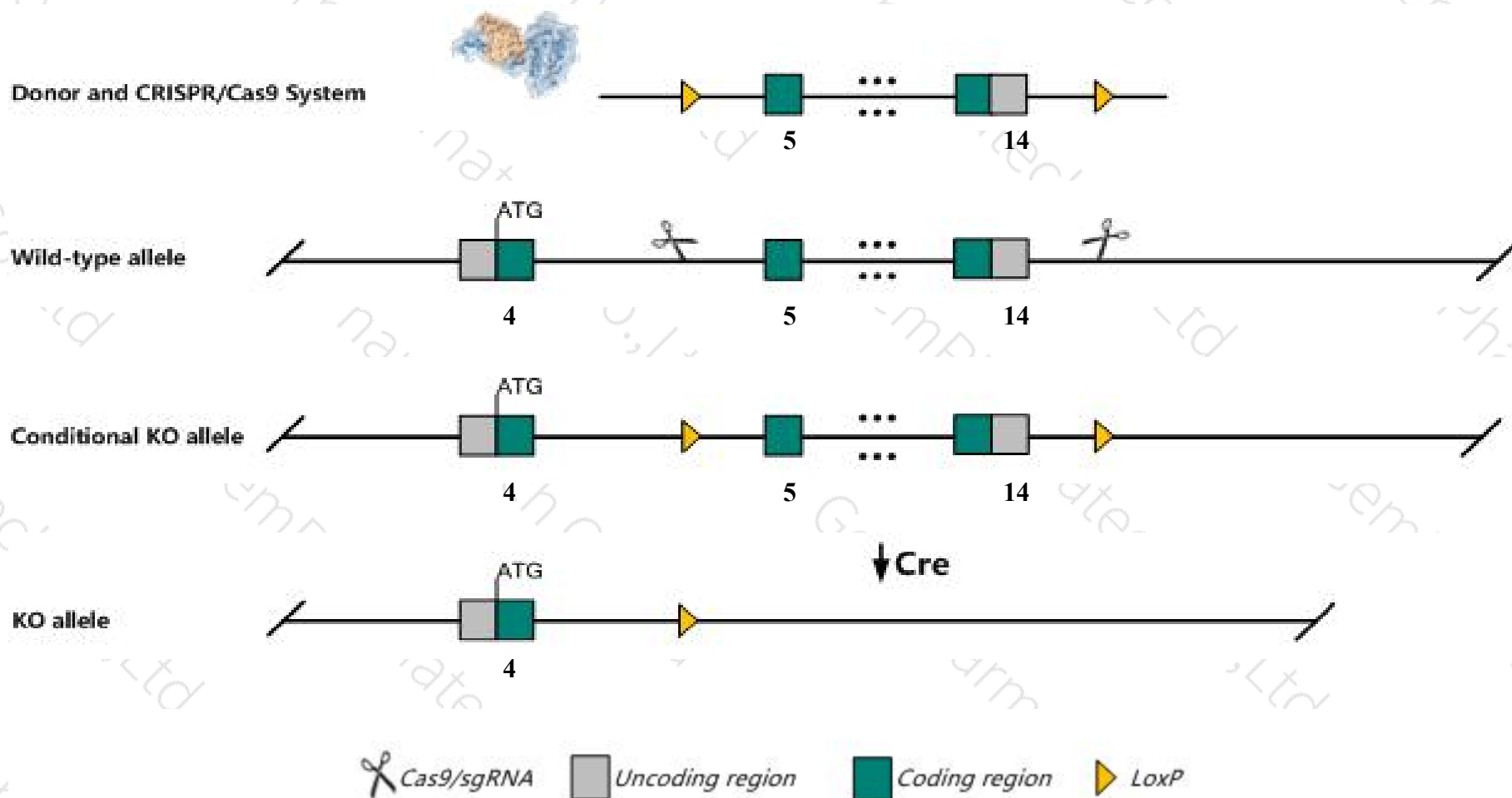|	
Project Name	*Stard8*
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Stard8* gene. The schematic diagram is as follows:

Technical routes

➢ The *Stard8* gene has 4 transcripts. According to the structure of *Stard8* gene, exon5-exon14 of *Stard8-201*(ENSMUST00000036606.13) transcript is recommended as the knockout region.The region contains most of the coding sequence.Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Stard8* 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 *Stard8* gene is located on the ChrX. 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 *Stard8*-204 is unknown.

➢ 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）

Stard8 START domain containing 8 [Mus musculus (house mouse)]

Gene ID: 236920, updated on 13-Mar-2020

▲ Summary

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Official Symbol	Stard8 provided by MGI
Official Full Name	START domain containing 8 provided byMGI
Primary source	MGI:MGI:2448556
See related	Ensembl:ENSMUSG00000031216
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	Dlc3, mKIAA0189
Expression	Broad expression in lung adult (RPKM 27.0), kidney adult (RPKM 19.8) and 20 other tissuesSee 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
Stard8-201	ENSMUST00000036606.13	4963	1019aa	Protein coding	CCDS30297	Q8K031	TSL:1 GENCODE basic APPRIS P1
Stard8-204	ENSMUST00000149999.7	631	57aa	Protein coding	-	B1AZJ1	CDS 3' incomplete TSL:3
Stard8-202	ENSMUST00000127361.1	759	No protein	Processed transcript	-	-	TSL:3
Stard8-203	ENSMUST00000145820.1	468	No protein	Processed transcript	-	-	TSL:2

The strategy is based on the design of *Stard8-201* transcript,the transcription is shown below:

Genomic location distribution
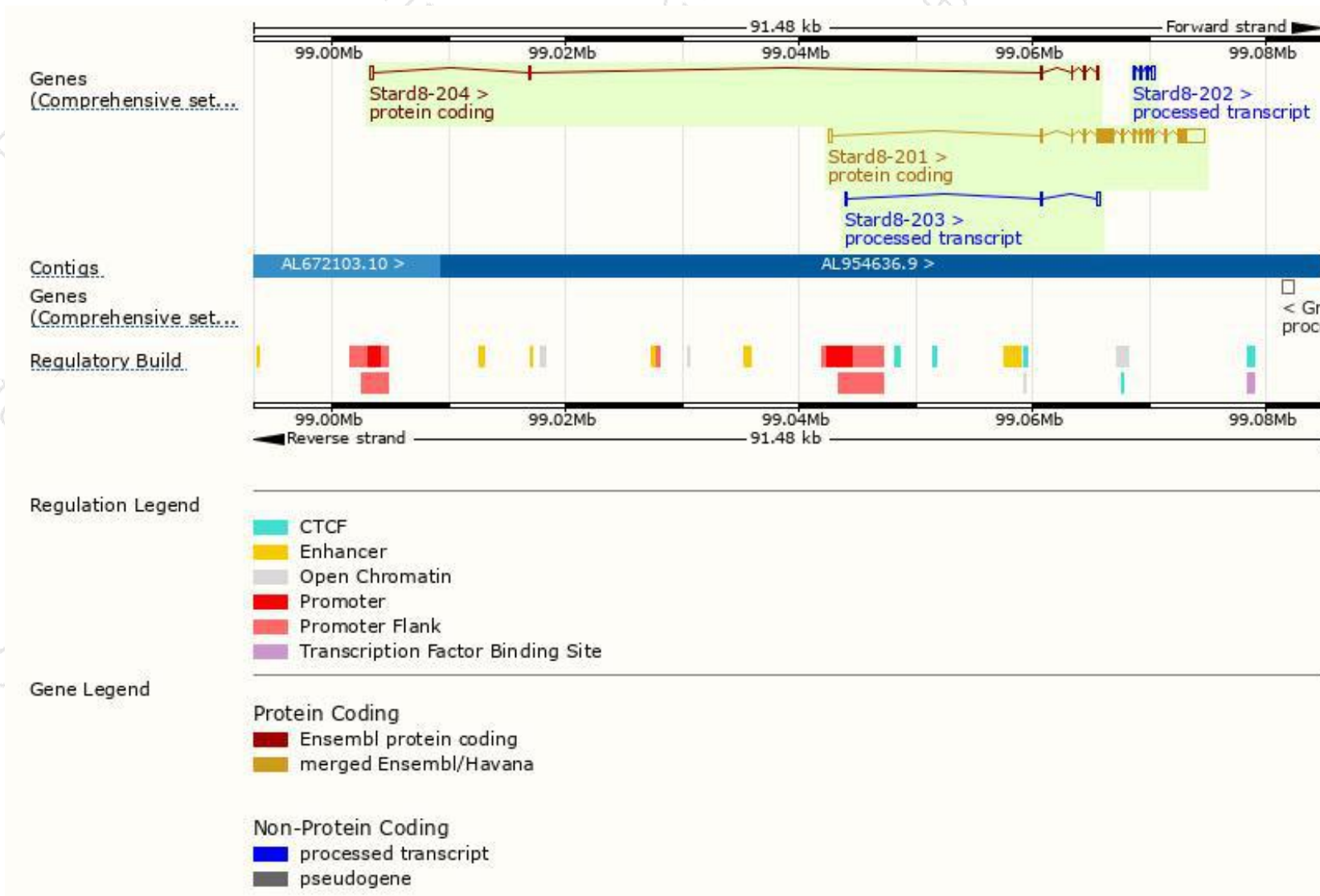
Protein domain

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